



INTERNATIONAL
ASSOCIATION
FOR THE PLANT
PROTECTION SCIENCES



HELLENIC
SOCIETY
OF PHYTIATRY



AGRICULTURAL
UNIVERSITY
OF ATHENS



**1-5
JULY 2024**

MEGARON ATHENS
INTERNATIONAL
CONVENTION CENTER

**ATHENS
GREECE**

XX International **Plant Protection Congress**

**Healthy Plants
Support Human
Welfare**



**Abstract
Book**



Welcome to the 20th IPPC



The International Association for the Plant Protection Sciences welcomes you to the **20th International Plant Protection Congress**

The first International Plant Protection Congress (IPPC) was held in Louvain, Belgium in 1946. It took 50 years before IAPPS became the official organisation responsible for subsequent Congresses.

The IAPPS Board consists of:

An Executive - President, Vice President, Secretary General, and Treasurer

Thirteen Regional Coordinators - Covering the major regions of the world, and

Six Industry and Institutional Representatives

IAPPS membership subscriptions (<https://www.plantprotection.org/subscriptions/>) consist of:

- Developed country
- Student
- Developing country
- Institutional

IAPPS communicates and interacts with its members, and the plant protection community at large, in the following ways:

- The Global Plant Protection News Service
- Involvement in Regional Symposia and Workshops
- Monthly IAPPS Newsletter - published online and in “Crop Protection” (IAPPS Official Journal)



- The IAPPS Website (www.plantprotection.org)



- An Education and Training section of the website including:
 - Plant Protection stories that illustrate the complex multi-disciplinary aspects of many plant protection issues
 - Online publications - including a recent review of digital identification tools for plant biosecurity
 - Digital pathway keys for important rice pests and beneficials in Asia and West Africa

Welcome address from the Chairman

Dear colleagues and friends,

The Hellenic Society of Phytiatry, my colleagues from the Laboratory of Plant Pathology at AUA, and I are honored by IAPPS to organize the XX IPPC in Athens. As chairman and organizational coordinator, I am proud to welcome you.



Having organized and participated in numerous international plant protection congresses, I understand the immense effort and pressure involved in ensuring smooth coordination and decision-making among organizing committees and participants. The COVID-19 pandemic significantly impacted attendance, overlapping with other scientific congresses, yet we greatly appreciate the participation of scientists from around the world.

With the excellent collaboration of Secretary General Short Heinrichs and the IAPPS Governing Board, we have secured outstanding chairs and speakers to enhance the quality of the congress. We are thrilled to have so many top Plant Protection scientists join us in Athens, the cradle of civilization, science, and art.

The program includes 11 keynote-plenary presentations, 550 invited/oral presentations, and 200 posters from over 700 scientists from nearly 60 countries. We are especially proud to welcome over 200 young scientists, including graduate and postgraduate students and postdocs, who will exchange scientific experiences and forge future connections.

We are pleased to host the 14th International Verticillium Symposium during the congress. The International Plant Protection Congress offers a valuable opportunity to exchange information, recognize efforts, and reward scientific achievements in plant protection.

We extend our gratitude to international organizations like FAO and EPPO for their contributions, and to our sponsors BASF, Syngenta, and Corteva for their substantial support. We also thank the IAPPS Governing Board, the local organizing committee, the Agricultural University of Athens, and the Ministry of Rural Development and Food for their assistance.

Special thanks to the congress secretariat led by Mr. Panagiotis Georgakopoulos and Global Events S.A. under Mrs. Bessy Paliouras for their tremendous effort in managing requests and communications.

Welcome to Athens and enjoy the XX IPPC and Greece!

Eleftherios (Eris) Tjamos

Chairman of the XX IPPC ATHENS 2024



XX International Plant Protection Congress Healthy Plants Support Human Welfare

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XX International

Plant Protection Congress Healthy Plants Support Human Welfare

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PROGRAM AT A GLANCE



Program at a Glance

Monday, 1 July

ROOM MC3.2	
14:00-18:00	IAPPS GB meeting
FOYER MEGARON ATHENS INTERNATIONAL CONVENTION CENTER	
16:00-20:00	Registration
BANQUET	
18:00 -20:00	Opening Ceremony - Welcome address
20:00-21:00	Opening Keynote Lectures
20:00-20:30	The effect of climate change on plant protection
20:30-21:00	Phytiatry as a distinct University science, for the benefit of global agriculture and forestry
21:00	Welcome Reception

Tuesday, 2 July

	BANQUET	SKALKOTAS	MC2	MC3	CONFERENCE I
08:30-11:00	1 Unlocking the potential of microbiomes for sustainable agriculture	2 Novel concepts and approaches in management of fruit flies (Diptera: Tephritidae)	3 Responses of plant genotypes to pathogens and development of novel techniques for improvement of crop resistance to diseases	4A Grapevine trunk diseases	5 Vascular wilts
11:00-11:30	Coffee Break - POSTER SESSION				
11:30-14:00	6A Advances with biopesticides to meet plant protection challenges	7A Understanding pathogen-vector-host interactions in globally important pathosystems	8 Networking tools to manage transboundary pests	4B Grapevine trunk diseases	9 New advances and technologies for postharvest diseases control
14:00-15:00	Light Lunch - POSTER SESSION				
15:00-15:30	Plenary Lecture Strategies for developing and implementing digital identification tools				
15:30-16:00	Plenary Lecture Enabling sustainable agriculture through understanding and enhancement of microbiomes				
16:00-16:30	Plenary Lecture Impact and control of transboundary/invasive banana wilt pathogen, <i>Fusarium oxysporum</i> f. sp. <i>Cubense</i>				



Program at a Glance

Tuesday, 2 July

	BANQUET	SKALKOTAS	MC2	MC3	CONFERENCE I
16:30-17:00	Satellite Lecture Effectiveness of Bait Sprays and Mass Trapping using Dacus Bait 100 for IPM of Bactrocera oleae: Field Results from Crete, Greece 				
17:00-18:00	6B Advances with biopesticides to meet plant protection challenges	7B Understanding pathogen-vector-host interactions in globally important pathosystems	10A Impact and control of transboundary/invasive pests		
18:00-18:30	Coffee Break - POSTER SESSION				
18:30-21:00	11 Canker disease of fruit, nut and vine crops	7C Understanding pathogen-vector-host interactions in globally important pathosystems	10B Impact and control of transboundary/invasive pests	12 Herbicide-Resistant Weeds - A Global Perspective	13 Incorporating indigenous knowledge into plant protection science

Wednesday, 3 July

	BANQUET	SKALKOTAS	MC2	MC3	CONFERENCE I
08:30 - 11:00	14 Microbiomes and their role in plant pathology	15 Components of IRM programs in an IPM framework	16 Diseases in tree crops and forests	17 NLR guided strategies for durable disease resistance in crop plants	18A Fungicide resistance: diagnosis, risk assessment and management, Integrated pest management
11:00-11:30	Coffee Break - POSTER SESSION				
11:30-14:00	19 Plant Health research coordination: an international endeavor	20A Biopesticides and biofertilizers	21 Selected highlights in plant protection	22 Management of biological Invasions in a changing world	18B Fungicide resistance: diagnosis, risk assessment and management, Integrated pest management
14:00-15:00	Light Lunch - POSTER SESSION				
15:00-15:30	Plenary Lecture Molecular diagnostics for rational use of pesticides and resistance management of agricultural pests				
15:30-16:00	Plenary Lecture Microbial pesticides: Discovery, piloting and scaling up in Africa				
16:00-16:30	Plenary Lecture Cross-kingdom RNA trafficking between plants and fungal pathogens				



Program at a Glance

Wednesday, 3 July

	BANQUET	SKALKOTAS	MC2	MC3	CONFERENCE I
16:30-17:30	Satellite Symposium BASF Axalion® active: redefining modern sustainable piercing® sucking insect control 				
17:30-18:00	Coffee Break - POSTER SESSION				
18:00-20:50	23 Plant Health - Phytiatry	20B Biopesticides and biofertilizers	24 Emergent forest diseases in a changing and interconnected world	25 Emerging pests with relation plant biosecurity and food safety	26 Environmental fate, ecotoxicology, risk assessment, and remediation of pesticide residues

Thursday, 4 July

	BANQUET	SKALKOTAS	MC2	MC3	CONFERENCE I
08:30 - 11:00	27A Biological control of insect crop pests	28 Sustainable mycotoxin management in a climate change future	29A Improving resistance - key to meet future challenges - Plant Breeding	30 Microbial interactions in ecosystems: negative or positive consequences on plant health	1 14 th IVS Recent Advances in Verticillium Biology through Genomics
11:00-11:30	Coffee Break - POSTER SESSION				
11:30-14:00	27B Biological control of insect crop pests	31 Tropical Pest Management - Challenges in the Post-Covid World	29B Improving resistance - key to meet future challenges - Plant Breeding	32 Eco-epidemiological and pathobiome perspectives on diseases caused by mycotoxigenic fungi	2 14 th IVS Recent Advances in Verticillium wilt Management
14:00-15:00	Light Lunch - POSTER SESSION				
15:00-15:30	Plenary Lecture Food security in Africa needs policy support for sustainable plant health management				
15:30-16:00	Plenary Lecture Weed Management Challenges and Opportunities to Close Yield and Profitability Gaps in Smallholder Rice Production in Asia				
16:00-16:30	Plenary Lecture Coordinated approach for transboundary plant pest and disease management				



Program at a Glance

Thursday, 4 July

	BANQUET	SKALKOTAS	MC2	MC3	CONFERENCE I
16:30-17:00	Satellite Lecture How Biologicals can support the transition to Regenerative Agriculture systems 				
17:00-17:30	Satellite Lecture Reklamel active: a novel tool for integrated nematode management - key learnings from a global nematocide development project 				
17:30-18:00	Coffee Break - POSTER SESSION				
18:00-20:30	33 Unveiling Nature's Arsenal: The Role of Volatile Compounds in Plant Protection	34 Advances in biosecurity measures for emerging and invasive pests	35 Chemical ecology and biological control	36 Challenges in sustainable plant disease management in a changing climate	3 14th IVS Recent Advances in Verticillium - plant interaction

Friday, 5 July

	BANQUET	SKALKOTAS	MC2	MC3	CONFERENCE I
08:30 - 11:00	37A Recent advances in plant virology	38 Precision agriculture and remote sensing for future plant disease management	39 Prevention and sustainable management of transboundary plant pests	40A Recent advances in vegetable IPM	4 14th IVS Biological Control of Verticillium wilt pathogens
11:00-11:30	Coffee Break - POSTER SESSION				
11:30-14:00	37B Recent advances in plant virology	41 Advances in nematode research and plant protection	42 Biological plant protection	40B Recent advances in vegetable IPM	43 Approach to integrated soil and plant health assessments in IPM systems
14:00-15:00	Light Lunch - POSTER SESSION				
15:00-17:30	44 Plant protection products	-	45 Xylella fastidiosa	46 Post-harvest pest and disease management	47 Molecular Plant Microbe Interactions
17:30-18:00	Coffee Break - POSTER SESSION				
18:00-20:30	48 Phytoplasmas and Viroids in world agriculture	49 Endophytes as bioinsecticides	50 -	51 Interactions between Plant Protection and Pollinators	52 Frost damage mitigation strategies for crops, organized by project LIFE-FROSTDEFEND



KEYNOTE LECTURES



BANQUET

Chair: Elvis “Short” Heinrichs (*IAPPS Secretary General, USA*)
Epaminondas Paplomatas (*Agricultural University of Athens, Greece*)

K01

THE EFFECT OF CLIMATE CHANGE ON PLANT PROTECTION

Bebber D.

University Of Exeter, United Kingdom

This talk discusses three major approaches to understanding how plant pathogens respond to climate change. Firstly, observational studies analyze statistical relationships between plant pathogen distributions and climatic variables, projecting potential future scenarios. Notably, large-scale studies highlight an expanding threat to global food security, with climate change implicated in the rise of damaging pests and diseases, particularly in major crop-producing regions like China. Moreover, meta-barcoding studies emphasize the influence of climate on soil fungal distributions, revealing temperature as a crucial factor. Secondly, mechanistic models aim to mathematically describe life cycle processes of plant pathogens under different abiotic conditions. These models, focusing on factors like temperature and moisture, suggest that global warming will cause a latitudinal shift in disease risk, affecting crop yields and potentially altering the suite of pathogens at specific locations. Thirdly, controlled experiments simulate disease impact under current and future climatic conditions. These experiments often examine interactions among multiple global change drivers, such as elevated atmospheric CO₂. Results indicate that higher CO₂ concentrations can increase fungal disease pressure in crops. Additionally, changing climate conditions impact plant disease management options, as demonstrated by improved efficacy of certain fungicides and biocontrol agents under experimental CO₂ and temperature increases. The overall conclusion is that crop pests and diseases, including fungi and oomycetes, respond to climate change due to their sensitivity to abiotic conditions. The methods used to study these responses have complementary strengths and weaknesses. Large-scale observational studies provide a holistic perspective, mechanistic models offer insights into life cycle responses, and controlled experiments reveal interactions among global change factors. Despite uncertainties, the text tentatively concludes that global change factors benefiting host crops may also benefit co-adapted pathogens, potentially negating any yield benefits unless adequately controlled.

K02

PHYTIATRY AS A DISTINCT UNIVERSITY SCIENCE, FOR THE BENEFIT OF GLOBAL AGRICULTURE

Eleftherios C Tjamos

Department of Plant Pathology, President of the Hellenic Society of Phytiatry, Greece

Correspondence: Eleftherios C Tjamos, Emeritus Professor in Plant Pathology, Agricultural University of Athens, 75 Iera odos str, 11855, Greece

Scientifically qualified specialists able to work as Phytiatry (plant medicine) doctors are needed world-wide. Several postgraduate programs worldwide offer studies on plant health specialization. However, phytiatry as a distinct University science starting from a bachelor's degree has not been introduced so far. Unfortunately, its establishment was unsuccessful regardless of the timeless scientific efforts. Obviously, Phytiatry doctors will be able to follow careers as diagnosticians in plant health clinics, as crop consultants, plant pest inspectors, extension specialists, phyto pharmaceutical industry consultants, small business owners, applied research specialists for research institutes, or other plant health professionals. This proposal constitutes a new pioneer movement for a universal effort to introduce Phytiatry in universities, so to elevate and uniform educational status and specialization, consequently creating a new attractive profession of Phytiatry – plant doctors. Phytiatry, as a University multidisciplinary science could include several agronomical and biological scientific disciplines, having as core phytopathology, entomology and nematology, weed science and phyto pharmacy, soil management and fertilizers etc. to install a five-year University course as stands for Veterinary Medicine internationally, offering to the future plant doctors the opportunity of obtaining the necessary skills and qualifications starting from an undergraduate level.



Keynote Lectures

Tuesday, 2 July

BANQUET

Chair: **Corne Pieterse** (*Plant-Microbe Interactions Group, Utrecht University, The Netherlands*)
Geoff Norton (*University of Queensland, Australia*)

K03

STRATEGIES FOR DEVELOPING AND IMPLEMENTING DIGITAL IDENTIFICATION TOOLS

Norton G.

University of Queensland, Australia

K04

ENABLING SUSTAINABLE AGRICULTURE THROUGH UNDERSTANDING AND ENHANCEMENT OF MICROBIOMES

Carrion V.

University of Malaga, Spain

K05

IMPACT AND CONTROL OF TRANSBOUNDARY/INVASIVE BANANA WILT PATHOGEN, *FUSARIUM OXYSPORUM* F. SP. CUBENSE

Arie T.¹, Kashiwa T.², Kodama M.³, Sasaki N.¹, Abe T.⁴, Asai S.⁴, Andress C.⁵, Aragon L.⁵, Gutierrez D.⁶, Cabrera R.⁶, Oscar C.⁷

1. Tokyo University of Agriculture And Technology (tuat), Fuchu, Tokyo, Japan, 2. JIRCAS, Tsukuba, Ibaraki, Japan, 3. Tottori University, Tottori, Japan, 4. RIKEN, Wako, Saitama, Japan, 5. UNALM, La Molina, Lima, Peru, 6. INIA, La Molina, Lima, Peru, 7. UNAS, Tingo Maria, Peru

The potato late blight famine caused by *Phytophthora infestans* lost 1 million lives in the late 1840s when the world population was 1.5 billion. It is comparable to the 6 million lives lost by COVID-19 today in the world population of 8 billion. Plant diseases, pests, and weeds have had a major negative impact on agricultural production, and indirectly have affected human lives. Although recent development of pesticides, resistant cultivars and transportation of the agroproducts seem to have reduced the impact of these issues, we have to anticipate pandemics of plant diseases related to the global warming in near future. Wilt, or, Panama disease, caused by *Fusarium oxysporum* f. sp. cubense (Focb) is an awful threatening of banana (*Musa* spp.) production. In the late 1980s, a new race (TR4) of Focb was reported from tropical Asia, and it spread to Southeast Asia including the Philippines, Australia, Africa, and now reached to the Latin America in 2019 and probably to Japan in 2022. Race TR4 isolates from Southeast Asia and other area form a unique cluster on the *Fusarium* phylogenetic tree based on rDNA-IGS, and are considered to have the same origin. In other words, seedlings, soil, agro-machinery, or human feet helped the transboundary pandemic of race TR4. Currently, we are fighting Peruvian Banana wilt in the SATREPS collaboration research project (supported by JICA/JST). The objectives are (1) analyzing the accessory chromosomes that control the pathogenicity of Focb, (2) developing and implementing LAMP primer sets to diagnose Focb and its races in about an hour, (3) developing race TR4-resistant bananas using eg. heavy ion beam irradiation, (4) establishing production of pathogen-free seedlings and educating agricultural organizations and farmers to use them, (5) analyzing microflora of disease suppressive soil, (6) establishing new biopesticides and plant activators to control the disease.



BANQUET

Chair: Sunday Ekesi (ICIPE, Kenya)
Dimitris Tsitsigiannis (Agricultural University of Athens, Greece)

K06

MOLECULAR DIAGNOSTICS FOR RATIONAL USE OF PESTICIDES AND RESISTANCE MANAGEMENT OF AGRICULTURAL PESTS

Vontas J.^{1,2}, Mavridis K.², Grigoraki L.², Roditakis E.³, Tsagkarakou A.⁴, Bass C.⁵, Van Leeuwen T.⁶

1. Agricultural University of Athens, Athens, Greece, 2. Institute of Molecular Biology and Biotechnology, Foundation for Research and Technology-Hellas (IMBB-FORTH), Greece, 3. Hellenic Mediterranean University, Department of Agriculture, School of Agricultural Sciences, Heraklion, Greece, 4. Hellenic Agricultural Organization "DIMITRA", Institute of Olive Tree, Subtropical Crops and Viticulture, Heraklion, Greece, 5. College of Life and Environmental Sciences, Biosciences, University of Exeter, Penryn, United Kingdom, 6. Department of Plants and Crops, Faculty of Bioscience Engineering, Ghent University, Ghent, Belgium

Pesticide resistant pests become increasingly difficult to control in agriculture, among others due to the dramatic reduction in the availability of active ingredients. It is therefore crucial to make rational decisions on pesticide use to ensure effective and sustainable pest control. However, resistance monitoring programs that inform on pest susceptibility and resistance are not yet common practice in agriculture. Modern molecular diagnostic platforms are urgently needed and they will only increase in importance. Several practical and modern molecular platforms have been developed for the detection of genetic traits and biomarkers, at very low cost. However, several factors determine the strength of a biomarker, while the significance and the interpretation of molecular diagnostic applications are critical for insecticide resistance management. Functional approaches for elucidating pesticide resistance mechanisms at the molecular level, including in vivo validation of the role of genetic traits when present alone or in combination in a pest using CRISPR/Cas9 methods, not only improve our understanding for the genetic basis of resistance, but also determine the diagnostic value of the genetic biomarkers in field populations. This research provides new opportunities for decision making in an operational meaningful way, for evidence base rational use of pesticides, within the frame of the EU Green Deal and integrated pest management strategies.

This work was supported by the European Union's HORIZON-IA (101136611 - NextGenBioPest) and the project "Innovations in Plant Protection for sustainable and environmentally friendly pest control, InnoPP - TAEDR-O535675 that is "Funded by the European Union- Next Generation EU, Greece 2.0 National Recovery and Resilience plan, National Flagship Initiative "Agriculture and Food Industry

K07

MICROBIAL PESTICIDES: DISCOVERY, PILOTING AND SCALING UP IN AFRICA

Ekesi S.

ICIPE, Kenya

K08

CROSS-KINGDOM RNA TRAFFICKING BETWEEN PLANTS AND FUNGAL PATHOGENS

Jin H.

University of California Riverside, USA



Keynote Lectures

Thursday, 4 July

BANQUET

Chair Emmanuel Tamo (*IITA Benin*)
Trevor Jackson (*Agresearch, New Zealand*)

K09

FOOD SECURITY IN AFRICA NEEDS POLICY SUPPORT FOR SUSTAINABLE PLANT HEALTH MANAGEMENT

Borgemeister C.

University of Bonn, Germany

K10

WEED MANAGEMENT CHALLENGES AND OPPORTUNITIES TO CLOSE YIELD AND PROFITABILITY GAPS IN SMALLHOLDER RICE PRODUCTION IN ASIA

Kumar V.

International Rice Research Institute (IRRI), Los Banos, Philippines

Rice is the staple food for over half of the world population, hence crucial crop for global food security. To meet the increasing global rice demand by 2050, an estimated additional 75 million tons of milled rice will be needed. The challenge is that this additional rice must be produced while using fewer resources and adapting to changing climate conditions to ensure food security, and environmental sustainability. Closing yield gaps caused by weeds could help in achieving this target. The drivers such as labor and water scarcity, rising production costs, and climate change are causing shifts in farming practices in Asian smallholder rice systems leading to changes in weed management practices. Farmers are transitioning from puddled transplanted rice (PTR) to more efficient systems like direct-seeded rice (DSR), flooded to alternate wetting and drying (AWD) irrigation system, and from hand-weeding to herbicide-based weed control. Weeds are the biggest constraint in the wide-scale adoption of these new systems (DSR and AWD) and many weed-related issues/challenges have emerged including (1) higher risk of yield losses due to higher weed infestation; (2) evolution of weedy rice which poses a serious threat to DSR sustainability; (3) shifts in weed flora towards difficult-to-control weeds; (4) increased dependence on herbicides resulting in the evolution of herbicide-resistant weeds; and 5) weed management becoming more knowledge-intensive. Additionally, large knowledge and information gaps exist in farmers' current weed management practices. A huge opportunity exists to close yield and profitability gaps of smallholder rice farmers caused by weed competition by (1) developing robust weed reconnaissance systems for monitoring troublesome weeds and characterize farmer's weed management practices at scale for problem targeting and early warning, (2) developing and deploying robust integrated weed management (IWM) programs, and (3) creating enabling environment for wide-scale adoption of IWM. Examples of these interventions will be presented.

K11

COORDINATED APPROACH FOR TRANSBOUNDARY PLANT PEST AND DISEASE MANAGEMENT

Fazil Dusunceli

FAO, Turkiye



**CONCURRENT
SESSIONS**



BANQUET

08.30-11.00 Concurrent Session 1

Unlocking the Potential of Microbiomes for Sustainable Agriculture

Chair: Altier Nora (*Instituto Nacional de Investigación Agropecuaria, INIA Las Brujas, Uruguay*)
Eduardo Abreo (*Inia Uruguay*)
Linda Kinkel (*University of Minnesota, USA*)

SE01 C01

UNLOCKING THE SECRETS OF PLANT MICROBIOMES

Victor Carrión

Department of Microbiology and Crop Protection, Institute for Mediterranean and Subtropical Horticulture 'La Mayora', Universidad de Málaga, Málaga, Spain

Plant roots are colonized by an astonishing number of microbes, not merely externally (rhizosphere) but also internally (endosphere). In the past years, extensive research has revealed the role of specific members of the plant microbiome to the improvement of plant tolerance to (a) biotic stress factors. However, the molecular mechanisms underlying these functions remain largely elusive. This in turn has raised new questions such as how endophytic bacteria colonize and protect plants against pathogens; and which genes are essential for being pathogenic or beneficial for the plant? In other words, are there genetic markers associated with specific microbial lifestyles? Here, our newest results are presented to shed light on these outstanding questions. We used in-depth metagenomic sequencing of the endosphere to discover that chitinase and novel NRPS and PKS genes were only activated in endophytic microbes upon pathogen infection. We also developed a new computational tool (MicroLife) to explore large-scale genomic datasets and to identify genes as well as biosynthetic gene clusters (BGCs) associated with specific microbial lifestyles. Using MicroLife, we were able to dig into the microbial dark matter to discover novel genes and BGCs involved in pathogenesis and endophytic lifestyles. Collectively, our results provide new exciting insights into the largely unexplored functional potential of the plant microbiome and the chemical secrets of the endosphere.

SE01 C02

EXAMINING THE MICROBIOME THROUGH THE LENS OF THE ONE HEALTH CONCEPT

Gabriele Berg

Institute of Environmental Biotechnology, Graz University of Technology, 8010 Graz, Austria; Leibniz-Institute for Agricultural Engineering Potsdam, 14469 Potsdam; University of Potsdam, 14476 Potsdam, Germany

Plant microbiomes are key components for ecosystem health as well as for one health. The latter is a concept integrating the health of people, animals, plants into their environment (WHO). The plant microbiota, which consist of bacteria, archaea, protists and fungi, is vertically transmitted by seeds and replenished horizontally from soil. All plants are holobionts and form a functional unit with its microbiome. Plant diversification and co-evolution shaped the plant

microbiome and designed their specific composition and functional interplay including natural biocontrol of pathogens. Human activities in the Anthropocene, and especially intense agriculture, are linked to a significant shift of diversity and evenness of the plant microbiota. This shift is characterized by a decrease of host specificity and symbionts, and an increase of r-strategic microbes, pathogens, and hypermutators. Findings from plant microbiome research over the past 20 years clearly call for management of the microbiome and joint attention to the crop holobiont. Plant microbiomes can be managed either directly by applying (i) microbiome transplants, (ii) microbes with beneficial properties, or (iii) microbiota-active metabolites, or indirectly by changing environmental conditions in a way that microbiomes also shift their structure and function from dysbiosis into a healthy state. Examples for the different strategies for plant protection will be presented, and risk associated with the technology will be discussed. Beyond, the plant microbiome is connected across systems and crucial for planetary health issues as well.

SE01 C03

WHAT DETERMINES THE FUNCTIONAL CAPACITIES OF ENDOPHYTIC AND SOIL MICROBIOMES?

Linda L. Kinkel

Department of Plant Pathology, University of Minnesota, St. Paul, MN, USA

Plant-associated microbiomes play critical roles in mediating crop productivity. The promise of microbiome management for agriculture is compelling, but our understanding of the factors that mold the composition, diversity and functional capacities of plant microbiomes remains limited. While much work has focused on the roles of plant host, management, or habitat on the composition and functions of plant-associated microbiomes, we know much less about the role of microbial species interactions in mediating the ecological and evolutionary dynamics of microbiome functional capacities. Competition for resources, as well as complex, multi-species antagonistic, mutualistic, and cross-kingdom signaling interactions can all impact the both the potential for selective enrichment of beneficial or pathogenic microbes in the microbiome, as well as the real-time functional capacities of specific microbes. Moreover, species interactions within the microbiome are themselves impacted by plant host, management, and habitat. Here I will explore the roles of microbial species interactions, including within soil microbiomes and among foliar endophytes, in determining the composition and functional capacities of rhizosphere and endophytic populations. Focusing on nutrient use phenotypes, resource competition, and antagonistic species

interactions among bacterial and fungal populations in the microbiome, this work will highlight the significant roles that microbial species interactions within the microbiome play in determining the functional capacity of the microbiome. Simultaneously, our work highlights the significant roles of plant host, management, and habitat in determining the trajectories of species interactions within the microbiome, thereby shedding light on practical strategies for managing microbial interactions to achieve beneficial plant outcomes.

SE01 C04

MANAGING SOIL AND ROOT-ASSOCIATED MICROBIOMES IN AGROECOSYSTEMS

Doreen Babin, Victoria Cerecetto, Ioannis D. Kampouris, Andrea Braun-Kiewnick, Bunlong Yim, Rita Grosch, Carolina Leoni, Kornelia Smalla

Julius Kühn-Institut, Braunschweig, Germany

Soil microorganisms are key players of soil and plant health. Harnessing this potential for microbiome-based solutions might contribute to reduce agrochemical inputs and improve agricultural sustainability. At first, this requires a better understanding of factors shaping the soil and rhizosphere microbiome in agroecosystems. By using long-term field experiments located in different climatic regions, we could show that not only soil (e.g. soil type) and plant (e.g. developmental stage, cultivar) characteristics but also agricultural practices (e.g. tillage, fertilization) affect the structure and functionality of soil and root-associated microbiomes. This highlights the potential of agricultural practices to steer the soil microbiome into a more beneficial state supporting soil and plant health. For instance, we observed in field experiments in Uruguay that soils under long-term conservation practices such as pasture, reduced tillage and organic fertilization exhibited a distinct microbiome, which differed from conventionally managed soils and likely contributed to the improvement of soil structure and plant yields. Another option to manage microbiomes is to add plant-beneficial microorganisms by inoculation. I will present examples from greenhouse and field trials, where we inoculated microorganisms as single strains or as consortia to promote plant growth or reduce disease symptoms caused by soil-borne fungal pathogens. The efficacy to promote plant performance was not only dependent on the strain but also on (a)biotic stress conditions (e.g. drought, pathogen concentration). Interestingly, the inoculation with plant-beneficial microorganisms resulted in a modulation of the indigenous rhizosphere microbiome which varied e.g. based on the agricultural history of the soil and likely contributed to the inoculants' efficacy. These interdisciplinary studies under close-to-practice conditions provide important insights into a better understanding of options and the ecology of microbiome management, paving the way towards microbiome-based agricultural solutions in the future.

SE01 C05

MICROBIOMES IN AGRICULTURAL SETTINGS: INVISIBLE PARTNERS REVEALED

Eduardo Abreo, Pablo Torres, Andrés Villar, Pablo Fresia, Elena Beyhaut, Nora Altier

Biological Inputs, Inia Uruguay

Plant microbiomes comprise a diversity of entities that, through multiple interactions, determine processes in the plant soil continuum. The massive amplification of 16S and ITS can help to uncover the invisible partners that can have a positive or negative effect on plant performance. In this presentation, we will explore different practical approaches based on metabarcoding metagenomics in two situations and crops. First, I will present how the soil rhizospheric bacterial community in a soybean crop is affected by phosphorus fertilization and the seed inoculation of a P-biofertilizer based on *Priestia megaterium* (ex *Bacillus megaterium*). Last, I will show how the fungal and bacterial community in Eucalyptus cuttings could be associated with the rooting process, as determinants of successful or failed rooting of the new plantlet. In the first case, metagenomics is being used "backwards", to understand the consequences of management options (superphosphate fertilization vs P-biofertilizer) on the microbial community of the rhizosphere and generate novel hypothesis. In the second scenario, metagenomics is used "forward" to uncover the microbial status of cuttings that could be used as a guide to improve rooting through variations in management, including the reduction of hidden pathogens associated with failure or the isolation of key beneficial species associated with successful rooting.

SE01 001

VARIABLE EFFECT OF MICROBIOME MANIPULATION IN THE SOUTHERN GREEN STINK BUG NEZARA VIRIDULA AND ITS IMPLICATIONS FOR THE SYMBIOTIC CONTROL

Prieto S.¹, Brunetti M.², Magoga G.³, Orrù B.¹, Gonella E.¹, Montagna M.^{3,4}, Alma A.¹

1. Department of Agricultural, Forest and Food Sciences, University of Turin, Grugliasco, Italy, 2. Department of Agricultural and Environmental Sciences – Production, Landscape, Agroenergy, University of Milan, Milan, Italy, 3. Department of Agricultural Sciences, University of Naples Federico II, Portici, Italy, 4. Interuniversity Center for Studies on Bioinspired Agro-Environmental Technology (BAT Center), University of Naples Federico II, Portici, Italy

Many insects in the Pentatomidae family are responsible for losses in agriculture due to their phytophagous feeding habit, affecting both the yield and the quality of seeds and fruits. Pentatomids establish beneficial symbiotic interactions with gut bacteria vertically transmitted by the females via egg surface contamination. The symbiotic control is a pest management strategy based on the disruption of symbiont acquisition by spraying the egg surface with anti-symbiont agents, still the insect response to symbiont loss determines the efficacy of this strategy. In here, the symbiotic control was tested in an Italian population of the southern green stink bug *Nezara viridula* L. (Hemiptera, Pentatomidae), both in field and laboratory trials, by performing an anti-symbiont treatment on the egg masses. Moreover, the nymphal gut microbiome was characterized after symbiont disruption. The symbiotic control had a low impact on *N. viridula* in field, however in the

laboratory the mortality increased in nymphs deriving from treated egg masses and the symbiont abundance declined. The microbiome of control nymphs with low mortality was dominated by the symbiont, while a dysbiotic scenario was observed in treated nymphs and in control nymphs with high mortality. Nonetheless, symbiont elimination was not always associated with high mortality and some control nymphs were as well symbiont deprived. Five variants of the symbiont taxonomically assigned to *Pantoea* were recorded, and non-treated nymphs with low mortality showed higher variant diversity. Altogether, this study suggests an incomplete dependence of *N. viridula* on its primary symbiont. Still, the anti-symbiont treatment increased mortality under laboratory conditions, suggesting that a symbiont-targeted control strategy may be incorporated as a supplementary tool against *N. viridula* in a high pest pressure condition. Further studies will address the variability in the insect response to anti-symbiont treatment in different geographical populations along with a symbiont whole genome description.

SE01 002

"CRY FOR HELP" UPON PATHOGEN ATTACK ACROSS THE BRASSICACEAE FAMILY

Uribe Acosta M.¹, Aragón Gómez M.², Carracedo Lorenzo Z.², Beschoren da Costa P.², Zhou X.⁴, Spooren J.¹, Gols R.², Kloth K.², Berendsen R.¹, Stringlis I.³, Dicke M.², Pieterse C.¹

1. Utrecht University, Utrecht, Netherlands, 2. Wageningen University, Wageningen, Netherlands, 3. Agricultural University of Athens, Athens, Greece, 4. Nanjing Normal University, Nanjing, China

Following infection, *Arabidopsis thaliana* recruits beneficial rhizobacteria that stimulate plant immunity against pathogens, a phenomenon termed "cry for help". It is unknown whether pathogen-triggered recruitment is conserved among different members of the Brassicaceae family. To assess this, we treated 22 *Brassicaceae* species with defense hormones methyl jasmonate (MeJA) and salicylic acid (SA) as pathogen attack mimics, and collected roots for microbiome analysis. To identify microbial taxa responsive to the treatment we performed (i) 5 differential abundance tests, (ii) neutral models and (iii) random forest analysis on each plant species comparing the control with the treated group and selecting only microbial families deemed responsive by all these analyses simultaneously. In the fungal communities no families were deemed responsive by all analyses; in contrast, 7 bacterial families were responsive. Comamonadaceae, Chitinophagaceae and Streptomycetaceae were responsive to both treatments, family Oxalobacteraceae only to MeJA and families Rhizobiaceae, Xanthomonadaceae and Xanthobacteraceae only to SA. None of these responsive microbes showed the same behavior across all plant species, and only one ASV, genus *Niastella*, was enriched in at least 19 of the 22 plant species upon MeJA treatment. Despite that, 9 genera were enriched in at least 60% of the plants, 7 of them only on one of the two treatments, but *Streptomyces* and *Niastella* in both MeJA and SA. These results show that while not all members of *Brassicaceae* recruited the same bacterial families, there are some clearly conserved recruitment patterns in more than half of the plant species. These bacterial genera potentially have a stronger and distinct

evolutionary relationship with the *Brassicaceae* family. Now we are analyzing whether there is a phylogenetic signature between the plants and these responsive microbial groups.

SE01 003

CONTINENTAL SCREENING OF THE BIOCONTROL YEAST *AUREOBASIDIUM PULLULANS* ACROSS EUROPE: THE CASE FOR LOCAL ADAPTATION AND MINING

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Studying the evolution of beneficial microorganisms in natural environments could provide a foundation for mining and implementing more effective biocontrol agents for sustainable agriculture. The black yeast-like fungus *Aureobasidium pullulans* is a suitable model species for studying the evolution of biocontrol potential features due to its presumed adaptability to different environmental conditions. *Aureobasidium* spp. are essential members of the European wild strawberry (*Fragaria vesca*) phyllosphere regardless of the environmental conditions of the habitat from which the plants were collected. This study aims to investigate the biocontrol potential of local *A. pullulans* wild strawberry plants across Europe. Over 200 genotypes of the host plant *F. vesca* from diverse habitats across Europe were collected, from which *A. pullulans* strains were isolated. Selected strains were investigated using a multi-locus-based approach (internal transcribed spacer [ITS] and partial elongase gene [ELO]) and in vitro dual culture plate assay to screen antagonistic activity against pathogenic *Botrytis cinerea*, which causes grey mould disease in strawberry. The *A. pullulans*-induced reduction in the growth rate of *B. cinerea* varied between 17% and 44%. Interestingly, *A. pullulans* strains from northern Europe varied widely in their antagonistic potential, while central and southern strains showed much lower variation. Most *A. pullulans* strains from central and southern Europe reduced the pathogen's growth by 22-32%. No interrelations between the groupings of the yeast strains based on ITS- and ELO-phylogenetic trees were discovered. However, the preliminary results suggest that local environmental factors correlate with the biocontrol potential of *A. pullulans* strains.

SE01 C06

FINAL CONSIDERATIONS: HOW DO MICROBIOMES CONTRIBUTE TO PLANT PROTECTION SCIENCES AND THE CONCEPT OF ONE HEALTH?

Nora Altier

Instituto Nacional de Investigación Agropecuaria, INIA Las Brujas, Uruguay

The management of plant-associated microbiomes offers the possibility of addressing plant protection and plant nutrition issues for sustainable agriculture within the One Health concept, to acknowledge that the health of plants, animals, people and the environment are interlinked and interdependent. Interactions within microbiomes have a deep impact on soil, plant, and agroecosystem health, which in



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turn affects soil fertility, crop productivity, and food quality and safety. Beneficial microbes have been usually studied in isolation as inoculants, endophytes, or applied microbial products, neglecting the complex networks that determine the species interactions within the agricultural microbiomes where they are embedded. Additionally, the factors that shape the composition, diversity, and functional capacities of plant microbiomes and the genetic factors involved in microbiome assembly remain limited. A better understanding of microbiome structures and functions will determine common threads leading to beneficial activity and will contribute to the redesign of production systems. The invited speakers have explored these topics, connecting fundamental studies on plant microbiome to practical strategies of Integrated Crop Management, addressing future needs and discussing pathways towards microbiome-based agricultural solutions. The concurrent contributions have shed light on new biological, chemical, and breeding strategies to assure an agriculture with reduced agrochemical inputs, without jeopardizing food production and security. The session has provided a unique opportunity for researchers to discuss research outputs from diverse agroecosystems and build collaborations to advance the understanding and enhancement of microbiomes and their role in plant health, while posing significant challenges for agricultural sustainability towards human welfare.



SKALKOTAS

08.30-11.00 Concurrent Session 2

**Novel Concepts and Approaches in Management of Fruit Flies
(Diptera: Tephritidae)**

Chair: Nikos Papadopoulos (University of Thessaly, Greece)
Souleymane Nacro (INERA, France)

SE02 C01

INTRODUCTION TO MANAGEMENT OF FRUIT FLIES

Nick Papadopoulos and Souleymane Nacro

SE02 C02

**ENHANCED SIT TO ERADICATE AND CONTAIN SMALL
OUTBREAKS OF FRUIT FLIES**

Antonios Avgoustinos

SE02 C03

NOVEL CONCEPTS IN STERILE INSECT TECHNIQUE

Marc F. Schetelig

*Justus-Liebig-University Giessen, Department of Insect
Biotechnology in Plant Protection, Giessen, Germany*

The Sterile Insect Technique (SIT) is a fundamental approach to eco-friendly pest management, effectively diminishing pest populations through disseminating sterilized male insects. Nonetheless, the broader adoption of SIT has been constrained by the necessity for effective and economically viable sexing methods to facilitate the release of only male insects. The creation of Genetic Sexing Strains (GSS) has significantly transformed these procedures, exemplified by the remarkable achievements with the Mediterranean fruit fly (Medfly) GSS. This achievement was realized through the enduring process of classical mutagenesis, extending beyond two decades. The evolution in genomics and gene editing technologies has equipped us with insights into the genetic framework of the Medfly GSS, culminating in the identification of temperature-sensitive lethal (tsl) factors. The unveiling of tsl factors has paved the way for replicating the Medfly GSS attributes in other species more efficiently and without deploying transgenic methods. This presentation will delve into the tsl factor's discovery in Medfly, its potential for fostering the development of novel GSS, and a comparative analysis between the traditional 'classic GSS' methodology and our updated 'neo-classical' strategy.

SE02 C04

**EFFECTIVENESS OF FOUR INTEGRATED PEST
MANAGEMENT APPROACHES IN THE CONTROL OF
FRUIT FLIES (DIPTERA: TEPHRITIDAE) IN MANGO AGRO-
ECOSYSTEMS IN THE SOUTH-SUDANIAN ZONE OF
BURKINA FASO**

**Issaka Zida¹, Karim Nébélé¹, Alizéta Sawadogo¹, Souleymane
Nacro²**

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Bobo-Dioulasso 01, Burkina Faso, 2. Centre Régional de
Recherches Environnementales, Agricoles et de Formation
de Kamboinsé / Institut de l'Environnement et de Recherches
Agricoles / 01 BP 476 Ouagadougou 01, Burkina Faso*

This study evaluated the effectiveness of four Integrated Pest Management (IPM) approaches in the control of *Bactrocera dorsalis* (Hendel) [Diptera: Tephritidae] and *Ceratitis cosyra* (Walker) [Diptera: Tephritidae] during two consecutive mango fruiting seasons (2018 and 2019) in the south-Sudan zone of Burkina Faso. These approaches, including sanitation + M3 bait station (SM), sanitation + protein GF-120 bait (SG), sanitation + Timaye + M3 bait station (STM) and sanitation + Timaye + GF-120 bait (STG), were implemented in 12 mango orchards in three provinces of the country. In each province, one mango orchard was used as control. Flies per trap per week (FTW) and damage indices were assessed in treated orchards compared to the control orchards. The efficacy rate of each IPM approach in protecting mango against fruit fly attacks was also determined. The STG approach was the most effective in reducing both *B. dorsalis* and *C. cosyra* FTW with the best efficacy rate. Further research should emphasize indigenous and affordable attract-and-kill tools for resource poor farmers.

SE02 C05

**CERATIS CAPITATA (DIPTERA: TEPHRITIDAE) INVASION
SUCCESS: INSIGHTS INTO COLD TOLERANCE AND
OVERWINTERING CAPACITY IN NOVEL ENVIRONMENTS**

**Georgia Papadogiorgou, Eleni Verykoui, Antonis
Papadopoulos, Nikos Papadopoulos**

University of Thessaly, Volos, Greece

The Mediterranean fruit fly (medfly), *Ceratitis capitata*, is an invasive pest, that is currently expanding its geographic distribution from the Mediterranean coasts to more temperate areas of Europe. Biological and physiological traits such as (a) thermal plasticity, (b) strong dispersal ability and (c) quick adaptation seem to enable medfly persistence and thriving in novel environments. Additionally, distribution and abundance



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of *C. capitata* depend on several abiotic and biotic factors. Given that temperature and host availability are primary determinants of *C. capitata* geographic range boundaries, especially in the Northern Hemisphere with pronounced seasonality, we investigated the latitudinal variation in a) acute cold stress response, and b) the overwintering capacity of different populations under a range of temperature conditions. To estimate the response to acute cold stress, 5-day-old adults acclimated at either 10, 20, or 30°C for 5 consecutive days were subjected to a standard cold stress of -5.5°C for 1h. The overwintering capacity was assessed in two key overwintering hosts (apples and bitter oranges) under three constant temperatures. Results revealed that acclimation to low temperatures improved survival to acute cold stress, with southern populations being more sensitive to subfreezing temperatures. Furthermore, climatic analysis of the area of the population origin showed combined effects of latitude, host and macroclimatic variables on immature survival and development rates. Survival rates varied among populations and hosts. Medflies from regions with higher temperature variation displayed prolonged developmental durations, indicating increased plasticity and higher overwintering capacity. Overall, our results contribute towards understanding the invasion dynamics of medfly and the biological traits that influence invasion success, with emphasis on range expansion to northern, more temperate areas of Europe.

SE02 C06

A NOVEL METHODOLOGICAL APPROACH TO DETECT LOW-DENSITY POPULATIONS OF THE MEDITERRANEAN FRUIT FLY TO SUPPORT EFFICIENT ERADICATION AND CONTAINMENT APPROACHES

Eleftheria-Maria Bali, Eleni Verykoui, Vasilis G. Rodovitis, Nikos T. Papadopoulos

Laboratory of Entomology and Agricultural Zoology, Department of Agriculture Crop Production and Rural Environment, University of Thessaly, Volos, Greece

Fruit flies are among the most important pests of fruit commodities worldwide. Conventional pest management approaches primarily focus on controlling high-density populations through insecticides, biological control, and cultural practices. However, effective detection and management of low-density populations are critical for preventing outbreaks and implementing timely targeted eradication and control measures to inhibit further spread and minimize economic damage. Adult trapping serves as the primary method for early detection of invasive tephritid populations and monitoring of established ones. In an effort to understand the detection level of low-density populations of the Mediterranean fruit fly (medfly, *Ceratitis capitata*) in ecological different field settings, we conducted release recapture studies using varying population levels and both males and females. Four 1 km² plots with different densities of host plants were selected. Five trapping stations consisted of a Tephri trap baited with Biolure (ammonium acetate, putrescine and trimethylamine) and a Jackson trap baited with trimedlure, were established in each plot following the well-established deployment plan considered in California, USA. Four release recapture trials were conducted in each plot with 5-day old adults. A low and a higher density with

differently color-marked adults were used. Trapping stations were inspected at regular intervals and captured adults were examined in laboratory under a stereoscope using ultraviolet light to determine their marking color. The density level of released flies did not affect fly captures. The recapture probabilities of medflies remain extremely low (0.7%). The percentage of recaptured males exceeded that of females. The distance between the randomly selected release point and trapping station was a significant predictor of fly captures with closer proximity resulting in higher capture rates. The importance of these findings to understand the biology of low-density populations as well as to address the effective implementation of eradication and containment strategies is discussed.

SE02 C07

THE FALLACY OF THE CLASSIC IPM PARADIGM IN MANAGING FRUIT FLIES OF TROPICAL ORIGIN

Slawomir A. Lux

inSilico-IPM, Poland

The Mediterranean fruit fly (Medfly), *Ceratitis capitata* (Wiedemann), is a pest of Afro-tropical origin that has adapted to the Mediterranean climate. In most locations it is severely decimated in winter, but a limited number of fertile adults or immature individuals can survive even in northern Italy, Croatia or Austria. To be detected, the overwintering Medfly must first reproduce to increase its population density to a detectable level, so, the farmer is warned late, when the Medfly has already entered the phase of exponential growth and is difficult to control by methods other than immediate-acting pesticides. Therefore, the classic IPM paradigm based on monitoring and economic action thresholds does not seem optimal for control of tropical fruit flies, such as Medfly, which warrants its critical reappraisal. The advantages of the postulated OFFf-season Medfly control were evaluated *in silico* using a «virtual farm» approach - simulations of the local Pest-Terrain-Weather-IPM system using the *PESTonFARM* model. The simulations were conducted for three hypothetical farms located within the 1,000 km latitudinal range of Medfly presence in Italy: Sicily, Lazio and Trentino-South Tyrol in the north. Our simulations show that preventive IPM applied very early in the season, using only the Attract&Kill baited panels, can significantly reduce pest pressure or even virtually nullify population build-up in late summer. This approach has not only proven to be effective compared to current pesticide-based practice but can eliminate or significantly reduce the use of pesticides and is economically viable, despite being more expensive. The postulated paradigm shift will also be applicable in the case of other invasive flies of tropical origin. Implementation of the postulated OFF-season IPM, carried out on farms in Greece, Italy and Spain, produced encouraging results and received positive feedback from farm owners and stakeholders.

SE02 001

DUAL TEPHRITID FRUIT FLY AUTOMATED MONITORING SYSTEM FOR ESTABLISHING PEST FREE PRODUCTION FOR THE GREEN HOUSE TOMATO

Chou M.^{1,2}, Lin H.³, Chuang Y.³, Yeh S.³, Lin S.³, Chen C.⁴, Chen C.⁴, Hsu J.⁵, Jiang J.³

1. National Chung Hsing University, Agricultural Extension Center, Taichung, Taiwan, 2. National Chung Hsing University, Department of Entomology, Taichung, Taiwan, 3. National Taiwan University, Department of Biomechanics Engineering, Taipei, Taiwan, 4. Animal and Plant Health Inspection Agency (APHIA), Ministry of Agriculture (MOA), Taipei, Taiwan, 5. National Taiwan University, Department of Entomology, Taipei, Taiwan

Tephritid fruit flies pose a significant threat to global agriculture, substantial economic losses, and quarantine restrictions. The conventional monitoring methods are labor-intensive and time-consuming. More importantly, it lacks real-time feedback for the decision-maker to detect early invasion. An innovative approach to monitoring oriental fruit fly (*Bactrocera dorsalis* Hendel) and melon fly (*Zeugodacus cucurbitae* (Coquillett)) through the implementation of an automatic monitoring system was tested for the greenhouse tomato production in central and southern Taiwan. The objective of the research is to establish an automatic data report system to meet the phytosanitary requirement for pest-free production of export commodities. The system provides the functions of continuous data collection and data reporting. It served as a detection tool in the greenhouse environment to provide real-time data collection and an early warning system. Surveys from 2020 to 2023 showed that two production sites have recorded more than two years of zero fruit fly invasion, and the production site that joined in 2023 has achieved one year of zero fruit fly invasion. The system provides high accuracy with an average rate for false positive detection of $5.05 \pm 2.25\%$ from 2020 to 2023, and it is reduced to 0% with an update on the system's design. The system also served as a management decision assistant tool to collect the fruit fly density data outside the greenhouses. Data collected from three testing sites showed that the system provided a high data accuracy rate of $90.32 \pm 2.87\%$. Up to $96.59 \pm 1.34\%$ of data is acceptable for pest management decision-making when we set the action level at 1 fly/trap/day. There was one event of detection error throughout the three-year trial. The average detection error rate of $0.57 \pm 0.57\%$ indicated that the system could sustain itself in a complex natural environment while providing robust and reliable data collection to support decision-making. The proposed automatic monitoring system represents a significant leap forward in tephritid fruit fly management. The system provides an efficient solution to address the challenges associated with conventional monitoring methods.

Table 1. 2021-2023年溫室內自動計數蟲桶斷訊、故障及誤觸紀錄

Machine type	Location	Year	N_sample	斷訊	故障	誤觸	虛報率	可能原因
V1	嘉義	2020	146	0	38	8	5.48%	雨
		2021	365	2	17	32	8.77%	
		2022	365	0	0	0	0.00%	
		2023	201	23	0	0	0.00%	
	雲林	2020	146	0	25	27	18.49%	蜘蛛網、熱老化?
		2021	365	1	48	11	3.01%	
		2022	365	0	1	1	0.27%	
		2023	113	0	0	22	19.47%	
V2	嘉義	2023	164	10	0	0	0.00%	
	雲林	2023	252	4	0	0	0.00%	
	彰化	2023	125	0	0	0	0.00%	

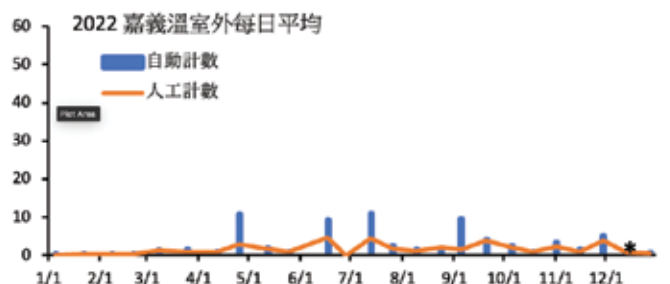
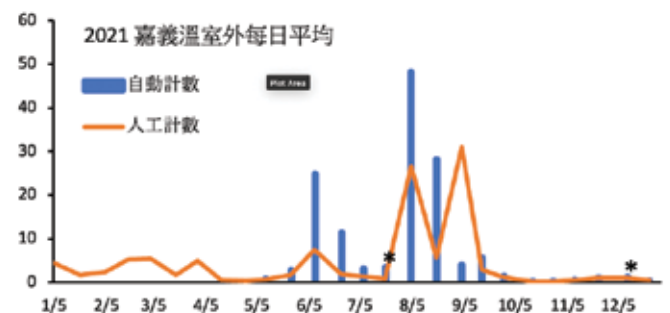
Table 2. 溫室外自動計數數據與人工調查數據比較

Location	Year	No. Sample	完全正確	可接受	偽陽 False positive*	偽陰 False-negative**	偵測錯誤 Detection error***
嘉義	2021	17	0	15	2	0	0
	2022	25	6	18	1	0	0
	2023	25	5	14	5	1	0
雲林	2021	13	0	12	0	1	0
	2022	25	2	20	1	1	1
	2023	24	5	14	1	1	0
彰化	2023	9	4	5	0	0	0

*不須噴藥但自動計數數據判斷需噴藥(自動計數每日蟲數平均 ≥ 1)

**需噴藥但自動計數數據判斷不需噴藥(自動計數每日蟲數平均 < 1)

***自動計數數據顯示0, 但人工計數有蟲



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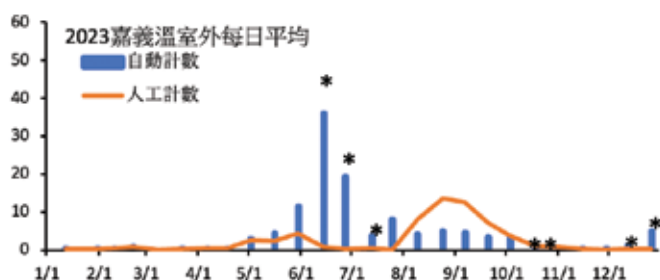


Figure 1. 嘉義生產點外瓜、果實蠅自動監測與人工計數比較
*不須噴藥但自動計數數據判斷需噴藥(自動計數每日蟲數平均 ≥ 1)
**需噴藥但自動計數數據判斷不需噴藥(自動計數每日蟲數平均 < 1)
***自動計數顯示0, 但人工計數有蟲

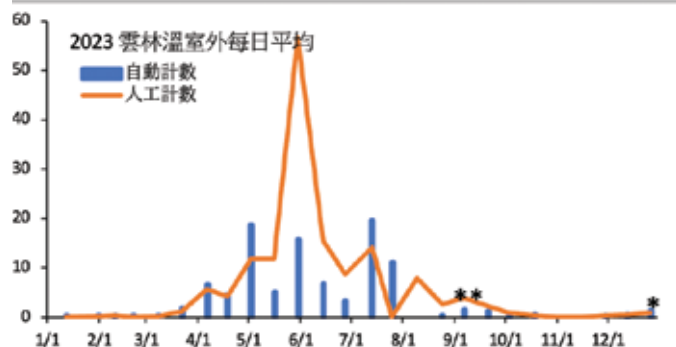
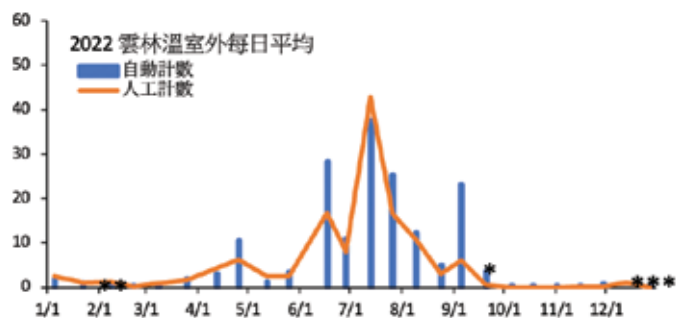
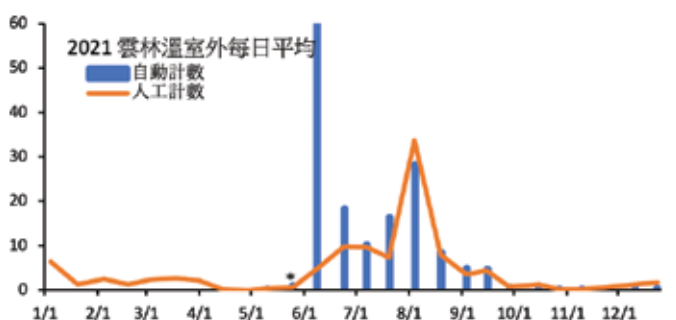


Figure 2. 雲林生產點外瓜、果實蠅自動監測與人工計數比較
*不須噴藥但自動計數數據判斷需噴藥(自動計數每日蟲數平均 ≥ 1)
**需噴藥但自動計數數據判斷不需噴藥(自動計數每日蟲數平均 < 1)
***自動計數顯示0, 但人工計數有蟲

SE02 002

USE OF THE PESTonFARM MODEL FOR THE MANAGEMENT OF CERATITIS CAPITATA (DIPTERA: TEPHRITIDAE) POPULATIONS

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Mediterranean fruit fly (medfly), *Ceratitis capitata* (Diptera, Tephritidae), is considered one of the most damaging pests in the world. It can attack several commercially important fruits causing considerable economic damage. The management of medfly infestation in fragmented and heterogeneous landscapes, such as in the European countries, has been done mainly with the use of synthetic insecticides. With the aim of contributing to the Integrated Pest Management (IPM) of this fruit fly, we report results obtained in 2022 and 2023 on the application of management scenarios generated by PESTonFARM model. This approach uses an agent-based simulation of the behaviour of medfly females based on the combined effects of farm structure, fruit phenology, spatio-temporal distribution and density of the pest, type of control techniques, and local weather patterns. The simulations obtained in the two years were based on data collected in three Italian farms, characterized as mixed orchards and with IPM based mainly on insecticide treatments. The produced scenarios include socio-economic analysis of the farms considering IPM costs and fruit value. Specifically, the following surveys were carried out: evaluation of host fruit phenology, identification of medfly overwintering resources, monitoring of the adult population, estimation of the level of fruit damage, and collection of basic socioeconomic information. The best scenarios proposed by the PESTonFARM model for the two years were used to control the medfly populations in the experimental farms. The scenarios produced were found to be adherent to the data collected in the field, showing that the model can be used to improve the medfly IPM. In particular, its application in both years allowed a reduction in the use of insecticides, in comparison with previous years and control farm, in favour of eco-friendly control techniques and improved the economic balance of the farms where the suggested scenarios were applied.



MC2

08.30-11.00 Concurrent Session 3

Responses of Plant Genotypes to Pathogens and Development of Novel Techniques for Improvement of Crop Resistance to Diseases

Chair: Shuxian Li (*Crop Genetics Research Unit, USDA, ARS, USA*)

SE03 C01

AN OVERVIEW OF RESEARCH ON EXPLORING RESPONSES OF PLANTSTO PATHOGENS AND DEVELOPMENT OF NOVEL TECHNIQUES FOR IMPROVEMENT OF CROP RESISTANCE TO DISEASES

Shuxian Li

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Many microbial pathogens attack crop plants resulting in significant yield losses that threaten global food security. The world's rising population exceeding 9 billion by 2050, along with climate change and the threat of new pathogens, pose significant challenges to the sustainability of world agriculture development. Understanding how plants perceive and respond to pathogens is of the utmost importance to ensure sustainable agricultural practices and mitigate crop losses. In addition, exploring the molecular, physiological, and genetic mechanisms underlying plant defenses against diseases hold great potential for developing novel strategies and techniques for disease management. The special session on "Responses of plant genotypes to pathogens and the development of novel techniques for improvement of crop resistance to diseases" invites speakers to delve into the different aspects of plant responses to pathogens and to provide insights into advancement in plant phenotyping and genotyping methods, as well as virus-induced gene silencing and CRISPR/Cas9-assisted engineering and other novel techniques as tools for the improvement of crop resistance to diseases.

SE03 C02

THE DISCOVERY OF TANDEM KINASE R-GENES: ORIGIN, FUNCTION, AND POTENTIAL IN RESISTANCE BREEDING

Tzion Fahima^{1,2}, **Liubov Govta**^{1,2}, **Tamara Reveguk**^{1,2}, **Hanan Sela**¹, **Andrii Fatiukha**^{2,3}, **Valentyna Klymiuk**^{2,3}, **Yinghui Li**^{1,2,4}, **Olga Borzov**^{1,2}, **Curtis Pozniak**³, **Gitta Coaker**⁵

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Plants employ numerous innate immune receptors to perceive various immunogenic signals associated with pathogen infection and activate defense responses. These receptors include cell-surface receptor-like kinases (RLKs), and

intracellular nucleotide-binding leucine-rich repeats (NLRs). Yet, pathogens continuously develop counter-defenses in an arms race with their hosts, destabilizing food production and sustainability, and resulting in global food insecurity. Thus, breeders are seeking durable broad-spectrum resistance (BSR) that can last for many years. We previously cloned the yellow rust BSR resistance gene, *Yr15*, derived from wild emmer wheat (Nature Communications 2018) that encodes a protein with a kinase-pseudokinase domain architecture, designated as Wheat Tandem Kinase 1 (*WTK1*). The cloning of *WTK1* led us to the discovery of a novel protein family with a unique tandem kinase protein (TKP) architecture, distributed across the plant kingdom. Phylogenetic analysis indicated that TKP family members are associated with RLKs and originated from either gene duplication or gene fusion events, implying a polyphyletic origin of the TKPs by convergent molecular evolution. We hypothesize that TKPs serve as decoys that counter-defend the suppression of RLKs by pathogen effectors. Whole transcriptome analysis of wheat near-isogenic lines revealed that *WTK1* activates defense-associated transcriptional reprogramming upon pathogen infection leading to disease resistance. The decoy model can explain how TKPs regulate the activation of local programmed cell death immune responses and provide support for TKP's polyphyletic origin and evolution. Currently, eight functional wheat TKPs were described by different groups, conferring resistance against rusts (*WTK1*, *WTK2*, *WTK5*, *WTK6-vWA*), powdery mildew (*WTK3*, *WTK4*, *Pm57*), and wheat blast (*Rwt4*) diseases. *Yr15* (*WTK1*) confers resistance to >2000 pathogen isolates from around the globe, and we developed functional molecular markers for marker-assisted breeding. Thus, TKPs have a great potential for resistance breeding. Further studies are underway to elucidate the mechanism of resistance conferred by this extraordinary protein family.

SE03 C03

THE POTENTIAL OF PLANT BREEDING INNOVATION FOR IMPROVING BIOTIC STRESS RESISTANCE IN AGRICULTURAL CROPS AND VEGETABLES

Dr Petra Jorasch

Euroseeds

Reconciling sustainability with agricultural productivity in the face of climate change relies strongly on the development of resilient, high-yielding crops of superior nutritional value that can be grown more resource efficiently. Therefore, innovation in plant breeding has gained unprecedented importance. Plant breeding has strongly contributed to increased and more stable crop yields and production in arable farming, and subsequently to improved market and trade conditions, increased food availability, higher economic prosperity and additional farm income while avoiding additional land use, greenhouse gas



Concurrent Sessions

Tuesday, 2 July

(GHG) emissions, and loss of biodiversity. With this, plant breeding in the European Union drives socio-economic and environmental sustainability. New pest and diseases due to climate change require plant breeders to react quickly and to efficiently introduce new biotic resistance traits that allow for stable yields under those conditions. Plant breeding therefore needs to be very efficient to provide farmers with enabling tools (seeds) to sustainably secure their productivity. This requires the availability of a comprehensive breeding toolbox for plant breeders including the latest breeding tools like genome editing to increase efficiency in plant breeding and address breeding goals with the most suitable tools available. However, the regulatory burden for the latest breeding tools including genome editing is high in Europe. The European Court of Justice (2018) confirmed that organisms obtained by targeted mutagenesis must be considered Genetically Modified Organisms (GMOs) and with this fall under the burdensome and highly politicized EU-GMO approval system under which only one GMO event has ever been approved for cultivation back in 1998. The EU Commission published a policy proposal for plants resulting from new genomic techniques (NGTs) in July 2025. This proposal suggests differentiating plants from certain new genomic techniques from the burdensome GMO legislation. The presentation will provide an overview of NGT applications for enhancing biotic stress tolerance in crops and will elaborate on the regulatory process and its impacts on enabling such innovations for the commercial seed market. It will also analyse the EU regulatory approach for plant breeding innovation against the practical needs of the seed and breeding sector and other policy approaches around the world.

SE03 C04

DEVELOPMENT OF HIGH-THROUGHPUT GENOTYPING ASSAYS FOR GENETIC MAPPING OF RESISTANT GENES AND ACCELERATING SOYBEAN AND COMMON BEAN BREEDING FOR BIOTIC STRESSES

Qijian Song

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Soybean and common bean are among the most important crops in the world. These crops are important sources of protein for animal feed and human food, respectively. Soybean is also an important source of vegetable oil and a top export commodity for the U.S. However, biotic stresses threaten soybean and common bean production by directly reducing seed yields and/or seed quality. Although different management strategies are utilized, development and utilization of varieties that contain "resistance or tolerance" genes from traditional and wild sources have proven to be extremely effective and sustainable. High-throughput DNA molecular marker assays are required for the discovery and characterization of the genes from the germplasm and their efficient transfer to new cultivars. We developed nested high-throughput soybean molecular marker assays such as SoySNP50K, 6K, 3K and 1K containing 50k, 6k, 3k and 1k SNPs, as well as common bean assays such as BARCBearSNP6K and 12k assays consisting of 6k and 12k SNPs, respectively, to characterize hundreds of soybean and common bean germplasm and recombinant inbred line populations. We have identified genomic regions or genes

that control many traits associated with biotic stresses, performed genomic selection of lines with the resistance in both crops and developed molecular markers for marker-assisted selection. Furthermore, using the genotypic datasets, we developed soybean core collections, providing diverse sets of germplasm for efficient screening of germplasm containing biotic stress resistant genes.

SE03 C05

CRISPR/CAS9-ASSISTED GENOME ENGINEERING FOR DISEASE RESISTANCE

Maeli Melotto

Department of Plant Sciences, University of California, Davis, CA, USA

Disease resistance can be controlled by hormones, such as jasmonates (JA), salicylic acid, and ethylene, whose mode of action can be tightly regulated at the transcriptional level. Notably, the jasmonate signaling pathway is largely regulated by a conserved family of transcriptional regulators known as JAZ (jasmonate ZIM domain) proteins. Specifically, JAZ proteins are repressors of the JA pathway that is required for resistance against insects, herbivores, and necrotrophic fungi. Thus, removing this repressor can potentially enhance plant resistance to diseases. Because plants in general contain multiple JAZ genes in their genome with prominent spatial-temporal functions, it is possible to fine tune the level of resistance by precise genome editing. The CRISPR/Cas9-based genome editing approach enables highly sophisticated gene structure-function testing without the typical caveats associated with transgenic approaches. Multiple engineered systems are routinely used to create targeted gene knockouts through double-strand breaks followed by non-homologous end joining, most commonly leading to indel (insertion/deletion) mutations that disrupt transcription. We have used this technology aiming at enhancing lettuce and tomato germplasms by directly removing susceptibility genes (such as JAZ) from breeding lines and commercial cultivars in support of breeding for disease resistance programs.

SE03 C06

STRATEGIES USING GENE SILENCING AND CRISPR TOOLS IN TOMATO TO UNDERSTAND AND IMPROVE PLANT DEFENSE AGAINST VIROIDS

Hammond, Rosemarie W.

USDA ARS NEA BARC Molecular Plant Pathology Laboratory, Beltsville, Md Usa 20705

Tomatoes are one of the world's most consumed vegetable crops with production of fresh market and processing tomatoes increasing in greenhouse, high tunnels, and the field. Several viroids (small, unencapsidated, single-stranded, covalently closed circular, highly structured, noncoding RNAs) are seed transmitted and easily spread mechanically in tomato; control relies on limiting introduction of the viroid to prevent disease outbreaks as there is no known resistance to viroids in commercial cultivars. Several strategies have been employed to suppress viroid infection using various molecular tools, but these approaches have not yet been employed in the field. Disease symptoms-including stunting, reduced vigor, flower abortion, reduced size and number of

fruits, and reduced root growth-can result in significant crop losses. The dramatic alterations in plant development are the result of differential gene expression patterns, including the transcriptional activation of genes encoding transcription factors and a protein kinase involved in phospholipid signaling. Silencing of the protein kinase gene in tobacco resulted in substantially increased vigor, including increases in plant and root biomass. Our studies involved a two-pronged approach to determine if symptoms of viroid infection (tolerance) can be mitigated to reduce crop losses in tomato: 1) silencing of key genes responsible for intracellular viroid movement and symptom production using virus-induced gene silencing (VIGS), transgenic plants, and CRISPR, and 2) grafting susceptible tomato cultivars onto either tolerant, engineered, or commercially employed rootstocks to evaluate changes in gene expression, water use efficiency, plant vigor, and the effects of viroid infection on fruit development. The continued research is important for developing practical approaches to disease control.

SE03 C07

COMBATING RICE TUNGRO-COMPLEX DISEASE IN ASIA

Gilda Jonson, Israel Dave Ambita, Genelou Atienza-Grande, Il-Ryong Choi and Van Schepler-Luu

International Rice Research Institute, Los Baños, Laguna, Philippines

Rice Tungro disease (RTD) is a major virus disease that threatened rice production in tropical Asia. RTD is complex diseases caused by two distinct viruses, Rice tungro spherical virus (RTSV) and Rice tungro bacilliform virus and are transmitted by *Nephotettix virescens*. RTSV is a helper for virus transmission while RTBV is largely responsible for symptoms. Efforts have been done in mitigating the disease by developing durable disease-resistant rice by resistance-gene identification, manipulation and allele mining for crop improvement. Using diversified rice germplasm and genetic resources at IRRI, genes controlling resistance to RTSV and RTBV were identified. *EIF4G* or *tsv1* gene provides resistance against RTSV (Lee et al., 2009) while a candidate gene, *AGO2* or *tbv* provides resistance to RTBV. Using CRISPR/Cas9 gene editing system, new RTD-resistant lines were generated by editing *EIF4G* in the RTSV-susceptible variety IR64 (Macovei et al., 2018). Moreover, the 3000 rice genomes were examined for natural allelic variation of *EIF4G* that are associated with RTSV resistance, focusing on the non-synonymous SNPs within the 30-nt region of *EIF4G*. Ten percent of 3K rice accessions carry resistance *EIF4G* alleles and about 90% of those rice accessions carrying homozygous resistance *EIF4G* allele were resistant to RTSV. We identified 11 types resistance alleles of *EIF4G* and primers for high throughput PCR Allele Competitive Extension genotyping were designed to facilitate the detection and introgression of these resistance alleles into elite rice varieties. Recently, combatting Tungro became more complex as we identified the coinfection of RTD and Rice orange leaf disease (ROL) caused by Phytoplasma transmitted by the same vector of tungro. The increased incidences of RTD and ROLP coinfection in Asia requires attention on developing new rice varieties with improved resistance to both Rice Tungro disease and ROLP.

SE03 001

IN SILICO PREDICTION OF PLANT NLR- AND PATHOGEN EFFECTOR INTERACTIONS PROVIDES A TARGETED APPROACH FOR FUNCTIONAL CHARACTERIZATION

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Nucleotide binding Leucine-rich repeat (NLR) proteins play an important role in effector recognition and the activation of Effector triggered immunity (ETI) in plants following pathogen infection. The identification of NLR-effector partners is critical for elucidating functionally important NLRs in different host-pathogen interactions, as well as advancing our understanding of NLR-triggered immunity. However, deciphering which NLR proteins recognize which pathogen effectors remains experimentally cumbersome. Thus, the aim of this study was to develop a method for predicting NLR-effector interactions *in silico*, which would provide a more targeted approach for further molecular validation. NLR-effector protein structures for all experimentally validated NLRs and pathogen effectors were predicted using AlphaFold2-Multimer. Binding affinities (BA) and binding energies (BE) were then predicted using 95 structure-based machine learning models. Predicted BAs (log(K)) and BEs (kcal/mol) for NLR-effector pairs known to interact showed little variation, while BAs and BEs for false NLR-effector pairs, which do not elicit an immune response, showed large variations. Combining the results from nine models resulted in the identification of the correct binding pairs with 90% accuracy, and could thus enable the identification of true NLR-effector pairs in understudied pathosystems. The results also indicated that NLRs are activated when effector binding induces protein conformational changes resulting in a Gibbs free energy change of ~ -14.4 kcal/mol. Furthermore, contact maps identified specific NLR amino acids participating in effector binding, which could be modified to increase the binding affinity to the relevant effector. This is the first study to provide a method for predicting true NLR-effector interactions *in silico*, which could be used to narrow down NLR-effector pairs in plant-pathogen interactions. This method will streamline research efforts in deciphering which NLRs play a role in ETI activation during pathogen infection of understudied plant species, as well as shed light on amino acids important for effector recognition.



Concurrent Sessions

Tuesday, 2 July

SE03 002

IN SILICO CHARACTERISATION OF THE WAK/WAKL GENE FAMILY IN AVOCADO AND IMPLICATION IN DEFENSE AGAINST PHYTOPHTHORA CINNAMOMI

Harvey A.^{1,2}, Van den Berg N.^{1,2}, Swart V.^{1,2}

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Persea americana (avocado) faces a threat from the hemibiotrophic oomycete pathogen *Phytophthora cinnamomi*. The Wall-Associated Kinases (WAK) and WAK-Likes (WAKL) can bind pectin to influence cell expansion during development or fragmented pectin due to pathogen penetration to activate downstream pathways during defense responses. This gene family was assessed to increase understanding of the *P. cinnamomi*-avocado defense interaction. The *PaWAK/WAKL* gene family was identified in four avocado rootstock genomes - including the partially-resistant Dusa®. The *PaWAK/WAKL* expression profiles were assessed following *P. cinnamomi* infection of RO.12 (susceptible) and Dusa® rootstocks using RNA sequencing. Phylogenetic and cis-acting element analyses, with the expression profiles, were used to implicate genes in defense. In silico analyses identified differences of this gene family across rootstocks. In total, 14 *PaWAKs* and 63 *PaWAKLs* were identified and characterised in silico with five implicated in defense against *P. cinnamomi*. *PaWAK4&13* and *PaWAKL10&13&32* showed significant upregulation post-inoculation at three or four time points in Dusa® while showing either no upregulation or only at 6 hours post-inoculation in RO.12. There was a moderate relationship between *PaWAK13* and tomato's *SIWAK1* which is suggested to contribute to callose depositions during defense. There were more phytohormone-responsive cis-acting elements in the Dusa® promoter region compared to the RO.12 equivalent for *PaWAK13*. It is hypothesized that *PaWAK13* is influenced by salicylic acid and methyl jasmonate (phytohormones), with downstream pathways involved in callose depositions. The *in silico* analyses across different rootstocks showed differences in gene member compositions, coding/protein sequences, protein domains and protein structure. This study was the first to characterise the *PaWAK/WAKLs* and to implicate five in defense against *P. cinnamomi*. In silico analyses highlighted the differences across rootstocks, suggesting that *PaWAK/WAKLs* contribute to the different efficiencies of the defense responses. These *PaWAK/WAKLs* could be used as markers in molecular breeding in the future.

SE03 003

MOLECULAR BASIS OF CONSTITUTIVE DEFENSE MECHANISMS UNDERLYING RESISTANCE TO VERTICILLIUM DAHLIAE IN THE AC18 CLONE OF WILD OLIVE.

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The use of host resistance is the most effective and practical control method for the management of Verticillium wilt (VW) in olive caused by *Verticillium dahliae*, which remains one of the major threats to this crop. However, most olive cultivars of agronomic and commercial interest are susceptible to *V. dahliae*, and particularly the highly virulent D pathotype. Previous research has shown that wild olive (*Olea europaea* L. subsp. *europaea* var. *sylvestris*) germplasm harbours resistance, which could be crucial for the development of resistant rootstocks and breeding new, resistant olive cultivars. We compared transcriptomes from uninfected stems of wild olive clones AC18 and AC15, previously shown as highly resistant and susceptible to D *V. dahliae*, respectively. GO-term enrichment analysis revealed terms related to systemic acquired resistance and the plant cell wall biogenesis and assembly. Classification of the gene families and their enrichment analysis showed gene families related to biotic and abiotic stress responses. qRT-PCR analysis of selected genes showed differences in the expression of phenylpropanoid and lignin metabolism-related genes between the two wild olive clones. The phenolic content of cell walls from stem tissues was higher in the resistant AC18, and Raman spectroscopy suggested a different monolignol composition although the total lignin content was similar in the resistant and susceptible clones. Differences in gene expression profiles were found in the stems of uninfected resistant AC18, compared to susceptible AC15. The lignin composition of the stem cell wall reflects these differences. This work identifies key genes that could aid in breeding olive cultivars resistant to D. *V. dahliae*. The research findings may contribute to the conservation and utilization of wild olive genetic resources to tackle future agricultural challenges, such as climate change and emerging diseases.



MC3

08.30-11.00 Concurrent Session **4A**

Grapevine Trunk Diseases

Chair: Epaminondas Paplomatas (Agricultural University of Athens, Greece)
Laura Mugnai (Dagri, University of Florence, Italy)

SE04 C01

RECENT ADVANCES IN THE MANAGEMENT OF GRAPEVINE TRUNK DISEASES

David Gramaje

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One of the primary concerns of the grapevine nursery industry is a broad range of taxonomically unrelated pathogens associated with grapevine trunk diseases (GTDs), for which there are no effective remedies once planting material is infected. Due to the rapid expansion of wine regions, GTDs are escalating, as pathogens can be introduced during propagation, and diseases are often associated with poor planting practices and stress. Production practices in nurseries provide many opportunities for infection, through poor sanitation practices or introducing infected asymptomatic cuttings from mother vines. Control in nurseries is limited to hot water treatment and fungicide (biological or chemical) dips, with mixed results. In mature vineyards, the prevalence of GTDs has significantly increased with changes in production practices, loss of effective chemicals, predominance of susceptible cultivars and ageing of vineyards. Pruning wounds are the main infection portal for these pathogens and inoculum sources include a wide range of alternative hosts such as fruit crops and many introduced and/or native tree species. A good understanding of the etiology, biology and epidemiology of GTD fungi has led to the development of effective management strategies in the vineyard, through wound protection and remedial surgery. Successful disease management requires a holistic approach from the nursery to the vineyard. In this presentation, I will describe the available and effective strategies against GTDs at each phase of the plant's life cycle, from the nursery to the mature vineyard.

are the most common GTDs. As reported worldwide, one of the most important sources of spread of GTDs pathogens is through infected propagation material. However, knowledge about the occurrence during the production phase in Italian nurseries can be improved, and mainly the relationships nursery - vineyards must be clarified. This research aims to overview the status of GTDs affecting table grapes in Italy, with emphasis on nursery material and young vineyards. Indeed, in recent years, an increase of the incidence of GTDs has been observed. Therefore, numerous surveys were conducted on hundreds of grapevine rootstocks, grafted rootstocks, and young plants (2-4-year-old) in the main regions for table-grape production (Apulia and Sicily). Internal symptoms resembling GTDs, brown to dark discoloration developing from the rootstock base and wood necroses at the graft union were frequently observed on nursery materials while wood necrosis, reduction of root hairs and slow decline were observed on young plants. Our results showed a variable incidence of GTDs with over 90% of nursery plant material affected by different pathogens in some years. Through a molecular approach, we identified the fungal species associated with symptomatic table-grape nursery material and young vines. The main fungal were species in the families Nectriaceae, Quambalariaceae, Botryosphaeriaceae, Diatrypaceae, Diaporthaceae, Phaeomoniellaceae and Togniniaceae. Identifying fungal community, also by metagenomics approaches, is essential for improving management strategies of GTDs. The next step will focus on the effect of innovative physical and biological tools on the microbial communities associated with table-grape vines in Italy.

Funding: New Therapeutic Approaches to Reinforce the natural Grapevine microbiome against Grapevine Trunk Diseases (TARGET_GTDs), P2022ENPCL, PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) Missione 4 "Istruzione e Ricerca" - Componente C2 Investimento 1.1, "Fondo per il Programma Nazionale di Ricerca e Progetti di Rilevante Interesse Nazionale (PRIN)".

SE04 C02

GRAPEVINE TRUNK DISEASES AFFECTING TABLE GRAPE: CURRENT KNOWLEDGE AND FUTURE PERSPECTIVES IN ITALY

Dalia Aiello¹, Simone Mavica¹, Chiara Di Pietro¹, Faretra Francesco², Gerin Donato², Pollastro Stefania²

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Grapevine trunk diseases (GTDs) cause serious economic losses to table-grape vineyards in many producing countries. Black foot disease, Eutypa and Phomopsis dieback, Botryosphaeria canker, Petri disease caused by *Phaeomoniella chlamydospora* and *Phaeoacremonium* spp. and esca caused by *Fomitiporia* spp. and *Cadophora* spp.

SE04 C03

CURRENT ETIOLOGY OF ASPERGILLUS VINE CANKER AND SOUR ROT OF TABLE GRAPES IN CALIFORNIA

M.I. Bustamante¹, K. Elfar¹, M. Arreguin¹, J. Kuzmenko¹, T. Zaninovich¹, G. Zhuang², T. J. Michailides¹, A. Eskalen¹

1. University of California, Davis, 2. UCANR-UCCE

Fungal taxonomy is in constant flux and the advent of reliable DNA barcodes has allowed to improve the accuracy of identification of cryptic species. In California, *Aspergillus* Vine Canker (AVC) and Sour Rot (SR) are two diseases that affect the wood and the fruit of grapevines, respectively, and their causal agents have been previously studied using morphological characters. During the last decade, the taxonomy of *Aspergillus* section Nigri has been revised and modified. In this study, we aimed to reassess the etiology

of AVC and SR using a combination of morphological and phylogenetic analyses. Thirty-two isolates were selected based on morphological characters from 266 isolates grown on malt extract agar. Upon DNA extraction, a fragment of the calmodulin (*CaM*) gene was amplified through PCR and sequenced using the primer pair CL1/CL2A. Results revealed that isolates associated with AVC recovered from recent detections correspond to *A. tubingensis*, whereas isolates from previous studies that were initially identified as *A. niger* and *A. carbonarius* were re-identified as *A. welwitschiae* and *A. carbonarius*. The isolates from table grapes with SR corresponded to *A. tubingensis*, *A. welwitschiae*, and *A. carbonarius*. Overall, our results indicate that *A. tubingensis* was the dominant species causing both AVC and SR, and representative isolates were able to cause disease in both wood and fruits of Red Globe grapevines.

SE04 C04

STRATEGIES FOR THE DEVELOPMENT OF A NEW FORMULATION FRIENDLY FOR THE ENVIRONMENT AND PRELIMINARY RESULTS FROM GREENHOUSE TO VINEYARD

Florence Fontaine, Vincenzo Mondello, Olivier Fernandez, Patricia Trotel - Aziz

University Of Reims Champagne-Ardenne, France

Grapevine Trunk Diseases (GTDs) are a threat in viticulture and their control from the nursery to vineyard remains a real challenge. Since 2016, the Italian company NDG S.r.l. (Natural Development Group S.r.l.) is working on developing formulations based on the carrier molecule carbonate-hydroxyapatite as aqueous suspension in water (Microsap®) with low copper content (3.5%) and plant extracts (LC2017). In this sense, our previous assays with LC2017 formulation showed efficient towards some trunk diseases (GTDs) pathogens in controlled conditions by in vitro and in planta tests, and a trend to limit the esca expression in Champagne vineyard. The Natural Agro project, funded by the LIFE EU program and lead by NDG, aims to improve the assessed efficiency of Microsap®-based PPPs using new formulations derived from the LC2017, to better control long-lasting GTDs in vineyard. In addition, these formulations will be also evaluated to control Downy Mildew (DM), another relevant grapevine disease. To reach one of the main goals of the Natural Agro project, the homologation of new PPPs for the control of GTDs and DM in vineyard, several formulations were preliminary tested in vitro and in planta to evaluate their ability against pathogens of Esca and of *Botryosphaeria*- and *Eutypa*-diebacks and *Plasmopara viticola*. The selection of the potential PPPs up to two formulations, with low- or no-Cu content respectively, will be then fully tested over the duration of the Natural Agro project (2024-2028) in French, Italian, and Portuguese vineyards.

*Natural Agro is financed by the CINEA LIFE 2022 Horizon 2020 Framework under the specific programme LIFE-2022-SAP-ENV-ENVIRONEMENT (GA No 101113781).

SE04 C05

ANTIFUNGAL EFFECT OF CU BASED NANOCOMPOSITES AGAINST FUNGAL TRUNK PATHOGENS

Marie Zahrádková¹, Kateřina Štůšková¹, Jan Wohlmuth¹, Milan Špetík¹, Lucie Frejlichová¹, Zuzana Bytešníková², Tomáš Kiss³, Jakub Pečenka¹, Lukáš Richtera², David Gramaje⁴, Aleš Eichmeier¹

1. Mendeleum—Institute of Genetics, Mendel University in Brno, Valtická 334, 691 44 Lednice, Czech Republic; 2. Department of Chemistry and Biochemistry, Mendel University in Brno, Zemedelska 1, 613 00 Brno, Czech Republic; 3. Department of Fruit Science, Mendel University in Brno, Valtická 334, 691 44 Lednice, Czech Republic; 4. Instituto de Ciencias de la Vid y del Vino (ICVV), Consejo Superior de Investigaciones Científicas - Universidad de la Rioja - Gobierno de La Rioja, Ctra. LO-20 Salida 13, Finca La Grajera, 26071 Logroño, Spain

Grapevine trunk diseases, caused by various pathogens, lead to significant damage to grapevines, resulting in substantial economic losses. Currently, there is no effective treatment for these diseases. In an effort to find a solution, six batches of nanocomposites containing graphene oxide and three batches based on CuO were evaluated in vitro for their efficacy against ten fungal isolates (*Pleurostoma richardsiae* MEND-F-035, *Dactylonectria torresensis* MEND-F-0360, *Fusarium oxysporum* MEND-F-0174, *Botryosphaeria dothidea* MEND-F-0196, *Diaporthe ampelina* MEND-F-0766, *Diplodia seriata* MEND-F-0190, *Neonectria* sp. MEND-F-0788, *Eutypa lata* MEND-F-0553, *Fomitiporia mediterranea* MEND-F-0473, *Phaeoacremonium minimum* MEND-F-0013). These fungi, isolated from grapevine or stone fruit wood exhibiting symptoms of trunk diseases in the Czech Republic, represent significant threats to grapevine health. The nanocomposites, particularly those in the CuO group (CuO-L-3, CuO-L-7, CuO-L-9), demonstrated high inhibitory effects, successfully suppressing the growth of pathogenic fungal species. Among them, CuO-L-9 emerged as the most effective nanocomposite. Further investigation through in planta experiments, involving grapevines of the cv. Sauvignon grafted onto SO4 rootstocks and inoculated with the fungi, revealed nuanced effectiveness. When treated with solutions of the CuO-based nanocomposites, CuO-L-3 showed efficacy against *Pl. richardsiae* and *Da. torresensis* but not *F. oxysporum*. Conversely, CuO-L-7 and CuO-L-9 were effective solely against *F. oxysporum*. Measurements of plant and root lengths in these experiments indicated that CuO-L-3 and CuO-L-9 positively affected the height of grapevines inoculated with *Pl. richardsiae*, and all three nanocomposites benefited the root length in grapevines challenged with *F. oxysporum*. This study underscores the potential of CuO-based nanocomposites as a promising approach to managing grapevine trunk diseases.



SE04 C06

THE OOMYCETE BIOCONTROL AGENT, *PYTHIUM OLIGANDRUM*: ROOT COLONIZATION IN WORLDWIDE VINEYARDS AND CONTROL OF GRAPEVINE TRUNK DISEASES

Lisa Chaboussie^{1,2}, Amira Yacoub¹, Bikal Ghimire¹, Renaud Travadon¹, Eléonore Attard¹, Florence Fontaine², Patrice Rey¹

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Grapevine trunk diseases (GTDs) are complex diseases that represent significant threats to viticulture globally. This study explores the use of the oomycete mycoparasite, *Pythium oligandrum*, as a potential biocontrol agent against certain fungal pathogens involved in GTDs. The first objective was to assess *P. oligandrum*'s capability to colonize grapevine roots for which 10 cultivars were collected from diverse wine-growing regions in France. *Pythium oligandrum* root-colonization ability was also assessed in vineyards from Austria, Hungary, Iran, Israel, Italy, South Africa, and Spain. The study revealed that, except for South Africa, most grapevine roots were colonized by *Pythium* spp., characterized by echinulated-oospore. ITS region sequencing identified 90% of the strains as *P. oligandrum*. Furthermore, the amplification of elicitor-like genes encoding the proteins oligandrin (OLI) and Cell Wall Protein (POD1), essential for inducing plant systemic resistance, showed that the majority (80%) of these strains had both genes. Notably, 99% of the collected samples were unique for the POD1-gene, while for the OLI-gene, 86% of the samples were identical, indicating that the OLI-gene is highly conserved across different strains. The study's second objective was to assess *P. oligandrum*-selected strains' protective efficacy against GTDs pathogens under greenhouse conditions. Grapevine roots colonized by *P. oligandrum* demonstrated a reduction in wood necrosis caused by *Neofusicoccum parvum* and *Phaeoconiella chlamydospora* by 65% and 50%, respectively. To understand the mechanisms occurring during this tri-partite interaction, transcriptomic responses of grapevine were analyzed at the wood level. Transcriptomic analysis revealed an intense up-regulation of PR genes (e.g. PR1, PR2 and PR10) and genes involved in jasmonic/ethylene pathways suggesting a priming effect on the plant's defense system following *P. oligandrum* root colonization. This research underscores *P. oligandrum*'s potential as an effective biocontrol agent, offering insights into its mechanism of action and its role in enhancing grapevine resistance to fungal GTDs.

circulation of potentially contaminated planting material, the industry shift towards cultivation systems that predispose grapevines to wood infections, and the lack of effective plant protection products. Despite the significance of viticulture in Cyprus, GTDs have not been studied before. Thus, we conducted a national survey of > 180 vineyards, with GTDs incidence found high, ranging from 3% to 83%. Based on morphological and multi-locus phylogenetic analyses of the collected isolates (> 600), combined with pathogenicity trials, the local GTDs-associated mycobiome was recorded. Interestingly, besides the established GTDs pathogens, other wood-colonizing fungi were also identified in relative high numbers and found pathogenic. As a result, the novel species *Seimatosporium cyprum* (*Sporocadaceae*) was described, while *Sporocadus kurdistanicus* and *Spo. rosigena* were also reported for the first time in Europe. Furthermore, *Kalmusia variispora*, *Neofabraea kienholzii*, *Paraconiothyrium variabile*, *Macrophomina phaseolina*, and *Diaporthe foeniculina*, were also found implicated, underlying the complexity of the local pathosystem. Our GTDs management efforts have mainly focused on the exploitation of endemic, beneficial microorganisms. Out of > 490 rhizobacteria, a *Bacillus subtilis* strain (M75) provided the best protection, arresting *Phaeoconiella chlamydospora* by 87% in a potted-vine experiment. Large scale experiments are currently ongoing to evaluate the potential of synthetic plant protection products (PPPs) and biological control agents, either commercial (*Trichoderma*-based) or experimental (M75), to prevent pruning wound infections by *Botryosphaeria dothidea* and *Ph. chlamydospora*. In preventative PPPs applications, 24h and 7 days pre-wound inoculation, Tessior® and Esquive© were the most efficacious in both application time sets, while the *B. subtilis* (M75) showed very promising results, considering its non-formulated field application.

SE04 C07

THE STATUS OF GRAPEVINE TRUNK DISEASES IN CYPRUS: ETIOLOGY AND DISEASE MANAGEMENT EFFORTS

Kanetis L., Oplos C., Efstathiou S., Makris G.

Cyprus University of Technology, Cyprus

Grapevine trunk diseases (GTDs) form an aggregate of fungal diseases, considered the most destructive biotic factor of grapevines, causing reduced longevity and profitable vineyard lifespan. Although long considered a deteriorating factor, GTDs' incidence has recently spiked, mainly due to the



CONFERENCE I

08.30-11.00 Concurrent Session **5**

Vascular Wilts

Chair: Vardis Ntoulakakis (School of Life Sciences, University of Warwick, UK)
Marc Valls Matheu (Universitat de Barcelona, Spain)

SE05 C01

ANALYSING THE BACTERIAL WILT DISEASE: BACTERIAL SURVIVAL STRATEGIES AND PLANT STRUCTURAL DEFENCES

Rocafort Mercedes¹, Zhang Qingshan¹, Jiménez-Jiménez Álvaro¹, Corominas Clàudia^{1,2}, Coll Núria S.¹, Valls Marc^{1,2}

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Ralstonia solanacearum, the causing agent of the bacterial wilt disease, is a devastating pathogen spreading worldwide. We present our studies on the adaptation of the pathogen to all environments occupied throughout its life cycle. *R. solanacearum* deploys distinct transcriptional programmes at different infection stages and in environmental niches outside of the plant. We found that the type III secretion system (T3SS) is induced throughout infection, especially at late stages and identified alkaline pH and nutrient scarcity as inducers. On the contrary, oxidative stress responses and genes for the use of alternate carbon sources, such as phenylacetate catabolism, were specifically upregulated when *R. solanacearum* lives in the soil. We will also present our spatio-temporal characterisation of the *R. solanacearum* colonization dynamics in susceptible and resistant tomato varieties. Our work revealed that structural constraints are key for resistance to bacterial wilt both in root and shoot tissues. We have investigated the physico-chemical nature of the induced plant barriers as ligno-suberin coatings and tyramine-derived hydroxycinnamic acid amines. In agreement with these findings, overexpression of the ligno-suberin pathway in a susceptible tomato enhanced resistance by restricting *R. solanacearum* movement inside the plant and delaying disease progression. Our findings represent valuable insights into the biology of *R. solanacearum* and its adaptation to unexplored habitats and open new avenues to engineer resistance against vascular wilt pathogens.

is a relatively new field. The biocontrol agent *Bacillus velezensis* K165 (formerly *Paenibacillus alvei* K165) has previously demonstrated its ability to protect *Arabidopsis thaliana* plants against *Verticillium dahliae*. In this study, we reveal that K165 also imparts inherited immune resistance to *V. dahliae*. Through a combination of histone acetyltransferases mutant screening, chromatin immunoprecipitation (ChIP) assays, and transcriptomic experiments, we demonstrate that histone acetylation significantly contributes to K165's biocontrol activity and the establishment of inheritable resistance to *V. dahliae*. Treatment with K165 primes the expression of immune-related marker genes and the cinnamyl alcohol dehydrogenase gene *CAD3* via histone acetyltransferases. Our findings suggest that offspring of plants treated with K165 exhibit primed immunity and enhanced lignification, both of which contribute to K165-mediated inherited immune resistance. This study highlights the potential of biocontrol agents in establishing inheritable resistance to agriculturally significant pathogens, offering new insights into the molecular mechanisms underlying plant-microbe interactions and the potential application of epigenetic regulation in disease management strategies.

Research work in the Phytopathology Lab (Agricultural University of Athens) was supported by the Hellenic Foundation for Research and Innovation (H.F.R.I.) under the "1st Call for H.F.R.I. Research Projects to support Faculty Members & Researchers and the Procurement of High-Cost Research Equipment Grant" (project number: 125).

SE05 C03

SNC1 GUARDS TOPLESS FAMILY MEMBERS THAT ENCODE SUSCEPTIBILITY GENES FOR FUSARIUM WILT IN TOMATO AND ARABIDOPSIS

Frank Takken, Thomas Aalders, Fleur Gawehns, Hanna Richter, Harrold. A. van de Burg

University Of Amsterdam, Netherlands

Fusarium oxysporum causes Fusarium wilt disease many agricultural important crops. The fungus manipulates its hosts using SIX effectors. We reported that the broadly conserved effector SIX8 targets specific members of the Topless (TPL) family (Aalders *et al.*, 2024 PBJ; 22, 248). TPLs are transcriptional co-repressors and SIX8-targeted tomato TPL1 and TPL2 proteins are important for susceptibility to the fungus. Tomato *tpl2* mutants are not affected in susceptibility, but a *tpl1* mutant developed less disease symptoms upon inoculation. Combining both *tpl* mutants resulted in near complete Fusarium resistance. Likewise, *Arabidopsis thaliana* *tpl1* and *tpl2* mutants showed mildly reduced susceptibility to *F. oxysporum*, while near complete resistance was observed in the double mutants. Hence, TPLs are genuine S genes whose inactivation can confer *F. oxysporum* resistance. To study SIX8 function the effector was expressed in *Arabidopsis Col-0*. The few transformants recovered exhibited a temperature-

SE05 C02

EPIGENETICS IN BIOLOGICAL CONTROL OF VASCULAR WILTS: THE CASE OF BACILLUS VELEZENSIS K165 AGAINST VERTICILLIUM DAHLIAE

Danai Gkizi², Anna González Gil³, Alonso J. Pardal³, Sophie J.M. Piquerez³, Chrysi Sergaki³, Eirini G. Poulaki¹, Vardis Ntoulakakis³, Sotiris Tjamos¹

1. Phytopathology lab, Agricultural University of Athens, Greece, 2. School of Food Sciences University of West Attica, Greece, 3. School of Life Sciences University of Warwick, England

While the role of microbes in plant disease biocontrol and the molecular interactions between plant-biocontrol agents (BCAs) and pathogens have been extensively studied in recent decades, research into the epigenetic effects of BCAs on plants



dependent growth phenotype, constitutive *PR1* and *PR2* expression, and local cell death, all indicative for constitutive immune signalling. *TPR1* interacts with *SNC1*, a temperature sensitive NLR-type immune receptor (Zhu *et al.* 2010 *PNAS*; 107, 13960-). To test involvement of *TPL*, *TPR1* and of *SNC1* and *SNC1*-signalling components (i.e. *eds1*, *pad4*, and *NahG*) *Arabidopsis* mutants were transformed with *SIX8*. Knockout mutants in *TPL*, *TPR1*, *SNC1* and *EDS1* were found to fully revert the *SIX8* phenotype, while the phenotype was partially reverted in *NahG* plants. These data indicate that *SNC1* guards *TPL* and *TPR1* and monitors their perturbation by *SIX8*, implying that *TPLs* act upstream of *SNC1*. Evolution of a guard to monitor *Topless* proteins provides additional support for an essential role of these proteins in disease resistance and susceptibility. A possible mechanism of how *Six8* triggers *SNC1*-mediated immune signalling and manipulates *TPLs* will be presented, providing new leads to reduce susceptibility to *F. oxysporum*.

SE05 C04

THE ROLE OF MYST HISTONE ACETYLTRANSFERASES IN PLANT IMMUNITY

Alexia Tornesaki, Anna González Gil, Ampntelnour Litsa and Vardis Ntoukakis

School of Life Sciences, University of Warwick, Coventry CV4 7AL, United Kingdom

Part of the immune responses to pathogens is reprogramming of gene expression. Approximately 10 per cent of the plant genome have been shown to have significantly altered expression levels after pathogen perception. One major mechanism controlling the partial expression of the genome is acetylation of the N-terminal tails of histones by histone acetyltransferases (HATs). Here we investigate the role of the *Arabidopsis thaliana* MYST-HATs, *AtHAG4* and *AtHAG5*, in immunity responses. Our results show that the two MYST-HATs interact with distinct set of transcription factors and have different functions. *AtHAG4* regulates immunity against the root pathogens *Verticillium dahliae* and *Fusarium oxysporum* while *AtHAG5* regulates immunity against the leaf pathogen *Pseudomonas syringae* showing that there is a tissue specificity in defence responses. Our data also highlight the role of histone acetylation in plant immunity against vascular pathogens and how chemical inhibitors of histone acetyltransferases can be used to translate our results to crop plants.

SE05 001

BIOCONTROL AGENTS MITIGATE MAIZE LATE WILT: ENHANCING CROP RESILIENCE FOR FOOD SECURITY

Matos D.^{1,2}, Cardoso P.^{1,2}, Figueira E.^{1,2}

1. University Of Aveiro, Aveiro, Portugal, 2. CESAM, Aveiro, Portugal

Magnaportheopsis maydis causes late wilt in maize plants, yield losses reported may reach 70% in sensitive maize hybrids. Thus, late wilt is a serious disease compromising the necessary increase in maize grain the production to meet cereals demand and ensure food security by 2050. The fungus infects the roots at an early stage of plant development, but wilt symptoms appear at flowering stage. Then the lower stem

becomes dry and has a hollow and shrunken appearance. There is no effective method to control the disease, being resistant cultivars the currently used practice, but which is offset by the development of increasingly virulent pathogenic variations of the fungus and frequently have lower economic and nutritional values. Thus, there is urgency in new and effective approaches to overcome the disease. Bacteria with antifungal properties are emerging as sustainable alternatives to mitigate the effects of late wilt. In this study, bacteria isolated from the roots of symptomatic plants were tested for their antifungal ability. Isolates were chosen for *in planta* experiments and physiological, biochemical, and nutritional status of infected and control plants were assessed. Two bacterial isolates were able to reverse the lower growth of infected plants. However, with different strategies, one having little impact on the plant physiological and biochemical status, and the other promoting the antioxidant response and increasing proline and phenolic compounds, thus inducing the plant response to infection. Alterations in nutritional status of treated plants compared to infected ones can evidence the effects of fungal inhibition in an early stage of the disease. The results obtained may support the inoculation of bacteria with antifungal activity as a viable and sustainable alternative in the control of phytopathogenic fungi.

SE05 002

THE ROLE OF ATP-DEPENDENT CHROMATIN REMODELLING COMPLEXES IN REGULATION OF PLANT IMMUNITY AGAINST FUSARIUM OXYSPORUM

Ampntelnour L.¹, Clarkson J.², Ntoukakis V.¹

1. School of Life Sciences, University of Warwick, Coventry, UK, 2. Warwick Crop Centre, School of Life Sciences, University of Warwick, Wellesbourne, UK

Fusarium wilt is one of the most destructive and economically important fungal diseases, severely affecting the quality and yield of crops worldwide. We have previously shown the involvement of ATP-dependent chromatin remodelling complexes (CHRs), of the Swi2/Snf2 protein family in *Arabidopsis* leaf immunity against bacterial pathogens. However, their role in regulating immunity against soil-borne pathogens remain unexplored. Using a reverse genetic screen, we identify *Atchr8*, *Atchr9* and *Atchr25* (yeast *RAD54* homologue) as negative regulators of *Arabidopsis* root immunity against *Fusarium oxysporum* f. sp. *conglutinans* (*Foc*). This CHR family was previously reported to be involved in facilitating the effectiveness of the DNA-damage repair (DDR) response. Currently, we investigate the involvement of these CHRs in maintaining genome integrity following double-strand breaks (DSBs) induced by *Foc* infection using genome wide transcriptomic approaches.



SE05 003

DISSECTING INTO THE EFFECT OF STARCH METABOLISM ON ARABIDOPSIS THALIANA DEFENSE AGAINST THE VASCULAR WILT PATHOGEN FUSARIUM OXYSPORUM**Kalogeropoulou E.¹, Tjamos S.², Aliferis K.³, Beris D.⁴, Vloutoglou I.¹, Papiolmatas E.²**

1. Laboratory of Mycology, Scientific Directorate of Phytopathology, Benaki Phytopathological Institute, Kifissia, Greece, 2. Laboratory of Plant Pathology, Agricultural University of Athens, Athens, Greece, 3. Laboratory of Pesticide Science, Agricultural University of Athens, Athens, Greece, 4. Laboratory of Virology, Scientific Directorate of Phytopathology, Benaki Phytopathological Institute, Kifissia, Greece

Fusarium oxysporum species complex (FOSC) causes destructive vascular wilt diseases in a broad range of hosts resulting in severe yield losses. Various strategies are currently employed to manage *Fusarium* wilts in economically important crops, with host resistance being the most effective, sustainable and environmentally friendly component of an IPM system. *BAM3* gene is the dominant contributor to starch degradation and synthesis of maltose, which is involved in sucrose and cellular metabolism. *In planta* assays showed that *Arabidopsis thaliana bam3* plants had significantly lower susceptibility to *F. oxysporum* f. sp. *raphani* (*For*) infection compared to the wild-type plants. The results of the bioassays were confirmed by the quantification of the fungal biomass in plant tissues using real-time PCR. Transcriptomic analyses using DNA microarrays and reverse-transcription quantitative PCR, and metabolomic analyses using gas chromatography-mass spectrometry revealed that regulation of starch metabolism after *For* inoculation leads to the regulation of sugar fluxes that, in turn, induces defense responses against *For* in *A. thaliana bam3* mutant. Additionally, comparative studies of the expression of cell wall-related genes, the polysaccharide composition of cell wall and the immunolocalization of cell wall proteins showed that the cell wall remodeling regulated resistance against the pathogen.



BANQUET

11.30-14.00 Concurrent Session 6A

Advances with Biopesticides to meet Plant Protection Challenges

Chair: Trevor Jackson (*AgResearch, New Zealand*)

Laura Villamizar (*Microbial Solutions, AgResearch Ltd., New Zealand*)

SE06 C01

OVERCOMING CONSTRAINTS AND MEETING THE POTENTIAL OF MICROBES FOR PLANT PROTECTION: TECHNOLOGY AND SUSTAINABILITY

Laura Villamizar, Trevor Jackson

AgResearch, Lincoln Research Centre, New Zealand.

Fungi can interact with plants in beneficial or detrimental ways and play a major role in agriculture sustainability. Fungi can be endophytes, antagonists of plant pathogens, entomopathogens, plant-growth-promoting agents, sequesters of carbon in the soil and remediators of contaminated soils among other roles. All of these properties have encouraged industry to produce fungi as commercial products such as biopesticides and biofertilizers which can replace harmful synthetic chemicals. Commercial bioproducts based on beneficial fungi have been successfully developed, commercialised and used in a number of countries, where technology development has been supported by special efforts in (i) selection of biocontrol agent (ii) formulation (iii) manufacturing and registration (iv) application methods. However, commercial bioproducts can be expensive and not readily available in the global market and commonly have problems of quality and shelf-life. For these reasons, and under specific conditions and quality standards, local, artisanal, or on-farm production of beneficial fungi has been successfully implemented in developing and emerging countries such as Colombia, Brazil and Kenya. However, to achieve sustainable and quality local production, intense training and support programs as well as rigorous protocols and practical tools are necessary to ensure standards of purity, viability, concentration, and efficacy. We are currently applying this approach to assist Papua New Guinea, Solomon Islands and Samoa in the control of the coconut rhinoceros beetle (*Oryctes rhinoceros*), an invasive pest threatening coconut and oil palm production. Production systems for the entomopathogenic fungus *Metarhizium majus* are under development and implementation in these countries, through joint work with government institutions and industry to control the pest. In this presentation, we will show the progress in this work together with other successful examples of implementing local/artisanal production of fungi or commercial biopesticides in other countries and the challenges and opportunities involved will be discussed.

SE06 C02

LEVERAGING THE POWER OF TWO BACTERIAL SYMBIONTS IN PLANT PROTECTION: PHOTORABDUS AND XENORHABDUS

Selcuk Hazir

Aydin Adnan Menderes University, Faculty of Science, Department of Biology, Aydin, TURKIYE

Xenorhabdus and *Photorhabdus* are insect pathogenic bacteria mutualistically associated with entomopathogenic nematodes *Steinernema* and *Heterorhabditis*, respectively. These bacterial species release several enzymes and secondary metabolites into insect hemocoel to kill insect host and to protect infected cadavers against competitors. Some of these natural products (NPs) are non-ribosomal derived peptides, polyketides, and/or hybrids known to have different biological properties such as antibacterial, antifungal, insecticidal, acaricidal activities and have direct effect on plant protection. Over the years a newly developed biotechnological approach called the easyPACId has been successfully used to determine the bioactive compounds in *Xenorhabdus* and *Photorhabdus* spp. Among these NPs, fabclavine, transcinamic acid, cabanillasin, stilbene derivatives, benzaldehyde and xenocoumacine reportedly exhibit strong antifungal activities against various plant pathogenic fungi. Benzylideneacetone synthesized by *X. nematophila* is effective against some plant pathogenic bacteria like *Erwinia amylovora* and *Phytophthora nicotianae*. NPs with insecticidal activities against herbivorous insects include TcdA, PirAB and Mcf toxins and xenocoumacine from *X. nematophila* are bioactive compounds that kill *Tetranychus urticae*. In addition to these activities, bacterial NPs such as 3,5-dihydroxy-4-isopropylstilbene, indole, fabclavines, rhabdopeptides, and xenocoumacins are highly toxic to plant parasitic nematodes. Such natural products can be used as novel biopesticides or their chemical structures can be utilized as templates in the design and synthesis of new alternatives that can replace current more toxic chemicals.

SE06 C03

ADVANCES AND CHALLENGES IN BIOPESTICIDE PRODUCTION - A PRODUCER PERSPECTIVE

Peters Arnes

e-nema GmbH, Schwentinental, Germany

Founded in 1997, E-nema has experienced the accelerated growth in the biopesticide business as a producer of its own nematode based biopesticides and as a toll-manufacturer for companies around the world. Liquid fermentation is employed for the majority of biopesticides. Bacteria, bacteriophages, fungi, nematodes or protozoans are produced in liquid culture. The active ingredient resulting from the fermentation can



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either be a living propagule which is expected to act and grow on the pest and hence bears the advantage of propagation and auto-dissemination, or a blend of metabolites produced during fermentation affecting the pest or triggering plant defense mechanisms. The production and formulation of living propagules is usually more challenging, since they may be susceptible to desiccation or heat and hence can only be dried under mild conditions. The most robust propagules are spores of *Bacillus* spp. and biopesticides based on *Bacillus* comprise the oldest as well as the most widespread biopesticides today. The focus on *Bacillus*, however, bears the threat of missing the huge potential in microbial diversity for biocontrol. Current legislation, especially the allowable levels for contaminating bacteria or fungi were copied from stringent food regulation without reflecting the difference in human exposure between food and pesticides. They are easy to satisfy when producing robust *Bacillus* spores but impossible to meet when a gentle drying process is required for e.g. yeast like fungi or gram-negative bacteria. For baculoviruses, manufacturers have successfully argued that meeting the strict food-guidelines is impossible. Subsequently, the limit was raised by a factor of 1000 illustrating how arbitrary this limit was settled. Similarly, innovative products based on gram-negative bacteria or fungi will require tailored specification limits for contaminating organisms. It is argued, that industrial production possibilities should be considered early in the development of a novel biological plant protection product.

SE06 C04

MULTIFUNCTIONALITY IN THE ENTOMOPATHOGENIC ENDOPHYTIC FUNGI METARHIZIUM SPP

Federico Rivas-Franco

National Institute for Agricultural Research (INIA), Las Brujas, Uruguay

Entomopathogenic fungi, characterized by their ability to infect and kill insects, play a crucial role in the biological control of insect pests. The genus *Metarhizium* stands out for its unique features. *Metarhizium* demonstrates environmental friendliness and ease of mass production of conidia, blastospores, or microsclerotia. Species belonging to *Metarhizium* naturally infect insects through the cuticle without requiring ingestion and, unlike chemically synthesized pesticides, it does not induce resistance in pests. The ecological impact and potential of *Metarhizium* as a bioinsecticide have been further amplified by recent discoveries of its ability to colonize plant roots, even sometimes as an endophyte. This association imparts various benefits to plants, including growth promotion, acquisition of insect-derived nitrogen, antagonism against phytopathogens, induction of plant resistance, and enhanced tolerance to drought and salt stress. These distinctive properties in *Metarhizium* are linked to the evolution of the different species within the genus, suggesting an origin from fungal plant endophytes and phytopathogens. Consequently, *Metarhizium* spp. harbor a useful set of enzymes and diverse secondary metabolites with activities against not only insects but also fungi, bacteria, viruses, and even cancer cells. Microbiome studies involving soil amendment with *Metarhizium robertsii* reveal its contribution to generate disease-suppressive soils, accompanied by an increased abundance of beneficial bacterial and fungal communities in plants. In summary, the

multifunctional properties of *Metarhizium* make it highly promising for development as a versatile agricultural bio-input. Its biofertilizing and bioprotective capabilities against insect pests and phytopathogens position *Metarhizium* as a valuable asset in sustainable agriculture.

SE06 001

SPRAY-INDUCED GENE SILENCING IN MANAGING SOYBEAN FUNGAL DISEASES

Chen Z.

Louisiana State University Agricultural Center, Baton Rouge, United States

Soybean (*Glycine max* L.) is one of the most important agricultural crops worldwide as well as in the US and Louisiana. The total value of soybean production in Louisiana is over \$773.4 million in 2022. However, soybeans are susceptible to attack by various fungal pathogens throughout their growing season, such as *Fusarium virguliforme*, *Macrophomina phaseolina*, and *Sclerotinia sclerotiorum*. Soybeans grown in the southern U.S. suffer from additional diseases, such as Asian soybean rust caused by *Phakopsora pachyrhizi*, and *Cercospora* leaf blight and purple seed stains caused by *Cercospora* cf. *flagellaris*, and frog-eye leaf spot caused by *C. sojina*. The traditional fungicide-based approach to manage these diseases becomes less effective as pathogens develop resistance in their populations. The recently developed RNA interference-based spray-induced gene silencing that involves foliar application of double-stranded RNAs (dsRNA) showed a great potential as a viable alternative to the fungicide approach in managing plant diseases. This approach utilizes the toxic information carried in the dsRNA to specifically suppress the expression of its target genes of the pathogens to achieve the disease control. It is environmentally safe and sustainable. The objectives of this study are to clone multiple genes from these pathogens for dsRNA production in a bacterial system, apply these isolated dsRNAs to soybean plants before inoculating with pathogens to identify the most effective dsRNA in reducing fungal infection or disease symptoms. We have produced over 20 different dsRNAs to target various vital genes from these pathogens. Several of them have demonstrated high efficacy in suppressing fungal infection and disease symptoms in our repeated growth chamber and greenhouse trials. We are currently exploring different ways to enhance the dsRNA uptake and extend their effectiveness. The long term goal is to develop a practical dsRNA-based approach to manage various soybean fungal diseases through foliar applications or seed treatments.



SE06 002

CONTROLLING OLIVE ANTHRACNOSE WITH ANTIFUNGAL METABOLITES FROM BACILLUS SPECIES: A BIOLOGICAL APPROACH

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Anthrachnose disease in olive, caused by the fungal pathogen *Colletotrichum acutatum*, is considered one of the most critical issues in olive orchards in Pakistan. This disease poses a significant threat as it results in infections that can lead to the complete damage of olive plants, affecting leaves, stems, and fruits in the field. Controlling this disease is particularly challenging due to the absence of an effective fungicide that does not pose risks to farmer health and the environment. To address this challenge, our study aimed to evaluate the antagonistic activity of a biosurfactant produced by the *Bacillus subtilis* PE-07 strain against the anthracnose-causing agent in olive plants. This strain was selected after screening of forty gram Positive rhizobacteria strains. Additionally, we assessed the heat stability, pH range, and toxicity of the biosurfactant produced by strain PE-07. Our results revealed that the biosurfactant exhibited maximum antifungal activity against *C. acutatum*. In vitro studies indicated that the biosurfactant could reduce fungal activity by inhibiting the spore germination of *C. acutatum*. Furthermore, the biosurfactant demonstrated a wide pH and temperature range, displaying antifungal activity at pH levels ranging from 5 to 10 and a temperature range from room temperature to 110°C. To evaluate the biosurfactant's safety, we conducted toxicity tests on zebra fish (*Danio rerio*). The results showed that the biosurfactant had minimal harmful effects, even at maximum concentrations. In conclusion, our study confirmed that the biosurfactant produced by *B. subtilis* exhibited high pH and heat stability with minimal harmful effects. Therefore, it presents a promising alternative to chemical pesticides for effectively controlling olive anthracnose in Pakistan.

SE06 003

NEW PROMISING "BIOFUNGICIDES" TARGETING THE FUNGAL CELL WALL TO CONTROL THE CUCURBIT POWDERY MILDEW PODOSPHAERA XANTHII

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One of the main limitations of cucurbit production is the powdery mildew disease, caused by *Podosphaera xanthii*. Although different management strategies are used to control it, the application of fungicides is the most effective. However, there are two major problems: the rapid emergence of resistance to fungicides by the pathogen and the strong restrictions on the use and diversity of phytosanitary products imposed at the European level. For this reason, novel targets and strategies are needed to develop new "biofungicides" for sustainable disease control. The fungal cell wall (CW) is a unique and essential structure, not present in human or animal cells, which makes it an ideal target for the development of new phytosanitary products. In this study, three *P. xanthii* cell

wall-related genes (coding a GPI-anchored membrane protein and two proteins involved in the synthesis of rhamnose) were studied. To understand their impact on the development of *P. xanthii*, RNA interference (RNAi) technology was used. For this purpose, dsRNA molecules were synthesized and several assays were performed. The infiltration and leaf disc assays on melon and zucchini cotyledons, respectively, showed an effective gene silencing effect, resulting in a significant reduction of gene expression, fungal biomass and area covered by powdery mildew disease. Microscopy analysis confirmed the impact on fungal penetration points. In addition, the application of dsRNAs, through spray-induced gene silencing (SIGS) assays, resulted in a significant decrease in disease development on melon plants. According to the results obtained, these genes could serve as potential targets for the development of new generations of antifungal agents. In addition, SIGS technology could be a promising strategy to incorporate into integrated management programs for this important disease.

This work is part of the R+D+i projects PID2019-107464RB-C21 and PID2022-1362400B-C21, funded by MCIN/AEI/10.13039/501100011033/FEDER. UE. I.P.-R., was supported by a Ph.D. fellowship from AEI, grant number PRE2020-093156.

SE06 004

INSECT PARASITIDS OFFER A NATURAL SOURCE OF NOVEL BIO-INSECTICIDES FOR SUSTAINABLE PLANT PROTECTION IN DIVERSE ECOLOGIES

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Ensuring sustainable and cost-effective food production is an urgent priority for agriculture. While synthetic insecticides are widely used for insect control, they are known to have detrimental effects on the environment and non-target organisms. Heavy reliance on these chemicals poses significant environmental and health risks. One promising alternative is leveraging biotechnology-based natural products or organisms for pest suppression. The diversity of indigenous insect parasitoids presents a promising avenue for discovering new genes and peptides that could serve as alternatives to synthetic insecticides in agriculture. This is particularly important given the escalating issue of insect pests in agriculture, alongside projections of a global population increase to 10 billion in the next four decades. Female parasitoid wasps inject various substances, including venom, ovarian fluid, and polydnavirus, which contribute to the mortality of harmful insects. By analyzing the venom proteins of the wasps species *Aeansius arizonensis* and *Bracon hebetor*, identified potential bioactive compounds through techniques such as SDS-PAGE and protease/trypsin digestion. The genes encoding these proteins were isolated using RT-PCR, and recombinant proteins were produced. The insecticidal activity of these proteins was then assessed on both target and non-target organisms. This foundational research has provided valuable insights that could be leveraged to develop novel insect control strategies, ultimately safeguarding the biodiversity of agricultural crops.

SE06 005

LIST OF SUCCESS OF OLIGONUCLEOTIDE INSECTICIDES: HEMIPTERAN PESTS UNDER CONTROL OF THE UNMODIFIED ANTISENSE DNA

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The idea of CUAD (contact unmodified antisense DNA) biotechnology and the use of oligonucleotide insecticides (briefly, olinscides, or DNA insecticides) was proposed in 2008. As the next-generation platform, it has a number of unique characteristics (short unmodified antisense DNA as contact insecticide, insect rRNAs as target, DNA containment as mechanism of action) that distinguish it from existing classes of chemical insecticides. The simplicity, flexibility, and effectiveness of CUAD biotechnology for hemipteran pests (aphids, psyllids, soft scales, armored scales, mealybugs, etc.) is amazing (Table 1). The use of species-specific conservative sequences of target rRNAs of insect pests will minimize the occurrence of target-site resistance. Certain combinations of nitrogenous bases in an oligonucleotide insecticide will make it well-tailored to a distinct insect pest. The use of unmodified DNA as a natural polymer will minimize the toxicity load on ecosystems where oligonucleotide insecticides will be used. Liquid-phase synthesis of DNA makes CUAD biotechnology very cheap already today (0.5 USD/hectare for aphids). Oligonucleotide insecticides are solving many problems of the 21st century and agricultural countries that implement these innovations will prosper.

Table 1. List of success of oligonucleotide insecticides on hemipteran pests

Species	Target	Sequence of olinscide	Concentration, ng/μL	Mortality (%), day
<i>Unaspis euonymi</i>	28S rRNA	5'-ATACCGACGAT-3'	100	99.2, 10 th
<i>Aonidia lauri</i>	28S rRNA	5'-ATGCCAACGAT-3'	100	98.2, 14 th
<i>Macrosiphoniella sanborni</i>	ITS2	5'-TGTGTTTCGTTA-3'	100	97.4, 7 th
<i>Trioza alacris</i>	ITS2	5'-GACACGCGCGC-3'	100	97.2, 14 th
<i>Coccus hesperidum</i>	28S rRNA	5'-CGACCGACGAA-3'	100	95.6, 12 th
<i>Cinara pinea</i>	ITS2	5'-TGTGTTTCGTTA-3'	100	91.1, 4 th
<i>Diaspis echinocacti</i>	28S rRNA	5'-ATCGCTGCGGA-3'	100	84.2, 14 th
<i>Dynaspidiotus britannicus</i>	28S rRNA	5'-ATACCGACGAT3'	100	82.4, 10 th
<i>Ceroplastes japonicus</i>	28S rRNA	5'-CGACCGACGAA-3'	100	78.8, 10 th
<i>Pseudococcus viburni</i>	18S rRNA	5'-GGAATCGAACC-3'	100	78.3, 3 rd
<i>Icerya purchasi</i>	28S rRNA	5'-ACACCGACGAC-3'	100	70.6, 10 th

SE06 006

EVALUATION OF ENTOMOPATHOGENIC NEMATODES VIABILITY AS AFFECTED BY SPRAY APPLICATION TECHNIQUES AND OPERATIONAL PARAMETERS

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Nowadays, one of the main challenges to plant protection is to achieve a successful pest management with a concurrent significant reduction of chemical pesticides. In this framework, a pivotal role can be played by biocontrol agents (BCA)-based formulations; however, the control efficiency of BCA strictly relies on the operating conditions of spray applications. Indeed, during pesticide treatments, BCA are subjected to physical and thermal stresses mainly due to the i) liquid pump recirculation, ii) passes through filter meshes and iii) pressures in the sprayer hydraulic circuit, with a possible reduction of their viability and therefore effectiveness. In this work, we assessed the effect of different components involved in spray application on commercially formulated entomopathogenic nematodes (EPN). First, a protocol to determine EPN viability was set up using three among the most used species in bionsecticides, namely *Heterorhabditis bacteriophora*, *Steinernema carpocapsae*, and *S. feltiae*. Then, two types of pesticide application equipment were tested using *H. bacteriophora*, which is commonly used for biocontrol of a major agricultural pest in northwestern Italy, i.e. the grubs of *Popillia japonica*. A preliminary trials using a conventional small-boom sprayer and an uncrewed aerial spray system (UASS) were carried out simulating spray application in static condition; the EPN viability was assessed on spray mixture sampled at different times. The results showed a progressive decrease of EPN viability from the first to the last sampling timing, with the highest drop of live nematodes for the UASS. Therefore, the conventional sprayer was selected for further tests of the effect of separate equipment components and spraying conditions on EPN viability and efficacy. The combination of pressure, temperature and time affected the percentage of live nematodes, suggesting that working parameters deeply influence the effectiveness of EPN spray application.

Work is part of the project NODES which has received funding from the MUR – M4C2 1.5 of PNRR funded by the European Union - NextGenerationEU (Grant agreement no. ECS00000036)



SE06 007

METHYLOBACTERIUM-INDUCED PLANT BLEACHING: ANALYZING MOLECULAR MECHANISMS TOWARDS THE ESTABLISHMENT OF R&D BASIS FOR MICROBIAL HERBICIDES

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Microbial herbicides offer a promising approach by reducing chemical herbicide use, managing herbicide resistance and being applicable in organic agriculture. However, conventional options face challenges due to their weak and slow-acting effects, necessitating both theoretical and technological studies. This study focuses on *Methylobacterium indicum* strain VL1 as a candidate for a microbial herbicide, owing to its potent and fast-acting activity attributed to its plant-bleaching capacity. It aims to elucidate the pathogenic genes, factors, and mode of action to understand the comprehensive molecular mechanisms involved. Hypothesis regarding the mode of action and chemical properties of the pathogenic factors were formed based on observation of symptoms. VL1 was inoculated into rice under continuous light/dark conditions to evaluate chlorophyll and carotenoid contents. The solvent extraction method and untargeted-metabolome analysis with LC-MS were conducted to estimate the chemical structure of the pathogenic factors. *In silico* analysis with antiSMASH and gene knockout with transposon mutagenesis and homologous recombination were conducted to investigate the pathogenic genes. Based on symptom observation, the hypothesis that water-soluble factors inhibit carotenoid biosynthesis was formulated. Inoculation tests demonstrated a simultaneous decline in chlorophylls and carotenoids under continuous light condition, and solely reduction in carotenoids under continuous dark conditions. Using the solvent extraction method, pathogenic factors were partitioned into the aqueous layer. The untargeted-metabolome analysis detected several specific peaks in VL1, deduced as peptides. Genome analysis revealed the presence of approximately 10 secondary metabolite biosynthetic gene clusters, including peptides. Disruption of the *luxR* gene resulted in the loss of pathogenicity. Based on the above, the *luxR* gene and water-soluble factors may be involved in pathogenicity via carotenoid biosynthesis. Future investigations, including transcriptome analysis, are crucial to uncover the comprehensive molecular mechanisms involved.

SE06 008

EVALUATION OF RHIZOBACTERIA AND BIOGENIC SILVER NANOPARTICLES FOR THE MANAGEMENT OF PSEUDOMONAS SRINGAE PV. SYRINGAE CAUSING BACTERIAL CANKER ON PLUM

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The current study was focus on the management of bacterial canker disease on plum using rhizobacteria and silver nanoparticles. To achieve the objectives of this study, the collection of one-year-old seedlings of plum cultivar was done from certified nurseries in Swat and Peshawar. Isolation and purification of rhizobacteria was done from collected rhizospheric soil of healthy plum orchards. The rhizobacterial properties viz. siderophore production, hydrogen cyanide production (HCN), P-solubilization and biofilm formation of isolated rhizobacterial isolates were evaluated. Selected rhizobacterial isolates were identification using PCR technique using 16sRNA primer followed by phylogenetic analysis. Biogenic silver nanoparticles (AgNPs) were prepared using 0.2%, 0.4%, and 0.6% concentrations of *Cannabis sativa* plant extract. *In-vitro* antibacterial activity was performed by the disk diffusion method by culturing the stock cultures of the bacteria in broth by inoculation and grown for 18 hrs at 28±2 °C temperature. Biogenic silver nanoparticles and rhizobacterial isolates were then evaluated on already established plum nursery against *P. syringae* pathovars. Results showed that the during inhibition zone technique in laboratory assay the maximum inhibition zone (24.32 mm) was recorded for the concentration 0.6% as compared with control. While in greenhouse experiment the minimum disease incidence was recorded in the treatment having consortia of two bacillus strains with 0.6% concentration of green AgNPs while in a consortia of *Bacillus* and *Pseudomonas* strains with 0.6% AgNPs the disease incidence was enhanced rather than decreased may be due to the non-compatibility of *Bacillus* and *Pseudomonas* strains. The results of the current study should be helpful in designing control strategies against bacterial diseases on plum. The usage of relatively resistant cultivars, effective bio-control agents and silver bio-nanoparticles should provide alternatives to synthetic pesticides.



Concurrent Sessions

Tuesday, 2 July

SKALKOTAS

11.30-14.00 Concurrent Session **7A**

Understanding Pathogen-Vector-Host Interactions in Globally Important Pathosystems

Chair: Shaonpius Mondal (University of Nebraska - Lincoln, USA)
Elizabeth Jeannette Cieniewicz (Clemson University, USA)

SE07 C01

UNDERSTANDING WHEAT CURL MITE TRANSMITTED VIRUSES AND THEIR CO-INFECTION DYNAMICS IN WHEAT

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Wheat is a staple food crop and is grown worldwide. The United States is a significant wheat producer globally, but US wheat production is constrained by several viruses, some of which are transmitted by wheat curl mites (*Aceria tosichella* Keifer). In the US, wheat streak mosaic virus (WSMV), Triticum mosaic virus, (TriMV), and High Plains wheat mosaic virus (HPWMOV) are major production constraints of wheat. Two genetically distinct wheat curl mites (Type 1 and Type 2) are present in US wheat fields. WSMV and HPWMOV are transmitted by Type 1 and Type 2 wheat curl mites. In contrast, TriMV is transmitted only by Type 2 mites. Since these three viruses are transmitted by wheat curl mites, mixed infections are often present in wheat fields. Co-infection of wheat by WSMV and TriMV induces disease synergism in wheat with exacerbated yield loss. The early stage of WSMV+TriMV synergistic interaction is asymmetrical and depends on the order of infection of plants by interacting partner viruses. WSMV alters Type 2 wheat curl mite transcriptome profile, positively contributing to their survival and development. In contrast, TriMV has a negative impact on the mite survival. HPWMOV, the other important virus, was first reported in wheat and corn fields during the 1990s and since then occurs in the field with varying severity. In recent years, HPWMOV became a problem to the seed companies due to the import restriction concerns during seed shipment. Ongoing research is comparing wheat curl mite microbiomes using metagenomics approaches to understand the differential transmission of these viruses by mites as well as their survival and development in coinfecting wheat. Another ongoing research project focuses on co-infection dynamics of WSMV, TriMV, and HPWMOV in singly, doubly, and triply infected wheat plant.

SE07 C02

EXPLORING THE ROLE OF POLLINATORS AND THRIPS IN SPREADING POLLEN-BORNE VIRUSES IN FRUIT CROPS IN THE SOUTHEAST U.S.

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Peach production is important to the economy of South Carolina, which is the 2nd largest peach producer in the United States. Peach trees are susceptible to numerous virus and viroid diseases, which are mainly transmitted through grafting and vegetative propagation. However, some of the most important viruses in peach production are Prunus necrotic ringspot virus (PNRSV) and prune dwarf virus (PDV). These are both ilarviruses and are transmitted through pollen and seed. Spread of pollen-borne viruses in stone fruit orchards has been studied several decades ago, but the mechanisms of pollen transmission and the role of insects are still not well understood. We have conducted several studies in orchards in South Carolina and Georgia, USA, to investigate the incidence of bees and thrips carrying PNRSV and PDV in orchards, assess virus movement between orchards and wild hosts, and assess the impact of these viruses on pollen health. We have conducted surveys of bees in thrips in orchards, and found that bees were carrying virus-positive pollen in ratios that were reflective of the virus incidence in the orchard, but that bees were also carrying virus-positive pollen when collected in clean orchards, illustrating their potential to transfer virus-positive pollen from orchard to orchard. PNRSV was detected in wild black cherry and Carolina cherry laurel near peach orchards, but phylogenetic analyses and data on bloom timing suggest that populations of PNRSV are not moving from wild reservoirs to peach orchards, but more likely the opposite. Both viruses have negative impacts on peach pollen health, resulting in shorter, wider pollen grains as determined by scanning electron microscopy and reduced viability and germination rates. Overall, these studies have increased our ecological knowledge on these viruses in peach orchard ecosystems, and have also spurred more questions to address in future research.



SE07 C03

IMPACT OF DOUBLE VIRUS INFECTIONS ON THE YIELD OF MELON PLANTS SUBJECTED TO WATER STRESS

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Climate predictions suggest a decrease in rainfall in the Mediterranean basin in the near future; this may cause extended periods of drought which will impact virus-vector-plant interactions, likely threatening agricultural production. In this study, we evaluated how the combination of water stress and vector-borne viruses affects melon biomass, flowering, fruit setting, marketable yield and viral infection symptoms among other physiological variables. Melon plants (cv. Monique) were grown in the greenhouse until harvest under two water regimes and infected with the non-circulative cucumber mosaic virus (CMV) and/or the circulative cucurbit aphid-borne yellows virus (CABYV). Plants were watered regularly to maintain two water regimes: 80% and 30% of field capacity. Plants were inoculated with CMV and CABYV at the 4-leaf stage by aphids one week after initiation of the water stress regimes. Water stress had the highest impact on melon yield components, reducing melon biomass, flowering and marketable yield. CMV-infection had a much higher impact on melon biomass and yield than CABYV-infection as shown by reduced growth and clear mosaic symptoms that appeared soon after virus inoculation. CABYV-symptoms appeared much later and were less evident on plants grown under water stress. In comparison to mock-inoculated controls, CABYV infection delayed the onset and increased the rate of flowering, leading to a 36% increase in final flower number. Double-infected plants had similar effects on plant biomass than those infected with CMV alone but melon fruit weight was lowest in plants with double infection. The impact of single and double virus infections on melon plants when grown under water scarcity will be discussed under the scope of a changing climate.

SE07 C04

MOLECULAR MECHANISMS UNDERLYING TRANSMISSION OF LIBERIBACTER BY PSYLLIDS

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Liberibacter is a complex of fastidious bacterial species that cause severe diseases in many important agricultural crops. Carrot Yellows (CY) is a devastating disease that has been a major constraint to carrot cultivation in many countries, causing significant yield losses. Candidatus Liberibacter solanacearum (CLso) is the causative agent of CY and is transmitted by the carrot psyllid *Bactericera trigonica*. Management of diseases caused by Liberibacter species including CY, the citrus greening disease (Huanglongbing), and the Zebra Chip disease depends on chemical control for controlling psyllid populations, leading to health, economic and environmental burden. Basic understanding the transmission of CLso and other Liberibacter species by their psyllid vectors is fundamental for devising sustainable

management strategies. Persistent transmission of vector-borne pathogens involves critical steps of adhesion, cell invasion, and replication inside the insect gut cells before passage to the hemolymph. Our previous studies have used microscopy and expression analyses and confirmed a role of the Endoplasmic Reticulum (ER) in inducing immune responses and subsequent molecular pathways that lead to programmed cell death (apoptosis) upon CLso infection in the insect gut. Key genes involved in ER stress-related pathways were found to be differentially regulated in CLso-infected psyllids. Feeding CLso-infected and uninfected psyllids with different ER stress agents resulted in significant regulation of PERK, one of the three sensors that activate the unfolded protein response (UPR). These results demonstrate that CLso actively regulates and manipulates gene expression in the insect gut during transmission, and this site is a strong candidate for targeting and regulating the transmission.

SE07 C05

BIOLOGICAL, EPIDEMIOLOGICAL, AND ENVIRONMENTAL FACTORS DRIVE THE PREVALENCE AND DOMINANCE OF WHITEFLY-TRANSMITTED VIRUSES IN CUCURBITS

William M. Wintermantel

USDA-ARS, Salinas, CA

Increasing populations and range expansion of the whitefly *Bemisia tabaci* in cucurbit-producing regions of the southern and western United States (U.S.) has led to an associated emergence of whitefly-transmitted viruses. Since the late 1990s, U.S. cucurbit production has witnessed the emergence of five new whitefly-transmitted viruses. These viruses have become important economic threats to cucurbit production, reducing yield and fruit quality, as well as plant vigor. The virus(es) that are most economically important vary by region and crop. Recent surveys conducted in the western, southwestern, and southeastern U.S. demonstrated that mixed virus infections, which create challenges for disease management and plant breeding, are common. These mixed infections often include the criniviruses cucurbit yellow stunting disorder virus (CYSDV) and cucurbit chlorotic yellows virus (CCYV), the begomovirus cucurbit leaf crumple virus (CuLCrV), and the ipomovirus squash vein yellowing virus (SqVYV). Differences in cropping and environmental conditions influence vector prevalence and virus abundance among regions. Recent research investigated reservoir hosts that are epidemiologically significant by region as well as the impact of infection timing and virus competitiveness on virus prevalence and dominance in mixed infections. Understanding these factors may aid in predicting which virus(es) may be most impactful within a region and season. During the fall of 2023, a new begomovirus was identified infecting melon and watermelon in the southwestern U.S. This whitefly-transmitted virus was found in fields throughout a region where CYSDV has traditionally been the most economically important virus for fall cucurbit production. Continued research and improved communication and stakeholder education are needed to mitigate the impact of established and emerging whitefly-transmitted viruses, including the new begomovirus, in the U.S., particularly in regions where the whitefly vector is well-established.



Concurrent Sessions

Tuesday, 2 July

SE07 C06

SEASONAL SPREAD OF COTTON LEAFROLL DWARF VIRUS BY THE COTTON APHID IN THE UNITED STATES

Alana L. Jacobson¹, John Mahas¹, Jessica B. Mahas¹, Charles Ray², Kassie Conner², Phillip Roberts³, and Sudeep Bag⁴

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Cotton leaf roll dwarf virus (CLRDV) has been reported to infect *Gossypium hirsutum* (cotton) in Asia, South America, and the United States. CLRDV is spread to cotton by *Aphis gossypii*, the cotton aphid, in a persistent and non-propagative manner. Research over the past five years has been conducted to better understand the epidemiology of this pathosystem. Studies examining weekly seasonal dynamics of virus spread by the vector in three distinct geographical locations showed that cotton aphids are active mid- and late-season, and virus spread occurs 3-15 weeks during the cotton growing season, depending on the location. Data from on-farm studies characterizing abundance of *A. gossypii* and CLRDV over a two-year period at 64 locations characterized the causal relationships between aphid abundance, CLRDV incidence, weed host presence and landscape features using directed acyclic graphs. Additionally, aphid gut content analyses were conducted to determine alternate hosts of the aphids prior to infesting cotton in the Southeastern United States Cotton Belt. A summary of the major findings from these recent studies as they relate to vector-virus-host relationships responsible for CLRDV spread will be presented and discussed.



MC2

11.30-14.00 Concurrent Session 8

Networking Tools to Manage Transboundary Pests

Chair: Daren Mueller (Iowa State University, USA)

Laura Iles (North Central Integrated Pest Management Center, USA)

SE08 C01

ENHANCING CROP PROTECTION: THE COLLABORATIVE IMPACT OF THE CROP PROTECTION NETWORK (CPN)

Daren Mueller¹, Kiersten Wise², Albert Tenuta³

1. Crop Protection Network, Iowa State University,
2. University of Kentucky, 3. Ontario Ministry of Agriculture

The Crop Protection Network (CPN) represents a multi-state and international collaboration among university and provincial Extension specialists, as well as public and private professionals. Rooted in the provision of unbiased, research-based information, the CPN aims to effectively communicate relevant knowledge to farmers and agricultural personnel, facilitating informed decision-making regarding the protection of alfalfa, cotton, corn, small grains, and soybean crops. Through multi-state and international efforts, the CPN not only enhances the visibility and success of agricultural extension but also serves as a vital conduit for valuable crop protection information to farmers, agribusinesses, and educators. By leveraging online tools designed for crop scouts and researchers, the CPN ensures efficient data collection and analysis, augmenting the accuracy and effectiveness of crop protection strategies. The network actively disseminates impactful content encompassing practical guidelines and cutting-edge research findings, empowering stakeholders with up-to-date information to effectively address crop protection challenges. At the heart of the CPN lies a robust network of more than 300 specialists, encompassing experts from 40 U.S. states and Ontario, Canada. This collaborative platform fosters interdisciplinary collaboration, research partnerships, and the seamless dissemination of scientific knowledge. Anchored in unbiased, research-based approaches, the CPN harnesses collective expertise to tackle complex challenges in crop protection, elevating the efficacy and sustainability of agricultural practices.

exchange. Their efforts focus on tailored strategies for crops, pests, education, outreach, and invasive species management to develop and implement targeted IPM solutions. Working groups enhance the impact and visibility of IPM practices in the region. Pest Management Strategic Plans (PMSPs) and Crop Profiles (CPs) play a pivotal role in the realm of integrated pest management (IPM) by facilitating the collection of crucial data from growers, commodity associations, specialists, processors, consultants, and other agencies. By soliciting input from growers, commodity associations, university specialists, food processors, crop consultants, and other agencies, PMSPs ensure a comprehensive representation of the industry's priorities, research needs, regulatory considerations, and educational requirements. PMSPs are a valuable resource for regulatory agencies and the broader IPM community. These plans incorporate vital information provided by growers and consultants, encompassing the utilization of pest control methods, the rationale behind their selection or exclusion, the efficacy of control measures, activity timelines during pest infestations, application techniques, and grower priorities. Importantly, PMSPs inform regulatory agencies about the active ingredients required to safeguard crop production and introduce potential solutions to combat resistance issues. This talk emphasizes the collaborative nature of working groups and PMSP development, which involves key stakeholders from various sectors. By integrating scientific knowledge, technological advancements, and practical experiences, they reduce pest risks while safeguarding human health, food supplies, and the environment.

SE08 C03

YOUTUBE TRANSFORMS IPM EDUCATION: FROM INSIGHTS TO ACTION

Brandon Kleinke

YouTube, Iowa State University

This presentation explores the power of YouTube in shaping and enhancing Integrated Pest Management (IPM) education. Set aside old textbooks and static slides – it is time to harness the dynamic nature of video to empower educators, researchers, and most importantly, stakeholders across various fields. Videos can dissect complex IPM concepts with clarity, showcase real-world applications that can be immediately implemented, and engage diverse audiences who may not have engaged with traditional learning media. Drawing insights from successful channels, this talk will unlock the importance of YouTube for IPM, crafting accessible, visually appealing, and interactive content that resonates with a broad audience. Additionally, the presentation addresses considerations such as the platform's potential for community-building, empowering educators, researchers, and practitioners to strategically utilize YouTube in advancing

SE08 C02

NETWORKING FOR EFFECTIVE INTEGRATED PEST MANAGEMENT: SPOTLIGHT ON WORKING GROUPS AND PEST MANAGEMENT STRATEGIC PLANS IN THE REGIONAL IPM CENTERS

Lynnae Jess¹, Laura Iles²

1. IPM Working Groups, Michigan State University, 2. Iowa State University

Working groups within the Regional IPM Centers play a crucial role in fostering collaboration and networking for effective integrated pest management (IPM). Comprised of researchers, extension specialists, industry representatives, growers/practitioners, and professionals, these groups address specific pest management challenges in the North Central region through collective problem-solving and knowledge



Concurrent Sessions

Tuesday, 2 July

IPM programs. This presentation isn't just about watching videos – it's about equipping you with the tools and strategies to turn YouTube into a useful tool.

SE08 C04

INTEGRATED WEED MANAGEMENT WITH GROW: GETTING RID OF WEEDS

Michael Flessner

Getting Rid of Weeds (GROW), Virginia Tech University

Herbicide-resistant weeds have become a significant transboundary economic concern in U.S. agriculture, prompting the need for integrated weed management (IWM) practices. GROW (Getting Rid of Weeds) is a scientist-led network that offers up-to-date, tested, and vetted resources to help farmers adopt IWM tactics. The GROW team, consisting of agronomists, weed scientists, communicators, and economists from various regions in the U.S., collaborates on research and outreach to help farmers combat herbicide resistance through the implementation of diverse weed control strategies that are practical and adoptable. This talk will delve into the role and impact of GROW in advancing IWM practices through both research and outreach across state boundaries. GROW currently has three major research themes: cover crops, harvest weed seed control, and precision weed management. This collaborative, multi-region research is featured on GROW's website and social media platforms alongside other innovative research across the U.S. GROW's outreach team also produces weekly news posts, Farmer Forums (farmer-led webinars), educational web pages, decision support tools, and farmer case studies to facilitate evidence-based decision-making and adoption of IWM practices. Any resources posted or promoted by GROW have undergone rigorous testing to ensure their reliability and effectiveness in real-world farming contexts. The presentation will underscore the benefits of adopting IWM practices, including improved weed control, reduced herbicide dependency, enhanced long-term sustainability, and increased farm profitability. By integrating multiple weed control tactics, growers can effectively manage herbicide-resistant weeds while mitigating the risk of further resistance development.

SE08 C05

DEVELOPING TOOLS TO EMPOWER STAKEHOLDERS FOR EFFECTIVE PEST MANAGEMENT: THE US NATIONAL PREDICTIVE MODELING TOOL INITIATIVE

Kaitlyn M. Bissonnette

Director of Agricultural Research, Cotton Incorporated

Agricultural production is a dynamic system encompassing multiple levels of inputs, outputs, and in season finesse to produce high quality feed, fiber, and products. In recent year, critical challenges are facing those making decisions at the farm level including extreme weather events, changes in pathogen and pest dynamics, and pesticide resistance. The National Predictive Modeling Tool Initiative (NPTMI), comprised of more than twenty five universities, government agencies, and private laboratories, aims to connect stakeholders with predictive tools for pest management at the farm level. Since its inception in mid-2020, NPTMI has focused on major and emerging diseases of corn, cotton, and

wheat in the US and has recently incorporated pulse crops. Through centralization of data management and coordination at the national level, commodity specific models have been developed for critical disease threats. This initiative is ongoing and further model development drives forward the goals of sustainable agricultural production through reducing pesticide applications and increasing stakeholder return on investment.

SE08 C06

NETWORKING TACTICAL SCIENCES IN THE UNITED STATES TO ENHANCE PLANT AND ANIMAL BIOSECURITY

Marty Draper

Tactical Sciences Network, Kansas State University

The Tactical Sciences Coordination Network (TSN) aims to enhance collaboration and strategic coordination among the tactical science biosecurity programs administered within the U.S. land-grant university system. The TSN brings together programs that address the threat response continuum through detection and diagnostics of agricultural plant and animal pathogens and pests, deployment of novel crop and livestock production and protection technologies, and regulatory systems support. The network also engages industry stakeholders served by those programs to identify opportunities to optimize responsiveness. The eight programs participating include the National Plant Diagnostic Network (NPDN), National Animal Health Laboratory Network (NAHLN), Extension Disaster Education Network (EDEN), the Crop Protection and Pest Management (CPPM), Regional Integrated Pest Management Centers, Specialty Crop Pest Management Program (IR-4), and Minor Use Animal Drug Program (MUADP). These programs may interact with industry and Federal government agencies at various stages along the threat response continuum from threat to early detection, diagnosis, response, management, and recovery, contributing to the safeguarding our food systems. Familiarity of the other programs opens opportunities for collaboration and communication across common interests. The great goal is for improved relationships across programs to enhance effectiveness, efficiency, relationship, and accountability. Through the TSN coordinated strategy and with leveraged customer insights, collective efforts to address plant and animal diseases and other threats and strengthened. This collaborative customer-centric approach enhances the visibility and value of TSN programs, ensuring effective responses to evolving tactical sciences challenges. By viewing customers broadly, uniting stakeholders with university program people and government regulators, aligning objectives, and leveraging customer insights, the TSN strengthens the collective response to plant and animal disease threats. This coordinated approach not only enhances the effectiveness of tactical sciences programs but also fosters trust, transparency, and mutual understanding among participants.

**SE08 C07****THE NATIONAL PLANT DIAGNOSTIC NETWORK: TRANSREGIONAL COLLABORATIONS FOR PLANT PROTECTION IN THE UNITED STATES****Zachary Schumm¹, Chelsea Harbach²***1. National Plant Diagnostic Network, Iowa State University,**2. Iowa State University*

The National Plant Diagnostic Network (NPDN), comprising over 125 diagnostic labs in five regions across the United States and its territories, plays a pivotal role in safeguarding national plant health. Diagnostic laboratories within the network are responsible for diagnosing plant health problems for diverse clientele and have a responsibility to report diagnoses to the NPDN National Data Repository to track pest incidences nationwide. Experts within NPDN laboratories span the fields of plant pathology, entomology, and nematology, collectively forming a dynamic effort armed with state-of-the-art and validated diagnostic technologies. Through rapid and accurate diagnoses, the NPDN and member labs can facilitate timely response strategies with federal agencies and state plant health regulators. Through diagnostics, knowledge-sharing, and in-network committees dedicated to NPDN progress, the NPDN fosters an environment of continuous improvement and adaptability. Moreover, the NPDN and its diagnostic labs serve as a vital resource hub for farmers, researchers, policymakers, and the public, providing critical information and insights for informed decision-making. Herein, we discuss the NPDN framework and demonstrate network and diagnostic laboratory relevance using the Iowa State University Plant and Insect Diagnostic Clinic as a model.

of growers recalled all primary SCN Coalition messages and reported 6% to 18% increases in use of SCN management tools between 2015 and 2020. Grower-reported increases in management and yield impacts due to The SCN Coalition activities resulted in financial gains to U.S. soybean growers far exceeding \$100M USD. The SCN Coalition serves as a case study and example of how a PPP with shared vision, resources and successes can mitigate the largest threats in modern agriculture.

SE08 C08**HOW A PUBLIC-PRIVATE PARTNERSHIP CHANGED THE MANAGEMENT OF A BILLION-DOLLAR PEST: THE SCN COALITION****Sam Markell***SCN Coalition, North Dakota State University*

The Soybean Cyst Nematode (SCN), *Heterodera glycines*, causes over \$1B USD in losses to the United States soybean crop annually. Barriers to reducing yield loss are both biological and social, including the nematodes continued geographic expansion and adaptation to genetic resistance and limited awareness and active management among growers. The scale of the SCN problem and barriers to its management limit the influence and impact that any individual organization can have on mitigation of the problem. In 2015, a public-private partnership (PPP) of universities, soybean grower organizations and agro-chemical and -seed companies to address SCN was envisioned. Over the next three years, dozens of partners committed to a shared vision of increased grower awareness, active management and reduced yield loss to SCN. Since its launch 2018, "The SCN Coalition" executed an extension educational campaign that generated over 80 million potential impressions in the U.S. agricultural media, tens of millions of potential impressions in social media, garnered over 2 million video views, and delivered thousands of trainings, sampling programs and activities for soybean growers. To measure effectiveness and impact, soybean grower awareness and behavior were measured by national market research conducted in 2015 and 2020. Over 50%

MC3

11.30-14.00 Concurrent Session **4B**

Grapevine Trunk Diseases

Chair: Epaminondas Paplomatas (Agricultural University of Athens, Greece)
Laura Mugnai (Dagri, University of Florence, Italy)

SE04 C08

AN OVERVIEW OF GRAPEVINE TRUNK DISEASES AND FUSARIUM ROOT-BASAL ROT IN TURKISH GRAPEVINE NURSERIES

Davut Soner Akgül¹, Nurdan Güngör Savaş², Murat Yıldız³, İzzet Bülbül⁴, Mümine Özarslandan⁴, Serkan Önder⁵, Josep Armengol⁶, Maela Leon⁶

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Grapevine trunk diseases (GTD) and root-basal rots are two major disease complexes in grapevine nurseries worldwide. They are primarily associated with Petri and black-foot pathogens, *Botryosphaeria-Eutypa-Phomopsis* dieback fungi, and soil-borne species such as *Fusarium*, *Rhizoctonia*, *Macrophomina*, and oomycete fungi. This study aimed to reveal the current status of GTD and *Fusarium* root-basal rot diseases and their related pathogens in Turkish grapevine nurseries. Dormant marketable plants were randomly sampled from 43 nurseries in seven geographical regions of the country. Fungi were isolated from the roots and internal basal tissues and identified based on PCR, gene-sequencing (ITS, TEF1- α , calmodulin, histone H3, RPB2), and phylogenetic analyses. Pathogenicity tests were conducted on one-year-old 1103 Paulsen rootstock plants in greenhouse conditions by inoculating the representative fungi. Virulence of the isolates was estimated by internal wood necrosis (in stem or basal tissues) and dry root weights. In total, 320 fungal isolates were obtained by standard mycological isolation procedure and isolation rates of the species assigned species composition and disease prevalence. According to identification and pathogenicity test results, six species (*Cytospora viticola*, *Diaporthe ampelina*, *Diplodia seriata*, *Lasiodiplodia brasiliensis*, *Neofusicoccum parvum*, *Truncatella angustata*) were related to dieback diseases, six species (*Cadophora ferruginea*, *C. luteo-olivacea*, *C. malorum*, *Phaeoacremonium minimum*, *P. tuscanicum*, *Phaeomoniella chlamydospora*) to Petri disease, seven species (*Cylindrodendrum alicantinum*, *Cylindrocladiella peruviana*, *Dactylonectria macrodidyma*, *D. novozelandica*, *D. torresensis*, *I. liriiodendri*, *I. robusta*) to black-foot, three species to *Fusarium* root and basal rot (*Fusarium annulatum*, *F. falciforme*, *F. oxysporum*) disease respectively. The prevalence of disease complexes was calculated as 27.9% for trunk disease, 32.5% for Petri disease, and 92.9% for black-foot and *Fusarium* root-basal rot diseases.

This study revealed that soilborne fungal pathogens were the main phytopathological problems underlying plant mortality in Turkish grapevine nurseries.

SE04 C09

UNDERSTANDING EARLY RESPONSES TO ESCA-PATHOGENS OF WOOD TISSUES: A WAY TO OPEN CURTAINS ON MECHANISMS OF ACTION OF BIOCONTROL AGENTS

Ana Romeo-Olivan¹, Justine Chervin^{2,3,4}, Virginie Puech-Pages², Sylvie Fournier^{2,3,4}, Guillaume Marti^{2,3,4}, Olivier Rodrigues¹, Thierry Lagravier¹, Bernard Dumas² and Alban Jacques¹

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To understand the early grapevine trunk responses to *P. minimum* and *P. chlamydospora* we compared global transcriptomic and metabolomic wood responses. The analysis of the global transcriptomic reprogramming allowed the identification of specific modules of differentially expressed genes associated to each pathogen. Functional analysis of these genes reveals main differences in Signaling, Hormonal signaling and Biotic stress response-related genes. Global metabolomics highlighted a group of flavonoids and stilbenoids that were overproduced in infected wood of *Vitis vinifera*. Our results showed both different gene expression reprogramming and metabolomic specific signature depending on the pathogen bringing evidence that grapevine trunk can differently perceive and respond to *P. minimum* and *P. chlamydospora*. To open curtains on mechanisms of action of biocontrol agents we used Vintec®, a *Trichoderma atroviride* SC1-based commercial formulation that showed efficiency of the esca disease treatment. We evaluated whether the pre-inoculation of the BCA Vintec® modifies early molecular responses to *P. minimum* and *P. chlamydospora*. Global transcriptional analysis allowed the identification of clusters of genes differently regulated in the absence or the presence of the BCA Vintec®. "Phenylpropanoid metabolism and stilbene biosynthesis-related genes are significantly represented among the genes differently expressed in the presence of the BCA Vintec®. A global metabolomic analysis highlighted clusters of plant compounds synthesized in response to pathogens and biocontrol. These compounds belong mainly to the stilbene class. Five relevant biomarkers were chosen

for in vitro evaluation of their antifungal activity on *P. chlamydospora*. The results suggest that these compounds may play a role in limiting the in planta development of esca pathogens. Altogether, our results suggest that the BCA Vintec® enhances trunk responses to esca pathogens.

SE04 001

EXPLORING CYTOSPORA CANKER DIVERSITY IN EUROPEAN GRAPEVINES: MOLECULAR AND PHYLOGENETIC CHARACTERIZATION

Da Cunha Maia Leal C.¹, Špetík M.², Eichmeier A.², Bujanda R.¹, Gramaje D.¹

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2. Mendeleum—Institute of Genetics, Mendel University in Brno, Brno, Czech Republic

Cytospora canker, a grapevine trunk disease (GTD) caused by various species of the genus *Cytospora*, has been extensively studied in the US but remains underexplored in Europe, and its incidence and associated species are not truly known. This study aimed to investigate the potential of *Cytospora* spp. to penetrate grapevine plants through pruning wounds and to molecularly characterize a diverse collection of isolates from this genus. The assay was carried out at a commercial vineyard located in Samaniego, Álava region (Northern Spain). The vineyard in Samaniego was planted in 2001 (19-years-old) with 'Tempranillo' cultivar grafted onto 110 Richter rootstock. On February 2020, 1-year-old canes of 30 vines were spur-pruned to three buds using secateurs, coinciding with the common pruning time in this region. The experiment was repeated the following seasons (2021–22 and 2022–23), with pruning on the same date. Canes were harvested from vines above the second bud (about 10 cm long pieces) approximately 12 months after pruning and brought to laboratory for GTD pathogens assessment by plating wood fragments onto culture media. Colonies resembling *Cytospora* spp. were transferred to PDA plates for their identification based on DNA sequence data of ITS region and *tef* gene. Preliminary analysis identified a total of 347 isolates, representing 8 different *Cytospora* species. Isolates belonged to *C. viticola* (n=311), *C. cotini* (n=25), *C. ceratosperma* (n=2), *C. hippophaicola* (n=2), *C. salicacearum* (n=2), *C. salicina* (n=1). Notably, we observed four isolates potentially representing two novel *Cytospora* species, highlighting the richness of fungal diversity responsible for this GTD. This study contributes to the understanding of *Cytospora* canker incidence and diversity. The identification of multiple *Cytospora* species, including potential novel taxa, highlights the need for continued research to effectively manage this fungal disease and ensure grapevine health and productivity. Ongoing efforts involve further species identification and phylogenetic analyses using additional genomic markers to enhance phylogenetic accuracy.

SE04 002

AROUND THE WORLD IN EIGHT WHITE ROT SPECIES: UNDERSTANDING WOOD DEGRADATION IN ESCA COMPLEX OF DISEASES

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The Esca complex of diseases (ECD) is widespread globally, varying in severity across regions and causing important economic losses. Recent studies have shown that some effective control strategies for ECD foliar symptom expression are greatly related to the presence of white-rot (WR) agents in the wood, thus raising awareness of this symptom and related causal agents. However, the only well-investigated WR agent in ECD is the European *Fomitiporia mediterranea*. Research shows that the pathogen, in addition to enzymatic degradation, has also access to a non-enzymatic chelator-mediated Fenton (CMF) pathway. The study aims to investigate the degradation capabilities of other ECD-associated WR agents such as *F. capensis*, *F. langloisii*, *F. polymorpha*, *F. australiensis*, *Tropicoporus texanus*, *Inonotus vitis*, and *Stereum hirsutum*. Production and activity of lignocellulosic enzymes (i.e. Laccases and Class II PODs) were evaluated from 1-week-old and 2-week-old grapevine-sawdust liquid cultures. The chemical spectrum of grapevine sawdust used for the cultures was obtained through FT-IR spectroscopy. Non-enzymatic degradation capabilities of ECD-WR species were assessed by investigating if the Low Molecular Weight (LMW) fractions of iron-deprived fungal liquid cultures were able to accomplish all the steps of the CMF reaction i) iron (III)-reduction, ii) hydrogen peroxide production during ferrous iron oxidation, iii) hydroxyl radical generation. LMW fractions were identified through HPLC analysis. Lastly, microscope analyses were performed to investigate the decay pattern of WR species. Preliminary results reveal differences in the secretome of ECD-associated WR agents in lignocellulosic biomass degradation. This study also shows and confirms that the CMF degradation pathway classically associated mainly with brown-rot degradation is widespread among ECD-associated WR agents belonging to different orders. Overall, the research provides new insights into the ability to degrade woody tissues in grapevine by ECD-associated WR agents. These findings are crucial for future ECD symptomatology evaluations and control strategy development.



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SE04 003

INFECTION ROUTES FOR PATHOGENS CAUSING GRAPEVINE TRUNK DISEASES IN CALIFORNIA NURSERY

Rolshausen P.¹, Todd C.², Garcia J.³, Cantu D.³

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Grapevine Trunk Diseases (GTD) are caused by several taxonomically unrelated fungal pathogens. Infection with GTD-pathogens occurs in vineyards and nurseries and leads to wood decay that compromises the biological function of the vascular system and results in loss of fruit yield and shortening of vineyard lifespan. In California nurseries, the causal agents have not been thoroughly described and infection routes remain hypothetical. The overarching goal of this project was to increase the quality of plant nursery stock by reducing GTD-pathogen load and identify the critical steps that are conducive to the spread of these pathogens during the propagation phase. We collected 1400 industry-standard vines from five California nurseries at four stages of the plant propagation pipeline, including cuttings from mother vines, as well as grafted callusing vines, green vines and dormant vines. Disease diagnosis from plant samples was done by traditional microbial isolation techniques on culture medium, coupled with Sanger sequencing for taxa identification and by amplicon-based Illumina sequencing to profile the mycobiome associated with vines. We also applied a culture-independent method to 115 environmental samples from four nurseries including water baths, callusing medium and planting soils for potting green vines to determine if GTD-pathogens were also present in these environments and could spread and contaminate plant nursery stock. Results showed that infections stemmed from three origins. First, several pathogens were commonly found at a high incidence and abundance in mother vine cuttings. The second origin was exogenous infections of GTD-pathogens from the various environmental substrates (soil, water) that were in contact with the vines during the propagation. The last origin occurred through airborne infections of green and dormant vines. This work will help with implementing adapted management practices to reduce the incidence of pathogens in plant stock below acceptable levels.

SE04 004

IDENTIFICATION AND CHARACTERIZATION OF FUNGAL SPECIES ASSOCIATED WITH GRAPEVINE TRUNK DISEASE IN SOUTH TYROL (NORTHERN ITALY)

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1. Free University of Bozen-Bolzano, Bozen-Bolzano, Italy, 2. South Tyrolean Extension Service for Fruit and Wine Growing, Lana, Italy

Grapevine trunk diseases (GTDs) pose a severe threat to European vineyards. The diseases are associated with a complex of fungi, which may differ among winegrowing regions. In recent years, extensive damage caused by GTDs has been observed in South Tyrol (northern Italy), but the

spectrum of involved fungal species is still unclear. Therefore, this study aimed to investigate the fungal species linked to GTDs in South Tyrol. Symptomatic wood samples of 217 vines from 43 vineyards were collected during dendrosurgery carried out in winter 2019/2020. Fungal isolations were performed on Potato Dextrose Agar. The 225 obtained pure fungal cultures were classified into phenotypic groups based on morphological characteristics of the colonies and determined by ITS sequencing. *Diplodia seriata*, generally associated with Botryosphaeria dieback, was the most common species (36% of the isolates). Another fungal genus, *Arthrrium*, represented 5% of the isolates, although it has never been related to any GTD. Consequently, the focus was further laid on these two fungi. The isolates were characterized by multi-locus sequence typing, followed by in vitro and in vivo assessment of their potential ability to infect grapevine. Assays on cellulose basal medium revealed that *D. seriata* and *Arthrrium* spp. can be classified as cellulolytic fungi, and both are able to grow on grapevine wood extract. While mycelium plugs of *D. seriata* inoculated on detached one-year-old canes of the cultivar Sauvignon blanc showed no visible lesions, several isolates of *Arthrrium* spp. induced discolorations of the wood after 28 days. In contrast, none of the isolates was capable of inducing lesions on the stems of potted grapevine plantlets, but the inoculation on green shoots resulted in necrotic lesions of the epidermis and cortex tissue. The understanding of the mechanisms involved in the pathogenicity of different fungal species is fundamental to a better management of GTDs.

SE04 005

DEVELOPMENT OF A BIOCONTROL-BASED STRATEGY FOR THE MANAGEMENT OF PHAEOMONIELLA CHLAMYDOSPORA IN GRAPEVINE NURSERIES

Tsoukas C.¹, Tzima A.¹, Bakasietas K.², Paplomatas E.¹

1. Laboratory of Plant Pathology, Department of Crop Science, Agricultural University of Athens, Iera Odos 75, 11855, Athens, Greece, 2. Vine Nurseries Bakasietas, Leontio Nemea, 20500, Greece

The viticulture and wine sector in Greece, is plagued by several trunk pathogens. Amongst them *Phaeomoniella chlamydospora* is considered as the most severe one. Over the past two decades, the widespread emergence of this fungus in grapevine nurseries has raised concerns about the future sustainability of viticulture, while the lack of agrochemicals and the unique lifestyle of this notorious pathogen, render its management quite challenging. The aim of this study was to develop a biocontrol-based strategy that could be incorporated into the standard production practices in nurseries, targeting the mitigation of *P. chlamydospora* infections in propagation material. Isolations of endophytic bacteria were conducted from roots of phenotypically healthy young and mature grapevines and were evaluated in terms of their in vitro antagonistic ability against *P. chlamydospora* in a modified dual culture assay. Four out of twenty-four bacterial strains inhibited the pathogen's growth by 8% to 23,33% and were further evaluated in greenhouse trials by employing a developed talc-based formulation. Strain ba4 was the most effective, leading to a disease reduction by 34,04% in artificially inoculated, and 57,28% in naturally infected vines. Moreover, ba4-treated vines exhibited reduced vascular



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discoloration by 42,55%, and pathogen's concentration was 75,93% and 33,97% lower compared to the artificially inoculated and naturally infected vines, respectively. In field trials, application of strain ba4 in grafted vines led to a reduced vascular discoloration by 25,71% and to pathogen's biomass decrease by 84,69%. Whole genome sequencing revealed that strain ba4 belongs to species *Bacillus halotolerans*. Sequence analyses successfully predicted the presence of several gene clusters that regulate the production of secondary metabolites with antifungal properties, speculated to induce grapevine defense responses. The findings of this study indicate that the grapevine endophytic bacteriome could be utilized for the control of trunk pathogens leading towards the sustainable production of high-quality propagation material.

This research has been financially supported by the General Secretariat for Research and Technology – Greece. RESEARCH-CREATE-INNOVATE national call: "IMPROVITO".



CONFERENCE I

11.30-13.00 Concurrent Session **9**

New Advances and Technologies for Postharvest Diseases Control

Chair: **Davide Spadaro** (Disafa, University of Turin, Italy)

James Adaskaveg (Microbiology and Plant Pathology, University of California, USA)

SE09 C01

POSTHARVEST LOSS MANAGEMENT THROUGH NOVEL TECHNOLOGIES - BRIDGING THE GAP BETWEEN RESEARCH AND PRACTICE

Gianfranco Romanazzi

Department of Agricultural, Food and Environmental Sciences, Marche Polytechnic University, Ancona, Italy

Food loss and waste (FLW), often caused by plant pathogens in the field and after harvest, is a global challenge recognised by international governments and organisations. Reducing FLW is key to sustainably ensure nutritional food security for an increasing world population. It is a target of the Sustainable Development Goals of the United Nations, and the Farm to Fork Strategy of the European Green Deal. Losses are considered the ones that occurs from the growers to the retailers, while waste occurs when the consumer is involved, from retailers to home or other food services. Postharvest diseases affect fruit and vegetables with an infection that can occur in the field, with latent infections that develops after harvest, or with pathogen that benefits from wounding occurring during the harvest and postharvest manipulation. After harvest, the fresh produce becomes weaker and defenses declines due to ripening and softening of tissues, that allows the pathogen to infect, leading to loss of single fruit or even of the entire box, when it is including in plastic containers or woody boxes. This loss or waste has a very high economic, social and environmental value, since the fresh fruit and vegetables reach this point after being grown, picked, transported, cold stored and exposed to shelf life, that can occur in few days or even months (e.g. for apples, pears or kiwifruit). An integrated list of preharvest and postharvest technologies are important to preserve the fresh fruit and vegetables from the cultivation to the consumer home, that integrate use of decision supporting systems in the field for proper application of synthetic fungicides or alternatives (biocontrol agents, natural compounds, etc.), with a monitoring of isolates resistant to fungicides in the field and in the packinghouses and alternation of fungicides with a different mechanism of action, both in the field and even for the few fungicides allowed for postharvest application. Harvest needs to be done when the temperatures are not too high, to prevent the stress that the fruit suffer when we have to remove the field heat. It is fundamental the keeping of cold chain in the storage rooms and even more during transportation with dedicated devices with remote control, to prevent opportunistic behavior along the food supply chain. Packinghouses should apply the proper treatment to reduce the wounding of fruit and vegetables, reduce close to zero the inoculum load and allow the fresh produce to have a slow ripening, since a faster ripening is more exposed to the infection. All treatments that can prevent the loss of water and create a barrier to gas exchange can be beneficial, and in a good bunch of fruit the use of edible (or non edible) coating is becoming increasingly important. Use of smart systems to record the onset of infections is desirable, although in this

field is not easy to move from the research to the practical application. Another important issue is the cooperation of actors along the supply chain. Request of retailers of fruit and vegetables with a residue of pesticides even lower than legal threshold (maximum residue level) and of a number of residues very low (even 3 to 5) requires complex strategies in the field and in the packinghouse to reach such limits with risk to have a less protected produce, and increase of FLW. Last, but not least, an appropriate behavior of the consumer is important to contribute with own actions to FLW reduction.

SE09 C02

OVERVIEW OF POSTHARVEST DISEASE MANAGEMENT PRACTICES ON APPLE: PREVENTION, MONITORING AND CONTROL

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Postharvest apple diseases are primarily caused by fungal pathogens, as *Penicillium* spp. and *Botrytis* spp. that enter through wounds. Increasingly relevant latent pathogens, such as *Neofabraea alba* and *N. kienholzii* (bull's eye rot), *Ramularia mali* (dry lenticel rot), *Colletotrichum* spp. (bitter rot), and *Alternaria* spp. (black rot), develop in postharvest. White haze, which affects fruit quality, is linked to basidiomycetous genera such as *Entyloma*, *Golubevia*, and *Tilletiopsis*, with new species like *E. mali* and *G. mali* identified. Molecular diagnostic tools are used for pathogen detection and monitoring. Fruit microbiome analysis indicates that white haze agents are epiphytic and appear just before harvest, while *R. mali* is endophytic three months before harvest and becomes epiphytic during storage. *R. mali* and *N. alba* are found in orchard environments early in the season. Postharvest disease management strategies include the use of two *Aureobasidium pullulans* strains against white haze, which showed efficacy comparable to chemical treatments at the end of shelf-life. These strains proliferate well on treated fruit both epiphytically and endophytically. Biofumigation with thyme, savoury, and basil essential oils (EOs) at varying concentrations was tested against *Botrytis cinerea*, showing that thyme and savoury EOs are effective in preventing grey mould rot. Essential oils not only inhibit pathogen growth but also induce resistance in the fruit. Slow-release EO diffusers of basil, oregano, savoury, thyme, lemon, and fennel were tested against blue mould caused by *Penicillium expansum*, with lemon and oregano EOs showing the highest efficacy after 60 days of storage and an additional 14 days of shelf-life.



SE09 C03

RECENT ADVANCES IN ALTERNATIVE TREATMENTS INCLUDING BIOCONTROLS, BIOPESTICIDES, INORGANIC SALTS, AND PHYSICAL TECHNOLOGIES FOR MANAGING POSTHARVEST DECAYS OF FRUIT AND VEGETABLE CROPS

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Fruits and vegetables are among the most important crops in the world for providing dietary nutrition and health benefits for humans and animals. These crops also have a vital role in trade and the global economy. In the future, the global population and economic welfare of many countries are expected to increase the demand for fruits and vegetables. Climacteric and non-climacteric crops are nutrient-rich and thus, extremely perishable with estimated postharvest losses of often 10 to 40%. Extending the shelf-life of these crops is a crucial challenge in meeting the demand. Conventional preservation of fruits and vegetables has been based on integrating the use of resistant cultivars, cold temperature management, sanitation, and conventional pesticides. With consumer concerns on human health and the environment with the use of conventional pesticides especially after harvest, other strategies have emerged that include using antagonistic microorganisms, natural compounds, inorganic salts, and physical treatments. The management of decays with any of these approaches has been extensively investigated and has revealed the limits of any single approach. Thus, complementation of alternatives may provide more effective solutions that are commercial acceptable. New insights in endo- and epiphytic microbiomes of crops have the promise of developing of new biocontrols based on microbial consortia, possibly in a matrix of natural compounds including biopesticides or inorganic salts that are only inhibitory to the target pathogens and not the microbiome.

stranded DNA or RNA molecules that fold into unique three-dimensional structures, allowing them to bind specifically to a target molecule with high stability. In this work, two DNA aptamers against the SOD1 protein of *B. cinerea* (BcSOD1) were developed. BcSOD1 is involved in the virulence/pathogenicity of *B. cinerea* as it catalyses the dismutation of the superoxide-ion, produced as a host plant defence system. To test the effectiveness of both aptamers, sensitivity assays (effect on conidia germination, detached leaf, and fruit assays), fungal biomass analysis and aptamer uptake studies were carried out. The results showed that both aptamers were taken by the fungus and inhibited the conidia germination of *B. cinerea* by 60%. Furthermore, it was demonstrated that both were able to reduce *B. cinerea* growth and fungal biomass by 50% and 60%, respectively, on tomato leaves and apple fruit. These results demonstrate the potential, for the first time in agriculture, of DNA aptamers to be novel candidates that could be included within the different strategies to control the gray mold disease.

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SE09 001

DNA-APTAMERS AS A NOVEL STRATEGY IN AGRICULTURE TO CONTROL THE GRAY MOLD DISEASE CAUSED BY BOTRYTIS CINEREA

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Botrytis cinerea, the causal agent of the gray mold disease, is one of the main limiting factors of horticultural crops production worldwide. Its control is very dependent on the use of fungicides; however, this fungus has been categorized by FRAC (*Fungicide Resistance Action Committee*) as a high-risk pathogen for fungicide resistance development. In addition, and according to the "farm to fork" strategy of the European Green Deal, the diversity of fungicides available to growers will be reduced by 50% in 2030. For all these reasons, new advances and technologies are needed to control this important harvest and postharvest disease. Aptamers, also called chemical antibodies, are small synthetic single-



BANQUET

17.00-18.00 Concurrent Session **68**

Advances with Biopesticides to meet Plant Protection Challenges

Chair: Trevor Jackson (AgResearch, New Zealand)

Laura Villamizar (Microbial Solutions, AgResearch Ltd., New Zealand)

SE06 009

IMPACTS OF α -SOLANINE AND α -CHACONINE ON KEY PLANT PATHOGENS AND BENEFICIAL ORGANISMS

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α -solanine and α -chaconine are glycoalkaloids found in varying concentrations in potato tissue and are important for plant defense. As previous studies have shown, both compounds have inhibitory effects on microbes and insects, indicating their promising potential as plant protection agents. The aim of this research is to investigate the effects of glycoalkaloids on economically important plant pathogens and beneficial organisms. Therefore, various fungi, bacteria and nematodes were exposed to media enriched with glycoalkaloids and their development, parasitism and survival were subsequently evaluated. The results indicate a clear effect of the compounds on the organisms studied. Both glycoalkaloids inhibited mycelial growth of the fungi *Fusarium graminearum* and *Rhizoctonia solani*, with α -chaconine showing consistent efficacy at a four times lower concentration than α -solanine. Similar results were also obtained for *Leptosphaeria maculans*. Here, 125 μ g/mL α -chaconine significantly reduced colony growth by 69 % compared to the controls. The infection rate of Arabidopsis plants by the plant-parasitic nematode *Heterodera schachtii* was significantly reduced by 67% when exposed to α -chaconine (10 μ g/mL). The number of cysts formed, but not the number of eggs contained therein, was also reduced. The growth of certain beneficial organisms was also significantly affected by both glycoalkaloids, while others were not sensitive. For example, the growth of the beneficial bacterium *Bacillus subtilis* was significantly reduced by α -solanine (31.25 μ g/mL) and α -chaconine (1.96 μ g/mL) by 14 % and 12 %, respectively. The results illustrate the different concentration-dependent efficacy of the individual glycoalkaloids, with α -chaconine generally having a stronger inhibitory effect on the growth of both pathogens and beneficial organisms.

SE06 010

BIOPESTICIDES AS TOOLS FOR GREEN PLANT PROTECTION IN MAIN CROPS OF EUROPE

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Biopesticides are not designated as a specified category in the EU legislation (Reg. EC 1107/2009) regarding placing on the market plant protection products. However, they have surged as promising alternative tools to synthetic pesticides towards green plant protection in the frame of the Farm to Fork strategy and the new Common Agricultural Policy. Biopesticides include microorganisms, semiochemicals, botanicals and other substances many of which are considered as "low risk" to humans and the environment. Today more than 40 active substances (a.s.) are authorized in the EU as bioinsecticides and more than 50 are biofungicides, while bioherbicides are limited. Herein we attempt a review on the registered biopesticides for plant protection in main crops in the EU such as apples, grapevine and vegetables. Bioinsecticides include a.s. of microbial origin such as bacteria (e.g., strains of *Bacillus thuringiensis*) and pheromones to control mainly Lepidoptera and Diptera pests; fungi (e.g., strains of *Beauveria bassiana*, *Akanthomyces muscarius*, *Isarea fumosorosea*, *Metarhizium anisopliae* var. *anisopliae*) against soft-bodied insects or mites; viruses (e.g., *Cydia pomonella* granulosus virus); substances of natural origin (e.g., botanical extracts/oils, azadirachtin) against many insects and mites. Biofungicides are mostly microbials belonging to the genera *Bacillus* and *Trichoderma* as well as botanicals (e.g., eugenol, geraniol, thymol, spearmint oil, clove oil), and other substances (e.g., laminarin, cerevisane) with various modes of action. They can be employed against plant pathogenic fungi and oomycetes (*Botrytis cinerea*, *Erysiphe* spp., *Sphaerotheca* spp., *Monilinia* spp., *Phytophthora* spp., *Fusarium* spp., etc.). Bioherbicides mainly include botanical substances such as pelargonic acid, lauric acid, capric and caprylic acid and oleic acid which control grasses and broadleaved weeds. The toolbox extends to available basic substances. Finally, the challenges and perspectives in plant protection of these crops are discussed in the light of biopesticide a.s. in the pipeline of evaluation for authorization.



SE06 011

CONTROL OF POTATO LATE BLIGHT BY EXPLOITING THE ANTIMICROBIAL AND BIOSTIMULANT PROPERTIES OF EXO-METABOLITES PRODUCED BY TRAMETES VERSICOLOR

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Potato late blight is a devastating disease affecting cultivated potato caused by the oomycete *Phytophthora infestans*. The use of biocontrol agents and plant resistance inducers are sustainable and alternative ways to fungicides to contrast the disease even at field level. In this study we tested the effect of the cultural filtrate (CF), derived from the growth in liquid culture of the basidiomycete *Trametes versicolor*, against potato late blight. The CF is rich of bioactive compounds with antimicrobial activities and capable to stimulate antioxidant system and promote plant defense. The CF can be used as crude culture filtrate (CCF), or by separating the components (i.e., polysaccharides, proteins, lipids). *In vitro* assays were developed to understand the effect of CCF and a protein fraction, precipitated with ammonium sulfate, on *P. infestans*. To investigate the antimicrobial capacity of CCF, 3-week-old plants were sprayed with CCF at different concentrations (0.25%-0.5%-1% w/v), detached leaves were infected with *P. infestans*, and the lesions were evaluated by trypan blue staining. To investigate the biostimulant capacity, CCF (0.25% w/v) was applied on potato plants by soil drench, and defense-hormones were evaluated by LC-MS/MS. CCF showed an antimicrobial capacity *in vitro*, by inhibiting sporangia germination and mycelium development, and the protein fraction seems to be the responsible for this activity. The same effect was noted in potato leaves sprayed with CCF, in which the treatment reduced the late blight severity about 40% respect to the control. For the biostimulant capacity, the plants irrigated with CCF showed an increase in the synthesis of the defense hormone (salicylic acid), suggesting the potential application to stimulate resistance towards *P. infestans*. In conclusion, the tests performed could be used to select the best strategy for applying CCF, or the single fractions, before attempting field trials.

SE06 012

ε-POLY-L-LYSINE, A FOOD-APPROVED PRESERVATIVE, MIXED WITH CINNAMALDEHYDE AS A POTENTIAL NEW AGRICULTURAL BACTERICIDE

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Lack of effective antibacterial treatments, limitation of copper usage, and increasing regulatory restrictions on the use of agricultural antibiotics stimulated our evaluations of new treatments to manage bacterial disease of plants. The antibacterial ε-poly-L-lysine (EPL) is a hydrophilic cationic linear homo-poly-amino acid typically composed of 25 to 35 identical L-lysine residues and was found to destabilize bacterial membranes. It is produced by fermentation of *Streptomyces albulus* and is used as a natural preservative of food products including soft drinks, cheese, fish, and salad dressings in Japan, the United States, South Korea, China,

and other countries. We conducted field studies with EPL for managing major bacterial diseases of tree crops over several years. Using a rate of 1000 mg/liter, efficacy against fire blight of pear caused by *Erwinia amylovora* or walnut blight caused by *Xanthomonas arboricola* pv. *juglandis* was variable, but sometimes similar to the industry standards streptomycin or copper-mancozeb, respectively. To improve the efficacy and consistency, mixture partners for EPL were initially evaluated in laboratory studies. In agar dilution tests, growth of *E. amylovora* and *X. arboricola* pv. *juglandis* was not or only slightly inhibited in the presence of 500 mg/liter EPL or 100 mg/liter cinnamaldehyde (an organically-approved, broad spectrum biopesticide). The mixture of the two, however, completely inhibited growth. In first-year field studies on the two diseases, this mixture was highly effective and therefore, can possibly be an efficacious natural product treatment with high potential for organic registration. We will continue to include EPL-cinnamaldehyde in future field studies using an agricultural formulation provided by a registrant.

SE06 013

THE DYNAMIC CHANGES IN OLIVE FRUIT PHENOLIC METABOLISM AND ITS CONTRIBUTION TO THE ACTIVATION OF QUIESCENT COLLETOTRICHUM INFECTION

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Universidad de Córdoba, Rabanales, Cordoba, Spain

Olive anthracnose disease, caused by *Colletotrichum* species, poses a significant threat to olive fruit production worldwide. Despite the innate immunity of developing olive fruits to the pathogen, susceptibility increases with ripening, albeit with variations among cultivars. This study investigates the phenolic metabolic pathways influencing resistance to *Colletotrichum* in olive fruits at different ripening stages. By analyzing two resistant ('Empeltre' and 'Frantoio') and two susceptible ('Hojiblanca' and 'Picudo') cultivars, we elucidate distinct phenolic profiles correlating with resistance levels. Resistant cultivars exhibit heightened synthesis of aldehydic and demethylated phenolic forms, which strongly inhibit fungal spore germination. Conversely, susceptible cultivars favor the production of hydroxytyrosol 4-O-glucoside, lacking antifungal properties. These findings underscore the pivotal role of phenolic composition in determining fruit resistance. Furthermore, this research highlights the significance of fruit ripening stages in assessing cultivar susceptibility. Notably, the turning stage emerges as crucial for discerning resistance levels and their association with phenolic profiles. Susceptible cultivars undergo rapid degradation of secoiridoids during ripening, contrasting with the sustained dominance of these compounds in resistant cultivars. Moreover, bioassays reveal the fungicidal efficacy of secoiridoid compounds, including oleuropein and oleocanthal, underscoring their importance in conferring resistance. Concentrations of total phenols above a threshold of 35 mg/g render green fruits resistant to pathogen colonization, emphasizing the role of phenolic content as a determinant of resistance. In conclusion, this study provides valuable insights into the interplay between phenolic metabolism and olive fruit resistance to anthracnose. Understanding these mechanisms is pivotal for developing strategies to mitigate disease incidence and enhance olive

Concurrent Sessions

Tuesday, 2 July

production sustainability. Figures:



Figure 1. Olive fruit rot caused by *Colletotrichum godetiae* in field

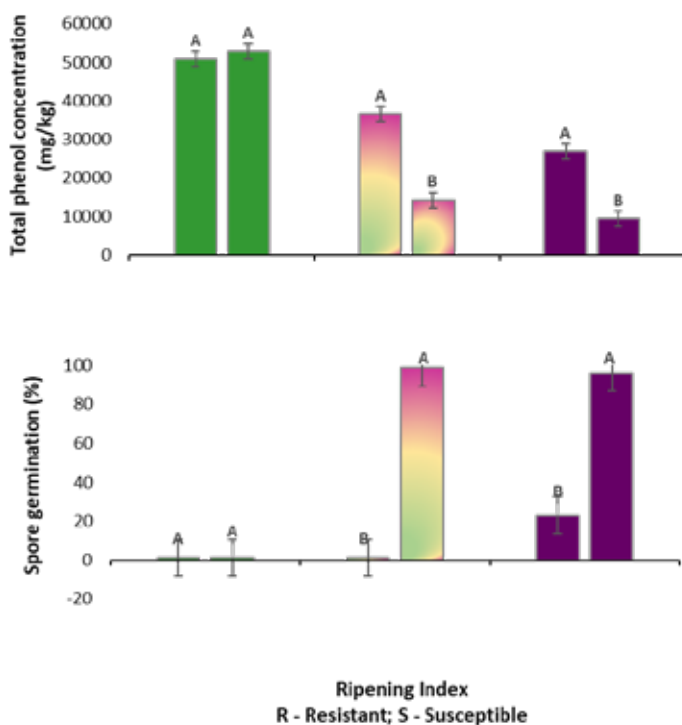


Fig. 2. Relation between total phenol concentration in olive fruits and percentage of spore germination of *Colletotrichum godetiae*. A. Mean total concentration of phenols in two resistant ('Empeltre' and 'Frantoio') and two susceptible ('Hojiblanca' and 'Picudo') cultivars. B. Percentage of spore germination in the presence of phenolic fruit extract during fruit maturation (green, turning, and ripe fruits) in the presence of phenolic fruit extracts. Means with the same letter are not significantly different according to the High Significant Different Test at $p \leq 0.05$.



Fig. 3. Pearson correlation analysis ($r = -1$ to 1) between the concentration of individual phenols in extracts from olive fruit at three ripening stages (green, turning, and ripe fruits) and the inhibiting effect in *Colletotrichum godetiae* spore germination. The correlations were based on the phenolic concentrations of two resistant ('Empeltre' and 'Frantoio') and two susceptible ('Hojiblanca' and 'Picudo') cultivars. The spore germination was evaluated with the total phenolic extract obtained from fruits at the three ripening stages. Significant Pearson correlations ($*p \leq 0.05$, $**p \leq 0.01$, and $***p \leq 0.001$).

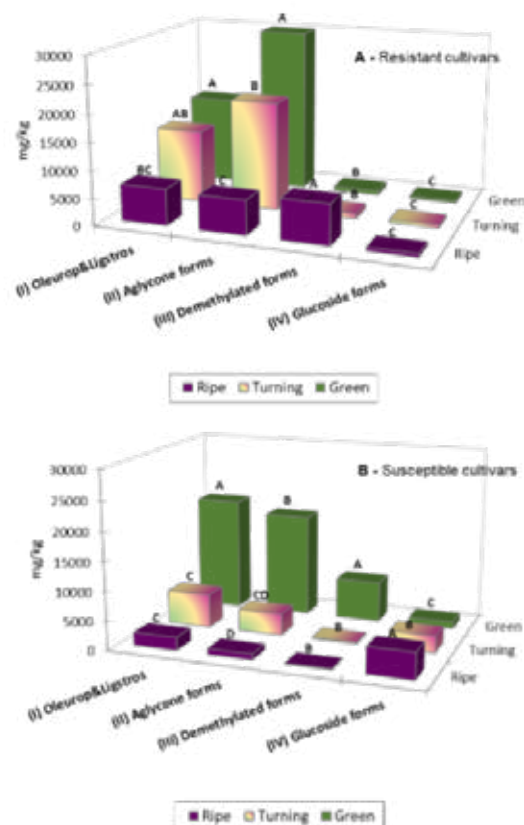


Fig. 4. Concentration of main phenols (mg/kg) at the three olive fruit ripening stages (green, turning, and ripe fruits). All phenolic concentration were estimated as a relative concentration to oleuropein standard. The fruit phenols are grouped according to the biotransformation process: I - Oleuropein and ligustrin, II - Oleuropein and ligustrin aglycone, III - Demethyloliguropein, oleocanthal and oleacein, IV - Hydroxytyrosol-4-O-glucoside and oleuropein glucoside. A - resistant to *Colletotrichum* cultivars ('Empeltre' and 'Frantoio') and B - susceptible cultivars ('Hojiblanca' and 'Picudo'). Means with the same letter are not significantly different according to the High Significant Different Test at $p \leq 0.05$.

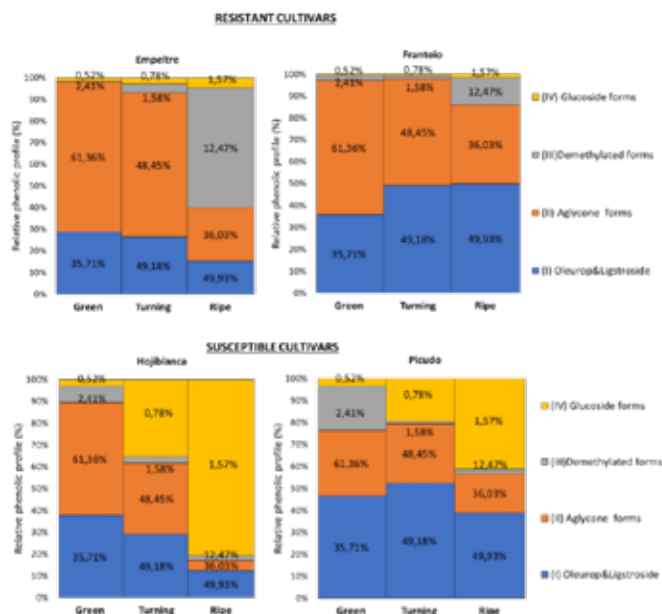


Fig. 5. Relative phenolic content expressed in percentage during olive fruit maturation (green, turning, and ripe fruits) in two resistant ('Empeltre' and 'Frantoio') and two susceptible ('Hojiblanca' and 'Picudo') cultivars. This include: I - Oleuropein and Ligstroside, II - Oleuropein and Aglycone Ligstroside, III - Demethylated forms, IV - Glucoside forms.

(cv. Kalamon), and the effectiveness of each treatment was assessed based on disease severity reduction and inhibition of conidial production. Molecular characterization results showed that the *C. acutatum* species complex is dominant in Greece. Pyraclostrobin and difenoconazole demonstrated high efficacy against all tested *C. acutatum* strains, while increased resistance levels were observed in trifloxystrobin, azoxystrobin, kresoxim-methyl, and dodine. The biological PPPs Serenade Aso®, Vacciplant®, and Mevalone®, along with the copper-based fungicides Nordox® and Cuprofix Dispers®®, as well as the synthetic PPPs Priors top®, Score®, Insignia®, and Ortiva top®, exhibited superior efficacy across all experiments. Regarding the sensitivity assays mentioned above. In conclusion, this research provides valuable insights for developing an integrated management system (IPM) for olive anthracnose disease.

Acknowledgments: The research is being funded under Action 2 "Implementation of the Operational Plan (project) of the ESK Operational Groups for the productivity and sustainability of agriculture" of Sub-measure 16.1-16.2 of Measure 16 of the AGRICULTURAL DEVELOPMENT PROGRAM 2014 - 2020 "Cooperation"



SE06 014

INTEGRATED MANAGEMENT SYSTEM FOR OLIVE ANTHRACNOSE

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Olive anthracnose (OA), caused by fungi belonging to the *Colletotrichum* genus, particularly *Colletotrichum gloeosporioides* and *Colletotrichum acutatum* species complexes, stands as a major global threat to olive drupes, leading to significant quantitative and qualitative deterioration of harvested products. Conventional chemical plant protection products (PPPs) have been widely used to manage the disease; however, their use poses risks to both the environment and human health while also potentially fostering pathogen resistance. This study aims to identify the *Colletotrichum* species associated with OA in Greece, investigate the sensitivity of *C. acutatum* strains to several active substances, and assess the efficacy of selected commercial biological and chemical plant protection products against OA. As part of the above objective, 100 *Colletotrichum* spp. strains were isolated and molecularly characterized, while some of them were evaluated for their sensitivity/resistance to six different active substances, assessing the inhibitory effect on mycelial growth and conidial germination. Subsequently, seven commercial, five copper-based, and nine biological PPPs approved for olive cultivation in Greece were evaluated. In planta experiments were conducted on detached olives



SKALKOTAS

17.00-18.00 Concurrent Session **7B**

Understanding Pathogen-Vector-Host Interactions in Globally Important Pathosystems

Chair: **Shaonpius Mondal** (University of Nebraska - Lincoln, USA)
Elizabeth Jeannette Cieniewicz (Clemson University, USA)

SE07 C07

THE MOLECULAR INTERPLAY BETWEEN PLANT VIRUSES AND INSECT VECTORS

Anna E. Whitfield

Department of Entomology and Plant Pathology, Emerging Plant Disease and Global Food Security Cluster Director, North Carolina State University

Plant viruses are a significant threat to global food security, and the majority of viruses that infect plants are transmitted from one host to another by arthropod vectors. Our research aims to understand the molecular and ecological interactions between viruses, vectors, and plants that result in virus transmission. Recent technological advances are breaking new ground in the study of negative-strand RNA viruses and their transmission by arthropod vectors. This talk will focus on the molecular mechanisms of vector transmission of plant rhabdoviruses and bunyaviruses. Due to the specificity of virus transmission by vectors, there are distinct steps in the transmission process that represent good targets for strategies to disrupt the disease cycle. A specific viral protein(s) is required for attachment and/or entry into the insect vector. We have used viral attachment proteins to reduce virus acquisition and to identify and insect host factors for virus infection of vectors. Transcriptome, proteome, and genome resources for thrips and planthopper vectors has enabled identification of insect proteins that interact with and respond to plant viruses. The long-term goal of this work is to develop effective and safe strategies for virus and vector control.

exhibited genetic variability predominantly in the ORF5/P5 region as well as the presence of tomato chlorosis virus (ToCV) in few pepper greenhouses. Aphid transmission parameters and host range of PeVYV-6 were studied in order to elucidate the factors affecting its prevalence in the field. Similarly to the other PYD-associated poleroviruses, *Aphis gossypii* was found to be a more efficient vector of PeVYV-6 even though several other aphid species were also identified as vectors. Pepper plants exhibiting PYD-like symptoms but with variable intensity were also reported in the Rhodes Island in Greece. Nevertheless, no polerovirus was identified. The application of HTS revealed the presence of two fungi-transmitted ophiioviruses, namely ranunculus white mottle virus (RWMV) and lettuce ring necrosis virus (LRNV). The *Olpidium* species, vector of the two viruses was identified and transmission trials were performed which unveiled its implication in the natural dispersion of RWMV and LRNV in the field. Taken together, our results indicate that although PeVYV-6 is the key-pathogen in PYD etiology in Greece other viral species from different genera with distinct transmission modes and epidemiology might also contribute to its development.

The research project was supported by the Hellenic Foundation for Research and Innovation (H.F.R.I.) under the 1st call for H.F.R.I. Research Projects to support Faculty Members & Researchers and the Procurement of high-cost research equipment grant (Project Number: 3719).

SE07 C09

EXPLORING THE SRBSDV-SOGATELLA FURCIFERA-RICE PLANT INTERACTIONS FOR REDUCTION OF VIRUS TRANSMISSION

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State Key Laboratory for Biology of Plant Disease and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing 100193, PR China

The Southern rice black-streaked dwarf virus (SRBSDV), transmitted horizontally and persistently by *Sogatella furcifera*, reduces the yield and quality of rice crops. Thus far, the effective way to manage SRBSDV viral disease is through manipulation of vector's virus transmission. Studies were conducted to evaluate the potential of silicon amendment, sublethal concentrations of pymetrozine, and rice varietal resistance to *S. furcifera* in reducing SRBSDV transmission. Virus transmission (both inoculation and acquisition) was measured against rice plants either amended with silicon, or treated with LC₁₀ and LC₅₀ pymetrozine, or resistant to *S. furcifera* in contrast to the controls. Vector preference, feeding amount, and probing behaviors were assessed using olfactometer, bromocresol green filter paper method, and electrical penetration graph. *S. furcifera* females showed non-

SE07 C08

ELUCIDATING THE ETIOLOGY AND EPIDEMIOLOGY OF PEPPER YELLOWS DISEASE

Vasileia Gavrilis, Nefeli Vasileiou, Maria Konsta, Leonidas Lotos, Nikolaos Katis, Varvara Maliogka

Aristotle University Of Thessaloniki, School of Agriculture, Plant Pathology Laboratory, Greece

Pepper yellows disease (PYD) affects pepper crops worldwide and it has been associated with a group of phylogenetically related poleroviruses which share key genetic traits but differ in their distribution and epidemiological parameters. PYD has been endemic in pepper crops in Crete (Greece) for more than 15 years and pepper vein yellows virus-6 (PeVYV-6) has been associated with it. The occasional identification of pepper plants in Crete exhibiting typical disease symptoms in which no polerovirus was identified has led to a deeper investigation of the etiology of the disease through the application of high throughput sequencing (HTS) analysis. HTS results revealed the presence of different variants of PeVYV-6, some of which



preference for and SRBSDV transmission was significantly reduced by 25%-66% in silicon-amended plants, which may be explained by the intensified silicification in the plants, the elongated non-probing duration and shortened phloem sap ingestion duration in the *S. furcifera* feeding on the plants. When plants were treated with sublethal pymetrozine, both SRBSDV transmission and vector feeding amount decreased significantly, total durations of pathway probing events were elongated whereas those of salivation and phloem-related activities were shortened. Among the varieties showing varying resistance, *S. furcifera* showed non-preference for and low feeding on the resistant plants. Both inoculation and acquisition rates were lower on resistant plants, which may result from the short durations of salivation and phloem-related activities and the low feeding amount of the vector on the resistant plants. A significant negative correlation between varietal vector resistance and SRBSDV transmission was observed. The results show that Si amendment, sublethal concentrations of pymetrozine, and rice varietal resistance to *S. furcifera* reduce SRBSDV transmission, thus these measures can be incorporated in the management protocol of the viral disease.

MC2

17.00-18.00 Concurrent Session 10A

Impact and Control of Transboundary / Invasive Pests

Chair: Moriyama Hiromitsu (Tokyo University of Agriculture and Technology, Japan)

Arie Tsutomu (Tokyo University of Agriculture and Technology, Japan)

SE10 C01

POTENTIALITIES OF MYCOVIRUSES INFECTING LOWER EUKARYOTIC ORGANISMS, PLANTS, AND INSECTS AS BIOCONTROL AGENTS

Hiromitsu Moriyama

Laboratory of Molecular and Cellular Biology, Graduate School of Agriculture, Tokyo University of Agriculture and Technology, Japan

Mycoviruses were first found in mushrooms, *Agaricus bisporus*, *Penicillium chrysogenum*, and brewer yeast killer strains, *Saccharomyces cerevisiae*, in the 1960s. A common characteristic of these earlier discovered mycoviruses is that their hosts are regularly cultivated or cultured by sophisticated and skilled technologies within well-organized fermentation process plants. After the 1970s, many mycoviruses were found among plant pathogens isolated in agricultural fields to yield crops, vegetables, and orchard fruits, supplied with agrochemical products to control plant diseases. Several mycovirus-infected phytopathogenic fungi exhibit hypovirulence traits that weaken the virulence of the host fungus. The hypovirus that infects the chestnut blight fungus (*Cryphonectria parasitica*) and attenuates the host fungus has established a leading position as a model for mycovirus research. On the other hand, there have also been several reports on mycoviruses that cause hypervirulence in host fungi. The genomes of these mycoviruses are either double-stranded RNA or single-stranded RNA, and no mycovirus whose genome is double-stranded DNA has been found. It is symmetrical compared to bacterial phages whose genomes are exclusively dsDNA but rarely RNA genomes. These mycovirus-related viruses have often been found as symptomless viruses in crops, vegetables, fruits, and livestock grasses. Among them, endornavirus is most dominant and well-known as an indigenous or endogenous high-molecular RNA replicon; however, a few knowledge are reported for their functions. Several mycovirus-related viruses have also been found in insects, such as mosquitoes, ants, and moths, associated with the phenomenon of late male killing that leads to female-biased sex ratios in *Homona magnanima*. Although mycoviruses propagate mainly to lower eukaryotes, they also exist endogenously in crops and insects, as if they were using these higher organisms as vectors. I introduce several examples of utilizing the mycoviruses as biocontrol agents and new insights into mycoviruses and related viruses as causal agents for biodiversity.

SE10 C02

CHALLENGES AND PROSPECTS FOR CONTROL OF CITRUS GREENING DISEASE IN JAPAN

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Huanglongbing (HLB), also known as Citrus greening disease, is caused by the pathogenic bacterium *Candidatus Liberibacter asiaticus* (CLas) and is transmitted by the Asian citrus psyllid (ACP, *Diaphorina citri* Kuwayama). In Japan, HLB has gradually expanded its distribution, reaching as far north as the entire Okinawa Prefecture and the Amami region of Kagoshima Prefecture. To push the current northern limit of the disease further south, our goal is to identify both the existing and potential risks of HLB occurrence. We attempted to implement four key tasks to achieve our goals: (1) estimating the areas suitable for ACP colonization in Japan, (2) developing a survey technique for the distribution of the host plant for ACP, (3) the orange jasmine *Murraya paniculata*, creating a novel detection method for ACP, and (4) enhancing the sensitivity of detecting low-concentration CLas. We visualized the hotspots of HLB occurrence by establishing the relationship between weather, geographical conditions, and potential settlement areas of ACP. This enabled us to estimate regions where HLB infestation could occur, even in areas where the disease has not yet observed. Furthermore, a dataset comprising drone-captured images targeting orange jasmine provided potential insights into the distribution of orange jasmine vegetation in HLB-free regions. Monitoring of ACP distributions was conducted by detecting ACPs and their symbiont DNA through environmental DNA, in addition to identifying the latent status of CLas with BioPCR. These combined efforts significantly contributed to our understanding of the risks associated with ACP and CLas transmission. In the future, our aim is to visualize the potential risk of HLB outbreaks and ACP invasion. This study will enable us to propose new strategies for urgently preventing and eradicating HLB in Japan.



SE10 C03

DEVELOPMENT OF MANAGEMENT TECHNIQUES AGAINST SUGARCANE WHITE LEAF DISEASE BASED ON EPIDEMIOLOGICAL RESEARCH AND COMPUTER SIMULATION

Youichi Kobori

Japan International Research Center for Agricultural Sciences, Japan

Sugarcane white leaf disease (SCWLD) is an insect-borne disease caused by a phytoplasma pathogen, and infected sugarcane dies after leaf chlorosis. SCWLD is spread by two leafhopper species, *Matsumuratettix hiroglyphicus* and *Yamatotettix flavovittatus*, and the planting of infected seedcane. The disease is considered to have the most severe impact on sugarcane production in Thailand, which is the second-largest exporter of sugar in the world. In addition, in the 2000s, many cases of the disease were reported in Southeast Asia. Therefore, this study aimed to develop a rational management technology against SCWLD based on epidemiological surveys and computer simulation results. Spatial distribution analysis of SCWLD and epidemiological analysis of spreading factors of SCWLD were conducted in sugarcane fields in northeastern Thailand, one of the major sugarcane production areas. In addition, an individual-based model was developed based on the ecology of the vectors and pathogen to simulate SCWLD spreading. The rational control methods were examined based on the simulation results. Field survey results showed that SCWLD-infected seedcane were probably planted on common farmer fields, and the use of healthy seedcane seems effective in reducing the spread of SCWLD. Moreover, it was difficult to propagate healthy seedcane in the spread area owing to frequent vector transmission. In addition, multi-generation propagation is required to produce sufficient quantities of healthy seedcane because sugarcane is a plant species grown through vegetative propagation and has low intergenerational propagation rate. Simulation results suggested that large-scale propagation of healthy seedcane coupled with pesticide treatment may enable low-risk seedling production. Based on the results of the study, we developed a "Healthy seedcane propagation and distribution manual against SCWLD".

laevis oocytes with the aid of cofactors. The responses to ligands of the expressed nAChRs were measured by voltage clamp electrophysiology. The contribution of each subunit to the neonicotinoid actions was analyzed by multivariate analyses. We found that five nAChR subunits coexisted in *Drosophila melanogaster*. Hence, 12 kinds of nAChRs formed by the subunits were expressed in *X. laevis* oocytes and the agonist activity of neonicotinoids was measured in terms of affinity and efficacy. Multivariate analyses indicated that the Da2 subunit had a negative impact on the agonist activity, and indeed, RNAi of the Da2 subunit gene expression resulted in hypersensitivity to neonicotinoids in adult fruit flies. It is suggested that the same thing will happen in the invasive species.

SE10 C04

TOWARD CONTROL INVASIVE INSECT SPECIES BY TARGETING NICOTINIC ACETYLCHOLINE RECEPTORS

Kazuhiko Matsuda

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Globalism has been accelerated, and non-native pest insect species can invade at higher frequencies than ever. We must be prepared for such an invasion with control measures and learn the resistance mechanisms as well as the mode of action of the control measures. Neonicotinoids are insecticides targeting nicotinic acetylcholine receptors (nAChRs). With good systemicity in plants, neonicotinoids enjoy 25% of the sales. Hence, it is crucial to understand their mechanism of action at nAChRs in detail yet it is largely unknown how they act on each nAChR subtype in insects. Here, the author addresses this question and perspective on invasive species control. *Drosophila melanogaster* nAChRs were expressed in *Xenopus*



BANQUET

18.30-21.00 Concurrent Session 11

Canker Disease of Fruit, Nut and Vine Crops

Chair: Themis Michailidis (UC Davis, USA)

SE11 C01

CANKER DISEASES OF NUT CROPS CAUSED BY BOTRYOSPHAERIACEAE IN CALIFORNIA

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The three major nut crops, almond, pistachio, and walnut, grown in approximately 0.92 million ha in California, are susceptible to canker diseases that can cause significant yield losses. The disease of almond is expressed as band canker, the pistachio as Botryosphaeria (Bot) panicle and shoot blight, and the walnut as Bot canker and blight caused by at least eight, ten, and ten species of Botryosphaeriaceae, respectively. The band canker attacks the trunk of young trees, causing band-shape cankers. Such cankers initiate from growth cracks or bark lenticels. Although initially this disease showed a diffusion spread pattern from the source of inoculum, in the last 7 years a uniform spread pattern occurred in the 1st to 3rd yr-old orchards. A qPCR assay for symptomless shoots of mother trees, nursery trees, and shoots from young orchards revealed the presence of latent canker-causing fungi, among which a few of the Botryosphaeriaceae fungi. The disease can kill young almond trees. Treating young trees with thiophanate methyl reduced the canker incidence. The Bot panicle and shoot blight can attack any green tissue of pistachio but does not kill mature trees. It can cause panicle (cluster) blight either from bud infections or from latent infections of nuts in the spring. The infection moves into the rachis of cluster and from there into last year's shoot causing cankers. The Bot canker and blight of walnut is very similar to the Bot canker of pistachio, i.e., infection and blight of the fruit move via the peduncle into the spur and from the spur into the shoot causing cankers. In pistachio, only pycnidia were found whereas in almond and walnut both pycnidia and pseudothecia can occur. Fungicidal control of Bot canker of pistachio and walnut is done with the use of carboxamide, strobilurin, and pre-mixtures of these fungicide groups. Detailed information on fungicidal control of these diseases can be found in: https://ipm.ucanr.edu/legacy_assets/pdf/pmg/fungicideefficacytiming.pdf.

major threats to the cherry industry in California. Main fungal canker diseases include *Calosphaeria canker*, *Cytospora canker* and *Eutypa dieback*, caused by *Calosphaeria pulchella*, *Eutypa lata* and *Cytospora sorbicola*, respectively. Managing canker diseases has been challenging for growers as no strategy alone suffices to control these diseases. Accordingly, the goal of this study was to improve the management of canker diseases affecting cherry by gaining new knowledge about disease biology. We investigated optimal temperatures for spore germination and colony growth of *C. pulchella*, *C. sorbicola*, and *E. lata* and compared the seasonal susceptibility of sweet cherry pruning wounds to these pathogens following early-summer and winter pruning. Overall, these studies indicated that *C. pulchella* favors warm temperatures (30°C) for spore germination and that pruning wounds made during winter were not to only poorly susceptible to infection by *C. pulchella* compared to pruning wounds made during summer. We also investigated the possibility that leaf scars, bud scars, and wounds resulting from fruit picking, could serve as infection courts for canker pathogens and performed artificial inoculations of these various tissues in the field. Results indicated that fruit picking wounds can serve as important infection courts for *C. pulchella*, *C. sorbicola*, and *E. lata*. Leaf and bud scars also were infection sites for *C. sorbicola*, although infection rates were relatively low. Finally, our laboratory evaluated the efficacy of different compounds to protect pruning wounds from infection by canker pathogens. Of the different fungicidal compounds tested, Thiophanate-methyl and Azoxystrobin + Propiconazole performed best against *C. sorbicola* and *E. lata*, allowing significant disease control. Biological, Trichoderma-based products provided significant protection of pruning wounds against all canker pathogens, and performed best at reducing infection by *C. pulchella*.

SE11 C03

FUNGAL PATHOGENS CAUSING TRUNK DISEASES IN NORTHERN ITALY, SPECIES DIVERSITY AND TEMPERATURE INTERACTION

Vladimiro Guarnaccia

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A broad range of collateral effects of intensification, diversification, and globalization have an impact on agriculture. The risk of disease dissemination via trade and contaminated propagation material is increasing. Moreover, climate change is affecting plant health by modifying the interactions among host plants, pathogens and the environment. Production of fruit, nut, and grapevine crops is

SE11 C02

EPIDEMIOLOGY AND MANAGEMENT OF FUNGAL CANKER DISEASES OF SWEET CHERRY IN CALIFORNIA

Florent P. Trouillas

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Fungal canker diseases of sweet cherry can be devastating to an orchard's productivity and longevity and thus constitute

increasingly threatened by fungal trunk diseases (FTD), and the consequent losses represent a major problem. Surveys were conducted in orchards located in different areas of Northern and Southern Italy to assess the fungal species diversity associated with FTD. Stem blight and dieback were observed as the most common symptoms with high incidence, and on several crops such as apple, citrus, berry fruit, hazelnut and grapevine. Different fungal pathogens such as *Botryosphaeriaceae*, *Diaporthaceae*, *Diatrypidae*, *Didymosphaeriaceae*, *Glomerellaceae*, *Nectriaceae*, *Phaeomoniellaceae*, *Sporocadaceae*, *Togniniaceae*, *Valsaceae* were found associated with the investigated affected plants. The fungal isolates obtained were identified at species level through multi-locus typing and morphological characters. Molecular analyses included different genomic regions selected on the base of the most recent fungal taxonomy studies. Pathogenicity tests were conducted with the aim to explore the virulence of the identified fungal species on the respective host. Effect of temperature on colony growth was assessed through in vitro tests. Moreover, the role of temperature on the severity of blueberry stem blight and dieback caused by *Neofusicoccum parvum* was explored by pathogen inoculation on plant cultivated at different conditions. The presented studies increase understanding of the fungi associated with FTD of several major crops, providing etiological and epidemiological knowledge, useful to develop sustainable disease control strategies.

SE11 C04

FUNGAL TRUNK AND CANKER DISEASES OF FRUIT, NUT AND VINE CROPS IN SPAIN: CAUSES AND SOLUTIONS

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Currently, the production of grapevines, and other fruit and nut crops is threatened by the increasing incidence of fungal trunk and canker diseases, which cause significant crop reductions and serious economic losses. Numerous fungal species, belonging to various taxonomic groups, can infect the woody tissues of these crops, mainly through wounds, both in nurseries and in the field, causing cankers, internal wood necrosis, and general symptoms of decay and death of branches. Severe attacks can lead to the death of the entire plant. In this presentation, the current situation of this problem in Spain is reviewed, which in addition to grapevine, has already been described and negatively impacted in other important crops such as almond, avocado, pistachio, olive trees, sweet cherry, and walnut. The causes that determine the increase in the severity of these diseases will be discussed, including the diversity of fungal phytopathogenic species and the cultural practices both in vineyard and field conditions. Among the control strategies, current research of those based on a better knowledge of the epidemiology of the pathogens and the search for alternatives to chemical control will be highlighted.

SE11 C05

STATUS OF CITRUS CANKER DISEASE IN SICILY

Giorgio Gusella, Greta La Quatra, Giuseppa Rosaria Leonardi, Dalia Aiello, Giancarlo Polizzi

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Citrus represents an iconic, very relevant crop for the Italian agriculture, especially for the territories of southern Italy, where its cultivation, particularly in Sicily, is almost predominant among the other fruit crops. This presentation aims to overview the status of citrus canker disease in Sicily. Citrus canker disease is characterized by the presence of abundant gummosis exuding from wounds and bark cracks mostly concentrated in the trunk and along the main branches of the trees. Under the bark is possible to observe severe wood discolouration. This disease was investigated many years ago in Italy and it was named as “*Dothoriella gummosis*”, “*Phomopsis gummosis*” or also “*Diplodia gummosis*”. At the time of the first investigations, the pathogens associated with the disease were identified as *Diplodia natalensis*, *Dothoriella ribis*, and *Phomopsis citri* but only a morphological approach was used for pathogens characterization. Taxonomy of fungi rapidly increased in DNA era and identification approaches based on multi-locus phylogeny rapidly revealed the hidden and huge diversity of fungal species. Canker diseases have been deeply investigated in the recent years and their etiology is appearing more complex than previously thought. Our field surveys have been conducted in different provinces of Sicily, mainly in lemon and orange orchards showing characteristic symptoms of gummosis and cankers. Our investigation is gradually revealing, through a multi-locus approach, the complexity of the etiology, since different species belonging mainly to the *Botryosphaeriaceae* and *Diaporthaceae* have been isolated. Our investigation is helping us to progress in the knowledge of the etiology of citrus canker disease, which is fundamental to understand the biology and epidemiology of the involved pathogens and to establish sustainable management strategies.

SE11 C06

FROM BRANCH CANKER TO “ESCUDETE” OLIVE DISEASES CAUSED BY BOTRYOSPHAERIACEAE SPP

María Isabel Márquez Pérez

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The olive tree (*Olea europaea* subsp. *europaea*) is the most extensive woody crop globally, covering 12 million hectares. Like other perennial crops, the olive tree is affected by numerous (> 20) *Botryosphaeriaceae* species, which usually cause a characteristic branch dieback, diminishing olive production. This re-emerging disease has been exacerbated by factors such as the expansion of olive cultivation, global warming, and the adoption of mechanized harvesting systems that cause numerous branch wounds. Olive branch dieback is an oligocyclic disease causing polyetic epidemics, whose control measures should be focused on reducing inoculum in the field and controlling the primary inoculum by fungicides. Since the behavioral plasticity of *Botryosphaeriaceae* species, olives suffer other diseases caused by these. For

instance, *Macrophomina phaseolina*, a soil-borne pathogen, induces significant losses in olive seedlings under water stress, particularly in highly infested soils following sunflower cultivation. Finally, we would like to highlight the olive “Escudete” (small shield), caused by *Botryosphaeria dothidea*, the most critical disease of table olive fruits. Fascinating the female of *Prolasioptera berlesiana*, a small Diptera whose larvae prey on the olive fly (*Bactrocera olea*) eggs, inoculates *B. dothidea* when it deposits its egg adjacent to the fly egg into the fly puncture. This allows the *Prolasioptera* larvae to feed on *B. dothidea* mycelium when the fly egg is absent; a need arises due to frequent false punctures (no oviposition) made by the fly to avoid predation. Our microscopic studies show that the *Prolasioptera* female has a particular structure (mycangia) to carry *B. dothidea* spores in the last abdominal segments close to the ovipositor, which suggests a mutualistic association between both agents. In summary, a comprehensive understanding of the biological cycles of *Botryosphaeriaceae* species is crucial for effectively managing these significant diseases in an environmentally friendly manner.



Figure 1. Olive diseases caused by *Botryosphaeriaceae* species. A) Branch dieback caused by *Neofusicoccum mediterraneum*. B) “Escudete”, caused by *Botryosphaeria dothidea*, in table olive fruit ready for consumption. Despite the small lesion, it significantly diminishes the fruit’s quality (Images by Juan moral)

SE11 C07

CANKER DISEASES IN OLIVE AND TREE NUTS IN SOUTHERN SPAIN

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Olive is the main cultivated woody crop in southern Spain, followed by tree nuts including almond, pistachio, and English walnut. Canker diseases are frequent in this geographic area. This speech aims to overview the different syndromes and the diversity of fungi associated with canker diseases in olive and tree nuts in southern Spain. Branch dieback and cankers in olive in Spain have traditionally been associated with *Neofusicoccum mediterraneum*. However, emerging canker diseases have been occurred due to olive crop intensification. Thus, olive leprosy caused by *Phyctemavagabunda*, and cankers and vascular streaking associated with *Cadophora luteo-olivacea*, *Phaeoacremonium* spp. and *Pseudophaeoniella oleicola* have been reported in the last few years. Regarding tree nuts, almond is the most affected crop showing a wide diversity of tree decline syndromes. More than 20 fungal species have been associated with almond canker diseases, with *Botryosphaeriaceae* fungi being the most frequent, and *N. parvum* being the most aggressive species. Almond wilt and crown rot caused by *Fusarium oxysporum*; and gumming

and sudden death caused by *Phytophthora niederhauserii* and *Phytophthora litorale* have been reported in specific growing areas. However, branch dieback associated with *Diaporthe amygdali* is not frequent in almonds in southern Spain, although it can be observed with high incidence and severity in rainy years. The etiology of branch dieback, shoot blight, and cankers in pistachio and English walnut has been associated with *Botryosphaeriaceae* and *Diaporthe* species, with the first being the most frequent and aggressive in both tree nuts. In addition, *N. mediterraneum* has been reported as the main causal agent of shoot and panicle blight in pistachio. All these studies gather the main advances in the knowledge of the etiology of olive and tree nut canker diseases, which are essential to understand the biology of the causal agents and to establish integrated management strategies.

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SE11 C08

BOTRYOSPHERACEAE DIEBACK AGENTS: UNDERSTANDING OF THEIR AGGRESSIVENESS AND RECENT ADVANCES IN MANAGEMENT

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Botryosphaeria dieback is a big threat for global viticulture. Both grapevine nurseries and vineyards are very susceptible to infections by *Botryosphaeria* pathogens due to several cuts and wounds made during the propagation process and the entire life cycle of grapevine, respectively. In the *Botryosphaeriaceae* family, many of its members can live as endophytes and turn into aggressive pathogens following the onset of environmental stress events. Their ability to cause disease may rely on the production of a broad set of effectors, such as cell wall-degrading enzymes, secondary metabolites, and peptidases. We show that these *Botryosphaeriaceae* genomes possess a large diversity of carbohydrate-active enzymes (CAZymes; 128 families) and peptidases (45 families). *Botryosphaeria*, *Neofusicoccum*, and *Lasiodiplodia* presented the highest number of genes encoding CAZymes involved in the degradation of the plant cell wall components. The genus *Botryosphaeria* also exhibited the highest abundance of secreted CAZymes and peptidases. Generally, the secondary metabolites gene cluster profile was consistent in the *Botryosphaeriaceae* family, except for *Diplodia* and *Neoscytalidium*. At the strain level, a focus will be done on *Neofusicoccum parvum* NpBt67 in terms of some CAZymes activities and 2 secondary metabolites recognized as phytotoxins, mellein and terreutin. Finally, recent research based on the use of biocontrol agents will be presented.



SKALKOTAS

18.30-21.00 Concurrent Session 7C

Understanding Pathogen-Vector-Host Interactions in Globally Important Pathosystems

Chair: Shaopius Mondal (*University of Nebraska - Lincoln, USA*)
Elizabeth Jeannette Cieniewicz (*Clemson University, USA*)

SE07 001

UNLOCKING THE SECRETS OF ROSE ROSETTE DISEASE: DECODING VIRUS DYNAMICS AND VECTOR COMPETENCE IN ERIOPHYOID MITES

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Rose Rosette Disease (RRD) represents a mayham for the U.S. rose industry causing losses measured in the tens of millions of USD. The interaction between the rose rosette emaravirus (RRV) and its eriophyoid mite vectors is pivotal to developing effective control strategies. However, understanding of how these mites transmit RRV and the factors enhancing their vector capacity is lacking, necessitating further investigation to fill these knowledge gaps. Employing advanced quantitative techniques, this study tracks the replication of RRV within two mite species, *Phyllocoptes adalius* and *P. fructiphilus*. This approach aims to illuminate the processes of virus replication and assess the effectiveness of these mites as vectors, providing critical insights into the dynamics of virus transmission. The study reveals that RRV replicates actively within *P. fructiphilus*, a confirmed vector, in contrast with *P. adalius* a non-vector. This finding is crucial for directing disease management strategies, underscoring the importance of accurately identifying and targeting vector species. Additionally, the observed variability in viral load in mites over time suggests that factors such as mites' developmental stages and behaviors might influence virus retention and replication. These insights highlight the complex interactions between virus and vector, emphasizing the need for a comprehensive understanding of these relationships. This research marks a significant step forward in decoding the complex mechanisms of RRV transmission by eriophyoid mites, laying the groundwork for the development of more precise and effective control strategies against RRD. Given the critical threat RRD poses to the rose industry, these findings underscore the urgent need for continued and focused research efforts in this domain, aiming to safeguard and ensure the longevity of rose cultivation.

SE07 002

IMPACT OF LOW-SUSCEPTIBLE WHEAT GENOTYPES ON THE EPIDEMIOLOGY OF WHEAT DWARF DISEASE BY MONITORING PLANT / VIRUS / VECTOR INTERACTIONS

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Wheat dwarf disease (WDD), one of the most important viral diseases on cereals, is caused by Wheat dwarf virus (WDV, genus *Mastrevirus*). WDV is transmitted in a persistent manner by the leafhopper *Psammotettix alienus*. Due to the lack of resistance/tolerance against WDV in cultivated cereals, main control strategies against WDV are based on cultural practices and/or chemical uses. However, these methods are not efficient enough to protect crops against high disease pressures. Investigating the effect of WDV-susceptible host genotypes on key parameters involved in the epidemiology of WDD could help for the development of management strategies based on the deployment of low-susceptible cultivars. In this work, vector-plant (i.e. host preferences, survival and fecundity) and virus-plant (i.e. infection rate, viral prevalence in a mosaic of wheat cultivars) interactions were evaluated under controlled conditions for 12 wheat cultivars available for French farmers. As a first step, data collected were used to cluster wheat cultivars according to their host quality for WDV and for *P. alienus*. Clustering analysis showed that among tested cultivars the cv. Filon presents a low host quality for both the virus and the leafhopper. To complete this analysis, latency period of WDV in cv. Filon was studied. Results showed that cv. Filon slightly but significantly decreases parameters linked to the transmission efficiency of WDV. Taken together, data acquire in this work highlight that introduction and spread of WDV in wheat fields would be lowered in the presence of cv. Filon. While the molecular basis underlying the observed phenotype should be assessed, results presented in this work must be considered for future breeding programs or for agronomic management strategies against wheat dwarf disease.



Concurrent Sessions

Tuesday, 2 July

SE07 003

ENDOPLASMIC RETICULUM (ER)- ASSOCIATED INTERACTIONS OF LIBERIBACTER SOLANACEARUM WITH ITS CARROT PSYLLID VECTOR

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Candidatus Liberibacter solanacearum (CLso) haplotype D is a phloem-restricted gram negative plant pathogenic bacterium and transmitted by the carrot psyllid *Bactericera trigonica* in a persistent and propagative mode, and causes the carrot yellows disease. Our previous studies confirmed that the expression of key genes in the ER-associated protein degradation (ERAD), were upregulated in CLso infected psyllids, suggesting induced ER stress. Prolonged ER stress, induces the unfolded protein response (UPR) which lies in the basis of orchestrated signaling pathways leading to the activation of apoptosis. Here we investigated the involvement of UPR in the interactions with CLso and its role in the observed induced apoptosis. By quantifying the expression of UPR and apoptosis related genes using qRT-PCR, testing a key UPR protein (PERK) expression and activity using immunostaining and investigating PERK's expression profile after using chemical ER stressors, we found that apoptosis is indeed induced in *B. trigonica* as a result of CLso infection. CLso affect the expression of PERK in the psyllids midgut and it significantly changes the expression profile of PERK in case of chemical induction of ER stress. These results indicate that PERK pathway and UPR are putatively involved in the observed induced apoptosis as an immune response following CLso infection. Though their exact role is yet to be determined, these findings are the first step for understanding the interactions between ER stress pathway, UPR, and CLso in the psyllids midgut. Understanding these interactions is crucial for the development of alternative management approaches for Liberibacter-associated plant diseases

and effective way of managing YDVs. To begin addressing this knowledge gap, we sampled symptomatic cereals and grasses from south- eastern Australia. All plant samples were tested for cereal viruses using a serological method, tissue blot immunoassay (TBIA). However, selected samples were also examined using high- throughput sequencing (HTS). Several YDV species that were not previously known to be present in Australia were found, while the genome sequences of YDV species long known to be present in Australia were quite different to other isolates of the same species from other countries. New molecular diagnostic tests have been designed and are being used to further examine the incidence and distribution of each of the YDV species that was identified by HTS. Additionally, a series of glasshouse experiments have been conducted to investigate the epidemiology of several of the YDV species that have recently been reported in Australia by examining their relationships with their plant hosts and the aphids that transmit them. The aim of this work is to reduce on- farm yield losses that result from infection with these widespread and damaging viruses.

SE07 005

MODE OF ACTION OF OLIGONUCLEOTIDE INSECTICIDES (DNA CONTAINMENT MECHANISM)

Gal'chinsky N., Oberemok V.

V.I. Vernadsky Crimean Federal University, Simferopol, Russian Federation

Contact unmodified antisense DNA (CUAD) biotechnology uses oligonucleotide insecticides that act through DNA containment mechanism comprised of 2 steps. At first step, antisense DNA oligonucleotide (oligonucleotide insecticide) complementarily interacts with target rRNA (in other words, 'arrests' target rRNA) and interferes with normal functioning of ribosomes; this process is accompanied with insect pest mortality. Apparently, target rRNA hypercompensation by the DNA-dependent RNA polymerase is the only way for the insect cell to fight for life in such a situation when target rRNA is 'arrested' by antisense DNA oligonucleotides. Both, 'old' blocked target rRNA in ribosomes (ribosomes apparently do not function properly in this situation) and 'newly' synthesized target rRNA, can be found by RT-PCR and, thus, we detect target rRNA hypercompensation. At second step, RNase H cleaves target rRNA and substantial decrease in its concentration occurs. It has not escaped our notice that antisense DNA oligonucleotides are likely to participate in defense systems (innate immunity) against ssDNA viruses for which hemipteran insects serve as major vectors and also against DNA viruses that normally infect them. Obviously, insect nucleases cleave specific DNA sequences of the invader producing ssDNA oligonucleotides and subsequently initiate degradation of target viral RNAs through DNA containment mechanism (action of viral ssDNA fragment). Oligonucleotide insecticides are highly effective against hemipteran pests with unique characteristics for green plant protection (high biodegradability, avoidance of target-site resistance, selectivity in action, low carbon footprint). Research results obtained within the framework of a state assignment V.I. Vernadsky Crimean Federal University for 2024 and the planning period of 2024–2026 No. FZEG-2024-0001.

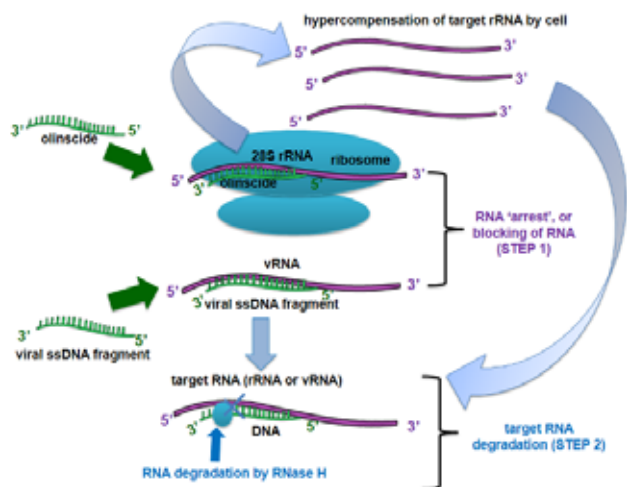
SE07 004

CEREAL VIRUS DIVERSITY AND EPIDEMIOLOGY IN SOUTH- EASTERN AUSTRALIA

Nancarrow N.^{1,2}, Lam S.¹, Rodoni B.^{3,4}, Kinoti W.³, Trebicki P.^{1,5}

1. The University of Melbourne, Parkville, Australia, 2. Agriculture Victoria, Horsham, Australia, 3. Agriculture Victoria, Bundoora, Australia, 4. La Trobe University, Bundoora, Australia, 5. Macquarie University, Sydney, Australia

Yellow dwarf viruses (YDVs) are transmitted by several aphid species and are important pathogens of cereals, both in Australia and worldwide. YDVs are the most prevalent, widespread, and important cereal viruses in Australia and can significantly reduce grain yield in critical cereal food crops such as wheat, barley and oats. Despite their importance and the description of new YDV species and variants from other countries, little is known about their diversity in the Australian broadacre farming landscape. However, this information is critical in the search for more targeted and effective virus-resistant cereal varieties, which is currently the most promising



Mode of action of oligonucleotide insecticides and viral ssDNA fragments (DNA containment mechanism)

MC2

18.30-21.00 Concurrent Session **10B**

Impact and Control of Transboundary / Invasive Pests

Chair: **Moriyama Hiromitsu** (Tokyo University of Agriculture and Technology, Japan)

Arie Tsutomu (Tokyo University of Agriculture and Technology, Japan)

SE10 C05

THE ROLE OF WEED SEED CONTAMINATION IN GRAIN COMMODITIES AS PROPAGULE PRESSURE

Yoshiko Shimono

Weed Science Lab, Graduate School of Agriculture, Kyoto University, Japan

The international grain trade is a major pathway for the introduction of alien plants because grain commodities can be contaminated with various weed seeds. To evaluate how alien weed seeds derived from imported grain commodities affect the local flora in international trading ports, we conducted a floristic survey at each of the 10 grain landing ports and non-grain landing ports throughout Japan and compared the flora between these two types of ports. We also surveyed weed seed contamination of wheat imported into Japan, and the contamination rate was calculated for each species based on our survey and previous studies on weed seed contamination. The flora clearly differed between the grain landing ports and the non-grain ports (Fig. 1). There was a tendency for the more abundant species at the grain landing ports to show higher contamination levels in grain commodities (Fig. 2). These results indicate that contaminant seeds spill from imported grain in grain landing ports and the most common contaminant species are likely to become established. We clearly show that weed seed contamination in grain commodities plays an important role in propagule pressure. Gathering information about the prevalence of weeds in grain-exporting countries and monitoring the weed species composition in imported grain commodities is becoming increasingly important for predicting the unintentional introduction of troublesome weeds and identifying effective weed management options.

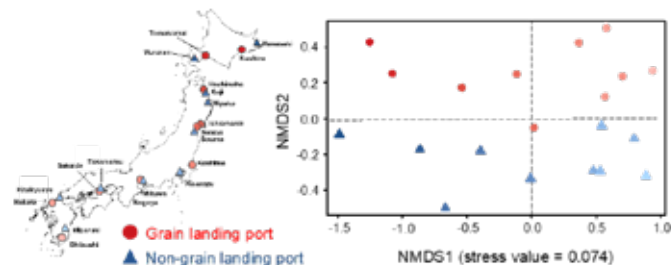


Fig. 1 Non-metric multidimensional scaling (NMDS) ordination diagrams based on the plant species compositions of ports.

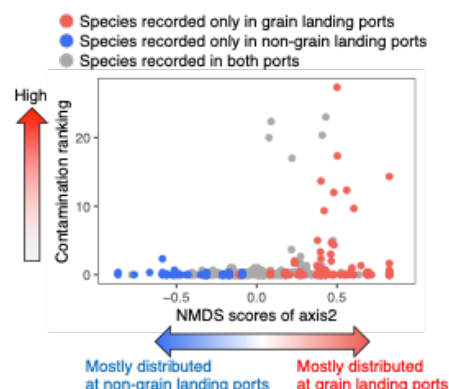


Fig. 2 Relationship between the distribution pattern and the average contamination ranking of each species. The x-axis shows the NMDS score of axis-2 of each species. Species with positive scores distributed more in the grain landing ports, and species with negative scores distributed more in the non-grain landing ports.

SE10 C06

CHALLENGE TO FUSARIUM WILT OF BANANA AND POD ROT OF CACAO IN THE PHILIPPINES

Kyoko Watanabe

Tamagawa University College of Agriculture, Tokyo, Japan

A joint project between a Japanese team and a Philippine team has been launched under SATREPS Program* as the "Banana and Cacao Diseases Management Project", which will start in 2020. Banana Fusarium wilt disease and cacao pod rot are the main disease management targets of the project. As the basis of disease management, the project mainly focuses on the correct diagnosis and field management to reduce the risk of disease. So far, the project has shown that not only *Fusarium oxysporum* f. sp. *cubense* TR4 is a pathogen for Fusarium wilt of Cavendish banana, but that other species are also involved, and pod rot of cacao is not only caused by black pod caused by the genus *Phytophthora* but also by the genus *Lasiodiplodia*, which causes VSD disease is widespread. In addition to these results, I will also present a simple diagnosis by LAMP method and disease control by ethanol soil reduction as part of the results of the project.

* SATREPS is a Japanese government program that promotes international joint research aimed at solving global issues.



SE10 C07

DISEASES OCCURRING IN BANANA CULTIVATION IN PERU AND THEIR CONTROL

Liliana Aragón

Institute of Agriculture, Universidad Nacional Agraria La Molina, Peru

In Peru, bananas and plantains are grown mainly in different coastal and forest agroecosystems. In Piura Region (north coast) the production of organic bananas for export is carried out. In the forest (although there is a diversity of forest ecosystems) bananas (moquicho, isla, seda and morado) and plantains (palillo, bellaco) are produced. In each agroecosystem, whose environmental characteristics (temperature, relative humidity, rainfall, soil) and host diversity (banana and plantain) different pathosystems can exist. Piura region is characterized by temperatures between 20°C to 35°C, precipitation is seasonal (during the summer months, January to March; it can extend until April). During El Niño years, precipitation volumes exceed the historical average. The production system in large areas is made up of few companies however small farmers predominate (less than 5ha); whose number exceeds 500 producers in the Sullana area. Disease problem reported is crown rot caused by *Verticillium theobromae*. In 2021, the presence of *Fusarium oxysporum* f. sp. *cubense* R4T was reported for the first time, in Sullana; which is the main problem, now. The Peruvian Forest can be divided in different ecosystems: the rain forest (correspond altitudes between 500 – 600 meters above sea level and is typical of the eastern slope of the Andes) and with differences between the northern, central and southern areas of the country. The lower parts or Amazonian savanna are characterized by having acidic soils, dominated by kaolinite clay. The presence of yellow and black Sigatoka is reported. Presence of *Fusarium oxysporum* f. sp. *cubense* race 4 subtropical is also reported in Chanchamayo area.

SE10 C08

IMPORTANT BACTERIAL DISEASES ON RICE, VEGETABLES AND FRUITS IN SOUTHERN VIETNAM AND THEIR MANAGEMENT

Nguyen Thi Thu Nga

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Vietnam has a tropical climate with a rainy season for a prolonged six months annually; this is a favorable condition for the aggressiveness of phyto-bacterial pathogens. In rice, bacterial leaf blight is caused by *Xanthomonas oryzae* pv. *oryzae*, grain rot caused by *Burkholderia glumae* are the main diseases causing rice yield losses. In vegetables, bacterial wilt caused by *Ralstonia solanacearum* is a severe disease in different crops. In addition, the genus *Xanthomonas* is also an important pathogen on foliar in almost all vegetables, e.g., *X. axonopodis* pv. *allii* causing leaf blight on onion, *X. vesicatoria* causing bacterial leaf spot on chilli and tomato. On fruits, huanglongbing and canker are the main important bacterial diseases in citrus. In jackfruit, two emerging new bacterial diseases cause economic losses, i.e. gummosis stem canker, giving quick dieback of branches and trunk (caused by *Pectobacterium carotovorum*) and jackfruit-bronzing (caused by *Pantoea stewartii*), characterised by yellowish-orange to reddish discoloration of the affected pulp. An integrated disease

management strategy is considered for controlling these bacterial diseases, where good cultural practices, induced disease resistance and biological control are combined with chemical control. In current, bacteriophages are promising agents for controlling bacterial diseases. For instance, soil drenching or foliar spraying with a bacteriophage suspension at 10^7 - 10^8 pfu/mL can significantly reduce soil bacteria and foliar bacteria pathogens, respectively, in many investigated crops. In addition, some promising chemical inducers such as calcium silicate, salicylic acid and benzothiadiazole has shown potential in activating plant defence against pathogens in several host-pathogen systems such. In conclusion, bacterial plant pathogens are important agents causing yield loss in almost all crops in the southern part of Vietnam, and biocontrol using bacteriophages and inducing resistance are promising environmentally friendly methods for disease management. These methods should be considered in plant protection in sustainable agriculture.

SE10 C09

SEX PHEROMONES: AN EFFECTIVE TOOL FOR SUSTAINABLE MANAGEMENT OF AGRICULTURAL HARMFUL PESTS IN THE MEKONG DELTA OF VIETNAM

Le Van Vang

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The Mekong Delta is the key agricultural production area in Vietnam. Due to agricultural production being heavily focused on producing yield, farming techniques have been carried out in a highly intensive manner, with an overreliance on the use of agrochemicals. In the long run, this practice has made the production unsustainable. Sex pheromones are semiochemicals that act as sexual behavior stimulants for other individuals in the same species (intraspecific activity). To date, sex pheromones have been identified in more than 1500 insect species; most of the studies were primarily aimed at pest management. The application of sex pheromones is considered environmentally friendly and a good alternative to the use of insecticides in the management of agricultural insect pests. In the Mekong Delta of Vietnam, research and application of insect sex pheromones have been carried out on 17 lepidopterous and one coleopteran species. Sixteen studied species are agricultural pests, including economically important species such as the sweet potato weevil (*Cylas formicarius*), the citrus leafminer (*Phyllocnistis citrella*), the tobacco cutworm (*Spodoptera litura*), the beet armyworm (*Spodoptera exigua*), the eggplant fruit borer (*Leucinodes orbonalis*), the citrus fruit borer (*Citripestis sagittiferella*) and the yellow peach moth (*Conogethes punctiferalis*). The remaining studied species is a weed biological control agent, the mimosa stem borer (*Carmenta mimosa*), which was imported into Vietnam to control the invasive *Mimosa pigra* weed. Sex pheromones have been applied for monitoring population dynamics (on 8 species), mass trapping (on 4 species), mating disruption (on two species), recognition of appearance (on one species), in the "push and pull" system (on two species) and as an entomopathogenic fungal transmission factor (on one species). Research and application of insect pheromones have positively contributed to the development of sustainable insect pest management in the Mekong Delta of Vietnam.

SE10 C10

STATUS OF BANANA AND CACAO INDUSTRY IN THE PHILIPPINES

Dionisio G. Alvindia

Director IV/Scientist IV, Philippine Center for Postharvest Development and Mechanization (PHilMech) and Affiliate Professor VI, College of Agriculture, Central Luzon State University (CLSU)

The second-largest producer and exporter of bananas in the world is the Philippines. Cavendish remains the most-grown and exported variety in the Philippines for 53%. The Philippines exported a total of 2.3 million metric tons (MT) in 2022 with a four percent decline from the 2.4 million MT shipped in 2021 based on FAO report. The Philippines aims to become the largest producer of bananas in the world by 2029 but was recently struggling with the widespread Panama Tropical Race 4, bacterial wilt disease, and Mosaic disease. The dreaded Panama disease have ravaged more than a quarter of all the banana plantations in Mindanao. Areas infected may have doubled from 15,000 hectares identified by the Department of Agriculture (DA) in 2015. Meanwhile, the local annual production of cacao is estimated at 15,000 MT, most of which is produced in Davao region of Mindanao, the most southern island of the Philippines. Local consumption is 50,000 MT, so there is a shortage of 35,000 MT, which the country imports. Some 15 years ago the Government of the Philippines started to invest in the cacao sector and production has grown steadily. Low survival and yield in the nursery and plantation due to pest and disease infestation, and inadequate preventive and control measures against pests and diseases; and low yield due to unproductive or old cacao plantations are some of the problems besetting cacao industry in the country.

and *Botourmiaviridae*. Further experiments focused on *Leptosphaeria biglobosa* quadrivirus (LbQV) 1. A direct comparison of the growth and virulence of LbQV-1-infected and -free isogenic lines illustrated that LbQV-1 infection caused hypervirulence in *L. biglobosa* and resulted in induced systemic resistance towards *L. maculans* in *B. napus* following lower leaf pre-inoculation with the LbQV-1-infected isolate. Analysis of the plant transcriptome suggests that LbQV-1 presence leads to subtle alterations in metabolism and plant defences. For instance, transcripts involved in carbohydrate and amino acid metabolism are enriched in plants treated with the LbQV-1-infected isolate, while pathogenesis-related proteins, chitinases and WRKY transcription factors are differentially expressed. These results illustrate the potential for deliberate inoculation of plants with hypervirulent *L. biglobosa* to decrease the severity of phoma stem canker later in the growing season.

SE10 C12

EXPLORING ALLELOCHEMICALS IN RICE VARIETIES FOR ECO-FRIENDLY WEED MANAGEMENT: A METABOLOMICS APPROACH

Ho Le Thi

Plant Protection Department, College of Agriculture - Cantho University, Vietnam

The intensive use of chemical herbicides for weed control in rice (*Oryza sativa* L.) production can lead to water contamination and the development of herbicide-resistant rice varieties. Bio-based allelochemical herbicides (bioherbicides) with novel modes of action offer a promising alternative to current chemical-based herbicides. This study utilizes UPLC-HRMS and the XCMS platform to detect and analyze potential allelochemicals in various rice varieties, including OM5451, OM380, OM3536, OM6976, OM4498, OM2395, OM-N406, OM5930, and OM7347. A total of 20 allelochemicals were identified across these varieties, with varying concentrations: 2,4-Dihydroxybenzaldehyde, 2,6-Dimethoxybenzoic acid, 3,4-Dihydroxybenzoic acid, 3,4-Dihydroxyphenylacetic acid, 3-Hydroxybenzoic acid, 4-Hydroxybenzoic acid, 5-Methoxysalicylic acid, 7-Oxostigmasterol, Benzoic acid, 2,4-Dimethoxybenzoic acid, 2,5-Dihydroxybenzoic acid, 3,4-Dimethoxybenzoic acid, 3,5-Dihydroxybenzoic acid, 3,5-Dimethoxycinnamic acid, Coumarin, Ergosterol peroxide, p-Hydroxycinnamic acid, Salicylic acid, and Vanillic acid. Four rice varieties, OM4498, OM3536, OM5930, and OM-N406, contained more than 50% of the total identified allelochemicals. These were followed by OM2395, OM5451, OM6976, and OM380, which had between 35%-45%, and OM7347, which had 20%. Among these nine varieties, OM5930 exhibited the highest number and concentration of allelochemicals. These findings underscore the significant potential of specific rice varieties in contributing to sustainable weed management practices. By leveraging the natural allelochemicals present in varieties like OM5930, OM4498, OM3536, and OM-N406, it is possible to develop bioherbicides that offer an effective, economical, and environmentally friendly alternative to traditional chemical herbicides. This approach aligns with the goals of promoting plant health and supporting human welfare, as it mitigates the negative impacts of chemical herbicides on water quality and ecosystem health. Further research should focus on

SE10 C11

A MYCOVIRUS INDUCES SYSTEMIC RESISTANCE IN OILSEED RAPE AGAINST PHOMA STEM CANKER

Ioly Kotta-Loizou

FHEA, Molecular Microbiology, Department of Life Sciences, Faculty of Natural Sciences, Imperial College London, United Kingdom

Brassica napus, also known as oilseed rape or canola, is a major source of vegetable oil, livestock feed and biodiesel worldwide. Phoma stem canker or canola blackleg is one of the most important diseases affecting *B. napus*, caused by the phytopathogenic fungi *Leptosphaeria maculans* and *L. biglobosa*. The aim of the study was to investigate the diversity of mycoviruses in *L. maculans* and *L. biglobosa*, together with their role in fungal pathogenesis. To this end, a panel of *Leptosphaeria* field isolates from *B. napus* were screened for the presence of mycoviruses using a small-scale double-stranded (ds) RNA extraction protocol. Mycoviruses were then purified, visualised, cloned, sequenced and classified. Virus-free isogenic lines were generated following treatment of virus-infected isolates with the protein synthesis inhibitor cycloheximide. Following mycovirus eradication, the growth and virulence of virus-infected and virus-free isogenic lines were compared in vitro and in planta. Screening for dsRNA revealed the presence of novel mycoviruses in *L. biglobosa* belonging to families *Quadriviridae*, *Polymycoviridae*



field trials to validate the efficacy of these allelochemicals in diverse agricultural settings and explore the mechanisms by which they exert their herbicidal effects.

SE10 001

ESTABLISHMENT AND APPLICATION OF EDNA-BASED MOLECULAR IDENTIFICATION METHODS FOR THREE IMPORTANT FRUIT-BORING MOTHS

Li A.^{1,2}, Li L.^{1,2}, Liu L.^{1,2}

1. College of Plant Protection, China Agricultural University, Beijing, China, 2. Key Laboratory of Surveillance and Management for Plant Quarantine Pests, Ministry of Agriculture and Rural Affairs, P. R. China, Beijing, China

Cydia pomonella, *Grapholitha molesta*, and *Carposina sasakii* are all high-priority control targets in orchards. The mixed infection of these moths not only brings difficulties to their prevention and control, but also greatly affects the national and international trade of the fruits. Traditional morphological and molecular identification method is body-dependent and time-consuming, for which fruit dissection is needed. Thus, a non-destructive, rapid and efficient identification technology for fruit-boring moths is urgently needed. As a non-invasive sampling technique, environmental DNA (eDNA) enables testing DNA from various media, including water, feces and soil. eDNA-based molecular methods have emerged as a successful tool for species detection. In this study, an eDNA tool was developed for the early detection of three fruit-boring moths. Firstly, the eDNA extraction method was optimized, including the method for sampling and the parameters (kinds and pore size) of filter membranes. And then the specificity and sensitivity of the primers were tested. Finally, the optimized eDNA-based detection technology was applied to detect the infection of three species of fruit-boring moths in different orchard. The results showed that the eDNA with the highest concentration and quality was obtained by filtration the victim pulp, which has been soaked in the water for 15 mins, using a combination of 10 µm mixed cellulose esters (MCE) membrane and 0.45 µm cellulose acetate (CA) membrane. The primers developed for the three kinds of moth exhibited high sensitivity. The eDNA based technology successfully detected the target moth species in the orchard, no matter the fruit was damaged by one or more than one kinds of moth. One eDNA-based technology was established and successfully applied in the field, which provided theoretical basis for the establishment of non-destructive fruit quarantine testing technology, and also provided references for eDNA studies of other species.

SE10 002

THE EFFECT OF INSECTICIDE SPRAY REGIMES ON FALL ARMYWORM DAMAGE AND MAIZE GRAIN YIELD

Otim M.¹, Ogwal G.², Adumo S.³, Adur S.¹, Jaggwe J.⁴, Gichuru L.⁴

1. National Agricultural Research Organization, National Crops Resources Research Institute, Namulonge, Kampala, Uganda, 2. Alliance of Bioversity International and CIAT, Kampala, Uganda, 3. National Agricultural Research Organization, National Agricultural Research Laboratories, Kawanda, Kampala, Uganda, 4. Alliance for a Green Revolution in Africa, Nairobi, Uganda

Spodoptera frugiperda is a significant maize pest in Uganda. Infestation and damage appear to be dependent on the environmental conditions. Assessing the effectiveness and profitability of insecticide application in different agroecological zones is vital for sustainable management. In this study, we assessed the effectiveness of Emamectin benzoate in reducing *S. frugiperda* leaf damage and abundance, and the benefit-cost ratio of the different insecticide spray regimes. The study was carried out in two rainy seasons of 2018 and the first rainy season of 2019 in three different agroecological zones. The experiment was laid out in a split-plot design with Emamectin benzoate (5% w/w) insecticide spray regimes as the main- and eight maize varieties as the subplot factors, respectively. The spray regimes consisted of two sprays applied at 40 and 50 Days after emergence (DAE), three-ten-day interval sprays starting at either 10, 20 and 30 DAE, one twenty-day-interval spray starting at 10 DAE, and a five ten-day-interval sprays starting at 10 DAE. An untreated control was included for comparison. Data were collected on leaf damage, abundance of *S. frugiperda* and grain yield. Maize variety did not influence the damage and abundance of *S. frugiperda* larvae. Early application of Emamectin benzoate at 10-day intervals starting at 10 or 20 DAE significantly reduced larval infestation and damage. Conversely, more egg masses were recovered in the sprayed plots. The delayed start of insecticide application (30 DAE onwards) resulted in higher leaf damage, larval abundance and grain yield. Insecticide application generally resulted into higher benefit-cost ratio (>1), although not in all seasons. Our results have demonstrated that spray applications reduce larval abundance and leaf damage, but did not always lead to higher benefit-cost ratio. It is thus important to scout and time control to reduce costs of fall armyworm management.



Concurrent Sessions

Tuesday, 2 July

SE10 003

THE INTERPLAY BETWEEN CLIMATE CHANGE AND PESTICIDE RESISTANCE: THE CASE STUDY OF THE TWO-SPOTTED SPIDER MITE

Regon P., Ben- Aziz O., Kliot A.

Volcani Institute Aro, Israel

The two-spotted spider mite (TSSM, *Tetranychus urticae*) is a generalist plant pest with worldwide distribution, known for its ability to quickly develop acaricide resistance. TSSM is highly heat resistant and is spreading and invading new areas where the climate is getting warmer. Developing resistance to pesticide can come at a cost to the organism, often in the form of reduced resistance to other stressors, such as heat. In our lab, we set out to study the effect warming climate has on pesticide resistance abilities of TSSM. To do so, we examined the effect of exposure to different temperatures for varying lengths of time on resistance abilities of field- collected TSSM populations with varying pesticide resistance abilities. We found that the heat significantly affects different aspects of TSSM pesticide resistance, including mortality rates, escape rates and fecundity. The genetic background of the mites, the timing of the heat exposure and the pesticide tested all can have significant effects on TSSM reactions. We are now in the process of utilizing our finding to better tailor pest management programs to control TSSM in the field in warm areas. We are also interested in studying the physiological aspect of our results to better characterise the effects of heat on the mites' physiology.



MC3

18.30-21.00 Concurrent Session 12

Global perspectives in herbicide-resistance and new integrated weed management approaches"

Chair: Baruch Rubin (Hebrew University of Jerusalem, Israel)
Ilias Travlos (Agricultural University of Athens, Greece)

SE12 C01

HERBICIDE-RESISTANT WEEDS: A CHALLENGE TO THE SUSTAINABILITY OF AGRICULTURE

Baruch Rubin

RH Smith Institute of plant Sciences and Genetics in Agriculture, Faculty of Agriculture, Food and Environment, The Hebrew University of Jerusalem, Rehovot, Israel

Herbicide-based weed management is the most effective and widely adopted practice in agriculture today. Although the adoption of bioherbicides is increasing, the use of herbicides in agriculture continue to expand. Overuse and misuse of herbicides increase the selection pressure on the weed population and enhances the evolution of herbicide-resistant weeds (HRW), which threaten the sustainability of the agroecosystem. In spite of the advances in understanding of herbicide resistance mechanisms, the number of HRW is constantly increasing causing severe economic and environmental damage. Climate changes, stricter regulations and increased number of banned herbicides further aggravate the situation, resulting in increased selection pressure caused by fewer herbicides acting with fewer mechanisms. Weeds confer resistance due to inherited alteration of the herbicide-binding site (TSR) occurring under strong selection pressure. The risk of evolved TSR is higher when the herbicide inhibits a specific and vital process. Non-target site mechanism (NTSR), often based on enhanced activity of different enzyme families catalyzing detoxification processes that may result in a multiple herbicide resistance. Obligatory exogamous weeds (e.g., *Lolium* spp.) disseminate the HR trait by pollen grains, are the most dangerous. Invasive weeds may "carry" HR traits and transfer them within and between countries and continents. Lack of workers and increased farm size increase over-dependence on herbicides so their use continues to increase. In order to reduce the detrimental impact of HRW and ensure food security, the ag community should be more proactive. Improved herbicide formulations should be developed to overcome uptake and translocation barriers; New multisite less-specific herbicides should be designed and novel synergists antagonizing metabolic detoxification processes are crucially needed. Above all, farmers should diversify the current control practices by adopting novel non-chemical management methods such as precision application and robotics.

SE12 C02

HERBICIDE RESISTANCE IN EUROPE: CURRENT STATUS, FUTURE TRENDS AND MANAGEMENT

Vaya Kati, Ilias G. Eleftherohorinos

School of Agriculture, Aristotle University of Thessaloniki, 54124 Thessaloniki, Greece

Herbicides remain the most efficient tool for weed control worldwide. However, populations of several weed species have evolved herbicide resistance (HR) to almost all modes of action (MOA), with the number of cases rising alarmingly. In Europe, more than 90 weed species developed resistance to 13 MOA, across 25 countries. Most problems occur in maize (35 species), cereals (31), orchards and grapevine (23), and several other crops. Herbicide resistance evolved mainly due to target-site alteration(s), while also non-target site mechanisms are suggested. Inline with the international trend, most HR cases in Europe occur to ALS-inhibitors (53 species, 22 countries), PSII-inhibitors (45, 17), ACCase-inhibitors (17, 14), glyphosate (9, 9), synthetic auxins (4, 8). Among the resistant grasses, *Alopecurus myosuroides* is widespread in central and northern Europe (12 countries) followed by *Apera spica-venti* (11), *Poa annua* (7), and *Avena fatua* (5), while *Avena sterilis* (6), *Sorghum halepense* (5) and *Lolium rigidum* (4) occur mainly in the south, and *Echinochloa crus-galli* (9) with *Lolium perenne* (8) throughout Europe. Resistant broadleaved weeds include *Chenopodium album* (17 countries), *Amaranthus retroflexus* (10), *Solanum nigrum* (10), *Papaver rhoeas* (10), *Stellaria media* (9), *Senecio vulgaris* (8), and *Tripleurospermum perforatum* (8). Several weed populations developed multiple resistance, while species like *A. myosuroides*, *L. rigidum* and *P. rhoeas* accumulated different resistance mechanisms concurrently in the same plant, thus surviving at each herbicide MOA and their mixture. As only few alternative MOA remain available for the control of resistant weeds in major crops, other management strategies need to be optimised for HR management and prevention. These include the combination of known agronomic practices like crop rotation, false seedbed, and enhancing the competitive ability of the crop, with novel tools, such as the use of fully autonomous weeding robots, laser, flame, electrocution, Harrington seed destructor, bioherbicides and crops with multiple herbicide tolerance.

The study was part of the project "Innovations in Plant Protection for sustainable and environmentally friendly pest control, InnoPP - TAEDR-O535675 that is "Funded by the European Union- Next Generation EU, Greece 2.0 National Recovery and Resilience plan, National Flagship Initiative "Agriculture and Food Industry"

SE12 C03

THE GOOD PROJECT FOR AGROECOLOGICAL WEED MANAGEMENT: CONCEPT AND LIVING LABS IN GREECE

Ilias Travlos¹, Nikolaos Antonopoulos¹, Ioannis Gazoulis¹, Panagiotis Kanatas²

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Weeds negatively affect the sustainability of EU farming systems with weed management relying to a large extent on herbicides. The reduction of pesticides' use and risk is the cornerstone of more sustainable and resilient farming systems. GOOD is a 4-year European project adopting a multidisciplinary approach, aspired to create, evaluate and optimize Agroecological Weed Management (AWM) systems. The core part of the project is being implemented by means of the establishment of 16 field scale Living Labs (LLs) in nine countries under different pedo-climatic conditions and in a wide range of annual (wheat, rye, triticale, maize, pea, cowpea, rice) and perennial (apple, olives, citrus, grapes, cherry) crops. Co-creation activities will be developed in conventional, organic and mixed farming systems. The experimental approach for both conventionally cultivated annual and perennial crops initially includes the use of different cover crops and their integration with different chemical and non-chemical weed management practices. During the second and third year, AMF inoculated cover crops will be used and their combination with AWM strategies will be assessed to increase the awareness, trust and adoption of successful AWM approaches. In 2023, two LLs were established in central and southern Greece with wheat and grapes, respectively. The first results and the observed differences build the basis for a feasible and efficient combination of AWM practices and their wider adoption in European agriculture.

emerged between spraying and initial seedbed preparation. Crop row spacing, treatments, and the interaction between the above factors significantly affected weed dry weight per unit area ($p \leq 0.05$). Treatment SSB 1 with 30 cm row spacing reduced weed biomass by up to 79% compared to treatment CS with 60 cm row spacing. Kenaf biomass yield was affected only by the factor of treatment ($p \leq 0.05$). For instance, treatment SSB 1 increased crop biomass yield by 35 and 65% compared to treatments CS and SSB 2, respectively. These results are consistent with multiple studies published in the recent literature. Such findings indicate that stale seedbed is a valuable tool to reduce weed pressure in biomass crops and improve their establishment especially when combined with other weed-suppressive cultural practices. Further research is required to evaluate more cultural practices for weed management in kenaf and other important biomass crops.

SE12 C05

THE POTENTIAL OF COVER CROP (CC) MIXTURES TO SUPPRESS WINTER AND SUMMER WEEDS IN CITRUS ORCHARDS

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Cover crops pose an attractive non-chemical weed management option in perennial crops. A field trial was conducted (2020 to 2021) to evaluate cover crop (CC) mixtures and monocultures, mowing and glyphosate applications for weed management in a citrus orchard in Western Greece. A randomized complete block design (RCBD) was established with nine treatments replicated four times. Treatment list included: oat, vetch, and white mustard CC monocultures, two-species CC mixtures of oat + hairy vetch and white mustard + hairy vetch, a three-species CC mixture of oat + white mustard + hairy vetch, two mowing passes, two glyphosate applications (720 g a.e. ha⁻¹), and an untreated control. CCs were established in autumn and flail-mowed at late flowering. Oat + white mustard + hairy vetch mixture produced the highest amounts of biomass followed by white mustard + hairy vetch. Glyphosate provided satisfactory control of winter weeds. A polynomial regression indicated that winter weed biomass decreased by increasing the number of species in the CC ($p \leq 0.001$). The three-species CC mixture suppressed weed growth by 87% compared to the control. Glyphosate provided excellent control of summer weeds while the three-species CC mixture suppressed summer weeds more than two-species CC mixtures and CC monocultures. A reciprocal linear regression was observed between CC biomass and summer weed biomass ($p \leq 0.001$). Such results have also been reported by other studies under similar soil-climatic conditions. Our results suggest that diversified CC mixtures can potentially contribute to the development of Integrated Weed Management (IWM) practices in important perennial crops in the Mediterranean region.

SE12 C04

CULTURAL PRACTICES FOR WEED MANAGEMENT IN KENAF CROP (*HIBISCUS CANNABINUS* L.)

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Kenaf (*Hibiscus cannabinus* L.) is a promising biomass crop. In order to evaluate cultural practices for weed management in kenaf, a two-factor field trial in a Randomized Complete Block Design (RCBD) was conducted at the Agricultural University of Athens. Two crop row spacings, 30 cm and 60 cm, were assigned to the main plots. Three treatments (with three replicates) were assigned to the subplots: conventional sowing (CS), stale seedbed with glyphosate application (720 g a.e. ha⁻¹) 15 days after seedbed preparation (SSB 1), and stale seedbed with glyphosate application (720 g a.e. ha⁻¹) 30 days after seedbed preparation (SSB 2). "Conventional sowing" refers to sowing the day after seedbed preparation. The term "stale seedbed with glyphosate application" means that the non-selective herbicide was used to control weeds that



SE12 C06

LONG-TERM STUDIES ON IWM EFFECTIVENESS IN MAIZE

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The Integrated Weed Management (IWM) has been applied with variable success in many crops since its establishment in 1991. Development of a weed control strategy based on a combined application of mechanical, biological, ecological and chemical measures, makes a system efficient and ecologically safe. The implementation of an IWM strategy without side effects on the profitability and the agroecosystem is still a challenge today. The IWM program is based on a few general principles: agronomic practices that limit the introduction and spreading of weeds, support the crop competitiveness over weeds, and use practices that keep weeds 'off balance'. Regarding the IWM promotion, work on the education of producers to encourage a long-term approach to weed control and introduce subsidies for to IWM application is required. Thus, research at the Maize Research Institute Zemun Polje was aimed to develop, test and promote efficient IWM. The long-term investigations are focused on a new approach highlights and contribution of combined application of crop rotation and lower rates of herbicides, different soil tillage systems, fertilizers and cover crops in reduction of maize weed infestation. After the application of the recommended and 0.5 of recommended rates of pre-emergence herbicides in the two- and three-crop rotation, the biomass of weeds in maize was significantly reduced compared to the continuous cropping and non-treated control, while the total biomass of weeds was significantly lower in conventional tillage than in reduced and no-tillage, with no significant effect of fertilization level. Maize continuous cropping also favoured perennial weed species over annuals, which is common for intensive production. Regarding alternative practices, different cover crops successfully reduced number of weeds and its dry mass in sustainable farming system in comparison to conventional and extensive farming. Those measures subsequently increase maize productivity parameters such as grain yield and its stability over time.

SE12 001

HERBICIDAL CONTROL OF WEEDS IN MAIZE HYBRIDS IN NORTHWEST REGION OF PAKISTAN

Khan R.

Department Of Agriculture, Bacha Khan University, Charsadda Pakistan, Charsadda, Pakistan

Recently the farming community of northwest area of Pakistan starts growing hybrid maize instead of traditional maize varieties due to its high production. But, meanwhile they are facing some serious issues regarding the pest control in these hybrids particularly the weeds issue. Still there are no proper recommendations for herbicides in the area and the research is still under process looking for recommendations. For this purpose, a field study was conducted to test different weedicides alone and in combinations against weeds in different maize hybrids. The tested weedicides included (Pendimethalin, S-Metolachlor, S-Metolachlor + Mesotrione +

Atrazine, Nicosulfuron, Halosulfuron + Mesotrione + Atrazine and Mesotrione + Atrazine. The maize hybrids Poiner-3025, Petal-CS200 and 70-Plus were evaluated against the stated herbicides in the field. The results showed both weedicides and maize hybrids gave significant responses for the recorded parameters. Among the weedicides, Nicosulfuron gave minimum weed density of 26.7 weeds m⁻² followed by Halosulfuron+Mesotrione+Atrazine gave 55.1 weeds m⁻² as compared to control that gave a weed density of 302.9 m⁻². Due to better control of weeds the same herbicide i.e., Nicosulfuron gave the tallest maize plants (186.1 cm) and maximum number of grains/ cob (398.3). Among different maize hybrids Petal-CS200 performed better and gave maximum plant height (156.7 cm), Number of ears plant (1.4) and 1000 grains weight. Hence, from the instant study is has been concluded that the weedicide Nicosulfuron performed better among the herbicides combinations as a results it also improved the production of maize hybrids. Further trials are recommended at different locations to confirm the present findings before recommending to the farming community of the area.

SE12 002

LUXIMO®: CONFIRMATION OF FAT AS SITE OF ACTION AND ITS IMPACT ON THE CONTROL OF RESISTANT WEEDS IN WINTER WHEAT

Johnen P.¹, Campe R.¹, Schlaefer S.¹, Sievernich B.¹, Hutzler J.¹, Hendriks J.², De I.¹, Betz M.³, Cornaciu I.⁴, Gerhardt A.¹, Pollmeier M.¹, Witschel M.³

1. BASF SE, Limburgerhof, Germany, 2. BASF Metabolome Solutions GmbH, Berlin, Germany, 3. BASF SE, Ludwigshafen, Germany, 4. ALPX-Services, Grenoble, France

Understanding the mode and site of action of a herbicide is key for its efficient development, the evaluation of its toxicological risk, efficient weed control and resistance management. Through a combination of chemo-proteomics, direct binding and inhibition assays, co-crystallization and by assessment of changes in the physiology and the metabolism, we identified the mode of action (MoA) of LUXIMO® in the lipid biosynthesis with acyl-ACP thioesterase (FAT) as the site of action (SoA). We will present new data on the analysis of downstream effects triggered by LUXIMO® on the cell biological, fatty acid and lipodomic level, that corroborate FAT inhibition as MoA and present herbicidal effects of LUXIMO® as clearly different to other herbicides that are used for the selective weed control in wheat. The discovery of a hidden cluster of FAT inhibitors formerly classified in the HRAC class O ('Unknown Mode of Action') further shows the unique features of LUXIMO®. A herbicide with a new SoA comes along with the potential to manage hard to control grass weeds. The analysis of hundreds of blackgrass and rye grass biotypes with known target and non-target site resistances showed that LUXIMO® indeed is highly effective for the control of resistant weeds. Together, the presented data shows that LUXIMO® is a new and efficient tool for the grass weed control in cereals.



CONFERENCE I

18.30-21.00 Concurrent Session **13**

Incorporating Indigenous Knowledge into Plant Protection Science

Chair: **Federico Tomasetto** (Agresearch, New Zealand)

Nick Roskrige (School of Agriculture and Environment, Massey University, New Zealand)

Trevor Jackson (Agresearch, New Zealand)

SE13 C01

A DUOPOLY TRANSDISCIPLINARY APPROACH BETWEEN MODERN SCIENCE AND INDIGENOUS KNOWLEDGE IS A POWERFUL TOOL FOR PLANT PROTECTION STRATEGIES

Federico Tomasetto

Agresearch, Lincoln, New Zealand

Globally, there are calls for increased integration of Indigenous Knowledge into decision-making relating to culturally sensitive food production and its security. Given this push, plant protection scientists and practitioners are starting to collaborate with Indigenous Knowledge-holders through transdisciplinary approaches. However, with this, there is some complexity. Indigenous Knowledge can often be unique and specific to quite small indigenous groups and it therefore may exist in multiplicity of forms. Further, the holders of such knowledge may be suspicious of modern science (and certainly its institutions), thus precluding the opportunity for the advancement of the management of plant pathogens and diseases. Conversely, the science community often lacks the expertise to recognise the valuable aspects of Indigenous Knowledge. Given this situation, the application of transdisciplinary approaches whereby Indigenous peoples partner with other researchers (including but certainly not only scientists) to provide results and solutions that are truly co-owned. Such a transdisciplinary approach can well apply to both small or large ecosystems, in all cases offering more holistic understanding of cumulative impacts and interconnecting systems. Part of this approach offers the potential for new data sources and observations that can be incorporated into quantitative models. Significantly, transdisciplinarity can be used to make regulatory decisions that are acceptable to the participating partners. Thus, communities may feel that their input is treated respectfully, and data sovereignty maintained. Broadly, the embracing of diverse ontological and epistemological approaches through such multipartner collaboration points to durable solutions and desired outcomes. In our presentation, we will explore how New Zealand researchers using transdisciplinary approaches are working with Mātauranga Māori (Māori knowledge) to establish effective plant pathogen surveillance (MMFS) that cause myrtle rust (*Austropuccinia psidii*) and kauri dieback (*Phytophthora agathidicida*).

SE13 C02

PLANT PROTECTION IN THE SOUTH PACIFIC

Nick Roskrige

Massey University, Palmerston North, New Zealand

The Pacific Ocean is a continent in its own right with a huge diversity in island nations, all dependant on local food production systems to add to the oceans' bounty. Achieving food security and food sovereignty through resilience is the desire of our South Pacific communities including Māori in New Zealand. External factors such as border biosecurity, climate change, land degradation and piracy of indigenous plant knowledge are recognised as leading issues affecting indigenous communities across the Pacific. Aside from commercial piracy of traditional knowledge, there is an increasing pressure on Indigenous communities and their food systems due to various biological pressures on crops and systems. As an example, the impact of climate change on pest and disease populations is becoming harder to manage without expensive inputs or technology and introduced biological threats at the 'border' are a continual issue across the whole of the South Pacific. The climatic extremes are impacting systems more regularly and increasingly out of season. The FAO "recognizes that control over the food system needs to remain in the hands of producers and is clearly focused primarily on small-scale agriculture of a non-industrial nature, preferably organic" (Gordillo & Jeronimo, 2013). This aligns to the South Pacific experience. However, local communities and food producers need support from the international science sector through identifying and developing systems inputs which recognise their local strengths and weaknesses, guarantees plant and crop health factors and contributes to achieving food security for all.

Gordillo, G & Jeronimo, O. M., (2013) Food Security and Sovereignty: base document for discussion, FAO (Food & Agriculture Organization of the United Nations), Rome.



SE13 C03

CULTURAL CHARACTERISATION THROUGH 'KNOWLEDGE INTEGRATION' AS A TOOL IN PROMOTING, PROTECTING, AND CONSERVING TRADITIONAL CROPS IN AOTEAROA (NEW ZEALAND)

Simon Apang Semese

Bioprotection Aotearoa, Lincoln University, Canterbury, New Zealand

Science has always been in the forefront of research as a means of mitigating, managing and finding critical answers to issues of interest. Plant (crop) science is one of the many areas with ongoing research due to population increase, food security, climate change and globalisation to name a few. However, focus in science underpins the traditional know-how of indigenous people far and wide. Neither of the two-knowledge systems of science and indigenous local knowledge (ILK) is supreme and so a systems change is imminent. This can be achieved through knowledge integration describe by Bohensky & Maru (2011) as 'the process of synthesizing multiple knowledge models (or representations) into a common model (representation)' and 'the process of incorporating new information into a body of existing knowledge.' They go on surmise that... 'this definition encapsulates the very dilemma knowledge integration faces in the arena of indigenous knowledge and science: whose knowledge is "new," whose is "existing," and who decides?'. Therefore, cultural characterisation through 'knowledge integration' and understanding of traditional crops in this case taewa (Māori potatoes) can be an added layer to not only protecting the crop from pests/diseases issues but also the conservation of crop biodiversity.

Bohensky, E. & Maru, Y. (2011) *Indigenous Knowledge, Science, and Resilience: What Have We Learned from a Decade of International Literature on "Integration"*, Ecology and Society 16(4): 6. <http://dx.doi.org/10.5751/ES-04342-160406>

SE13 C04

THE PACIFIC RESPONSE TO INVASION OF THE COCONUT RHINOCEROS BEETLE (CRB) (*Oryctes rhinoceros*) IN THE WESTERN PACIFIC ISLANDS

Trevor Jackson¹, Sean Marshall¹, David Tenakanai², Eremas Tade³, Emad Jaber⁴, Francis Tsatsia⁵, Armstrong Sam⁶, Mark Ero⁷, Tanya Robinson⁸

1. AgResearch, New Zealand, 2. NAQIA, Papua New Guinea, 3. KIK, Papua New Guinea, 4. PNG-OPRA, Papua New Guinea, 5. Biosecurity Solomon Islands, 6. Biosecurity Vanuatu, 7. LRD SPC, Fiji, 8. MFAT New Zealand

In 2007 an unusual beetle was found in a swimming pool in Guam. It was identified as coconut rhinoceros beetle (CRB) (*Oryctes rhinoceros*) and rapidly spread around Guam causing heavy damage to coconut palms. Genetic analysis showed that the beetle belonged to a new variant of the beetle (CRB-G, Clade I^b) related to those that have been found in other recent highly damaging outbreaks in Papua New Guinea (PNG), Hawaii, Solomon Islands (SI), and Vanuatu. An emergency response was called for by CRB scientists and stakeholders from the affected countries and supported by MFAT New Zealand. The Pacific Response to CRB-G aims to contain and control the new variant of CRB to reduce damage and support the livelihoods of Pacific islanders. Initial steps have

been to work with local partners with links to industry and communities to define the problem and organise response. The invasive variant is now well established on PNG, SI and Vanuatu, causing heavy damage and, in all cases, has arrived without the biocontrol *Oryctes nudivir*. An awareness programme has alerted communities to the danger and led to prompt reporting of fresh outbreaks and advice to prevent spread and clean-up breeding sites. The variant appears resistant to the traditionally used strains of the biocontrol virus. New strains of the virus are being sought and tested and alternatives, like the biocontrol fungus *Metarhizium majus*, being tested in the field. Local communities are involved for improved surveillance and selection of appropriate controls for implementation in local conditions. A coordinated response is necessary to contain the pest, dampen the impact and prevent the spread to vulnerable communities reliant on coconuts throughout the Pacific islands.



BANQUET

08.30-11.00 Concurrent Session **14**

Microbiomes and their Role in Plant Pathology

Chair: **Corné Pieterse** (Plant-Microbe Interactions Group, Utrecht University, The Netherlands)

Ioannis Stringlis (Agricultural University of Athens, Greece)

SE14 C01

THE ROOT MICROBIOME AND PLANT HEALTH

Corné M.J. Pieterse and the Utrecht Plant-Microbe Interactions team

Plant-Microbe Interactions, Utrecht University, Padualaan 8, 3584 CH Utrecht, the Netherlands

In nature, plants are attacked by a multitude of pathogens and pests that cause major crop losses in agriculture. To protect themselves, plants can activate a sophisticated immune system. Moreover, plants nurture a large community of root-associated microbiota, which provide them with essential services, such as enhanced nutrient uptake, growth promotion, and protection against pathogens. Research in the Plant-Microbe Interactions group at Utrecht University is focused on understanding plant-beneficial functions encoded by the root microbiome and the role of plant genes facilitating these functions. Recently, we demonstrated that upon foliar infection with the downy mildew pathogen, *Arabidopsis* roots recruit a consortium of synergistic microbes to their rhizosphere that in turn trigger an induced systemic resistance (ISR) in the whole plant body and create a so-called soilborne legacy of microbiota in the soil that also protects a next generation of plants against pathogen infection. We discovered that coumarins in root exudates play an important role in the chemical communication between plant roots and the root microbiome and in the onset of ISR. With our research we aim to provide a rational basis for developing sustainable microbiome-based strategies for disease resistance in next-generation crops that maximize profitable functions from the root microbiome. For instance, we developed a microbiome-informed prediction model that is based on the potato seed tuber microbiome profile and can accurately predict the vigor of seed potatoes in the next growing season. Understanding the mechanistic basis of plant-microbiome interactions will provide a firm knowledge basis for the development of future microbiome-assisted crop systems that produce more with less input of fertilizers or pesticides.

a class of secondary metabolites, coumarins, can affect the assembly of the root microbiome and favour the growth of beneficial microbes over that of phytopathogenic ones. Other than coumarins, different structural (cutin, suberin, callose and lignin) and chemical (camalexin and glucosinolates) defense components could act as gatekeepers to maintain a balanced plant-microbiome interaction. To understand the role of these components, we initially screened a range of *Arabidopsis* mutants *in vitro*, with the aim of studying their role in the interaction between *Arabidopsis* and the model beneficial rhizobacterium *Pseudomonas simiae* WCS417. We found that some mutants had reduced beneficial effects and impaired root colonization by the bacterium. Transcriptomic experiments revealed that camalexin affects bacterial chemotaxis, while other defense components influence a balanced growth-defense trade-off during colonization. We also studied the effect of these defense components on plant-associated microbiome assembly. We grew mutants in soil and harvested unplanted soil, rhizosphere soil and roots for microbiome analysis to identify microbiota enriched in each defense mutant compared to wild-type and in which compartment. Our data highlight a compartment-specific effect of defense components on the microbiome and the enrichment or depletion of different microbial taxa when the function of a defense component is misregulated. Our goal is to better understand how these defense components contribute to the plant-microbiome homeostasis and use this knowledge to develop microbiome-assisted agricultural strategies.

SE14 C03

THE ROLES OF FUNGAL EFFECTORS IN MICROBIOTA MANIPULATION

Hanna Rovenich, Nick C. Snelders, Ciaran Kelly, Jan Wieneke, Jinyi Zhu, Yukiyo Sato, Anton Kraege, Eva Schnell, Heidrun Haeweker, Zoe Prockl, Bart P. H. J. Thomma

Institute for Plant Sciences, University of Cologne, Cologne, Germany

Fungi thrive in virtually any environment on this planet where they typically encounter a myriad of other microbes. Many fungi live freely as decomposers of organic material, whereas others engage in various types of symbioses with multicellular human, animal, insect or plant hosts. Fungal survival in these niches depends on well-studied manipulation strategies of host physiology mediated by the secretion of effector proteins to facilitate colonization. However, we increasingly appreciate that during colonization fungi not only interact with their hosts but also with host-associated microbiota. The soil-borne plant pathogenic fungus *Verticillium dahliae*, for instance, employs effector proteins that display selective antimicrobial activity. These effectors are secreted in a

SE14 C02

PLANT-MICROBIOME COMMUNICATION IN THE RHIZOSPHERE

Ioannis A. Stringlis^{1,2}, Jiayu Zhou^{1,3}, Melissa Uribe-Acosta¹, Ronnie de Jonge¹, Corné M.J. Pieterse¹

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Plant roots interact with a mesmerizing diversity of microbiota in the soil. Despite the presence of pathogenic or potentially harmful microbes in their microbiomes, plants in nature are usually healthy. Previously, we have found that



life stage-specific manner to target bacterial and fungal antagonists in various host tissues as well as in soil microbial communities. Considering that such effectors are not only encoded in the genomes of (plant) pathogens but also in those of mutualists, endophytes, and even free-living saprotrophs, it is conceivable that effectors with antimicrobial activities may have evolved in fungal ancestors that colonized terrestrial and aquatic habitats before the emergence of multicellular hosts. These early microbe-manipulating effector-like molecules may have been maintained and subsequently functionally been co-opted as fungi adapted to novel niches over time. Therefore, microbiota-manipulating effectors are likely to act in diverse contemporary contexts where fungi occur.

SE14 C04

FINDING BALANCE IN THE PLANT MICROBIOME

Omri Finkel, Amal Mhalwes, Raphael Sapir

Institute of Life Sciences, The Hebrew University of Jerusalem, Israel

Plants rely on sophisticated defense and immunity mechanisms to protect themselves from pests and pathogens. It is now established, however, that the first layer of defense is provided to the plant by other organisms – the plant microbiota. Accumulating data suggest that the protection provided by the microbiota constitutes not just the plant's first line of defense, but possibly its most potent one, as disruptions to the microbiota render plants susceptible to otherwise asymptomatic infections. To understand how this layer of defense is deployed, we have been applying a realistically complex and fully tractable plant-soil-microbiome microcosm. This system provides a platform for the discovery of novel plant-beneficial traits, which only emerge within a microbial community context. In order to identify which components of the plant microbiota are critical for plant defense, we deconstructed this microcosm top-down, removing different microbial groups from the community to examine their effect on the plant. Applying this method reveals a high redundancy in the microbial protection of plants. Taxonomically disparate microbial consortia have similarly beneficial effects. Further dissection of the microbiota however, started to reveal unique roles for different microbes. In previous work, we discovered the protective role of the genus *Variovorax*, which removed excess microbial auxins from the rhizosphere. Here, we have applied a community deconstruction approach to understand which members of the root microbiota contribute most to its protection from two very different pathogens: The bacterial leaf pathogen *Pseudomonas syringae* and the necrotic fungal pathogen *Botrytis cinerea*. This approach revealed that each pathogen is controlled by different elements within the microbiota. While *P. syringae* is controlled mostly by the genus *Bacillus* (phylum Bacillota), *B. cinerea* is controlled mostly by Betaproteobacteria (phylum Pseudomonodota). Using RNA-seq, we further show that the plant, defense-related genes in particular, responds very differently to the presence of Bacillota and Pseudomonodota in the root microbiota. We conclude that microbial protection of plants is layered, and that labor is divided among different members of the community. These findings also demonstrate the power of microcosm deconstruction as a tool in unraveling the complexities of plant-microbiome interactions.

SE14 C05

PLANT MICROBIOMES: FROM MOLECULAR MECHANISMS TO ADDRESSING PHYTOPATHOLOGICAL ISSUES IN AGRICULTURE

Kalliope K. Papadopoulou

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Beneficial microbiota in the rhizosphere can have a diverse range of symbiotic relationships and can support, enable and enhance diverse functions and the adaptability of their host-plants under challenging conditions. The mechanistic details that underlie these capabilities are not well understood. The endophytic fungus *Fusarium solani* strain K, F_sK, has been used as a case study to elucidate the mechanistic details of the establishment and function of the beneficial microbe in various host plants species as well as to develop microbial inocula solutions for plant protection. Plant and fungus responses were studied by integrating omic approaches. Mutant plant lines and genetic reporter systems were used. Expression of genes were followed by molecular analysis and microscopy. The beneficial phenotypes against pathogens and pests in multi-trophic systems was recorded at low to higher tier, up to the greenhouse and field level. Major modifications occur both in the plant and fungal side at the molecular and at the cellular level. Gene expression and metabolic profiles strongly indicate the involvement of well-known hormonal networks, highlight differences with pathogens and supports novel RNA-mediated communication pathways. These results were used for a data-driven application of single and low-diversity inocula as alternative means of plant protection in tomato crop plants with positive outcomes. Deciphering the genetic factors and the environmental context are indispensable parts for the development and application of root microbiota as microbial inocula in agricultural settings towards a more sustainable agriculture.

SE14 C06

FROM SUPPRESSIVE COMPOST TO TARGETED INOCULANTS: SYNTHETIC MICROBIAL COMMUNITIES PROMOTE TOMATO GROWTH AND DISEASE CONTROL

Maria-Dimitra Tsolakidou¹, Ioannis A. Stringlis², Natalia Fanega-Sleziak¹, Stella Papageorgiou¹, Antria Tsalakou¹, Iakovos S. Pantelides¹

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Soilborne pathogens such as *Fusarium oxysporum* and *Verticillium dahliae* represent a hard-to-control threat for many economically important crops. Suppressing composts represent an eco-friendly approach to control soilborne plant pathogens and enhance plant growth. This study aimed to design functional synthetic microbial communities (SynComs) promoting tomato health by harnessing insights from a previous exploration of suppressive compost microbiota. Two SynComs were constructed using representative bacteria isolated from the rhizosphere of tomato plants grown in a suppressive compost. SynCom1 consisted of a taxonomically



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diverse bacterial mix, while SynCom2 included only *Bacillus* strains. Pot experiments evaluated the effects of SynCom application via two methods: root drench in sterile substrate and seed biopriming. Root drench application revealed that SynCom1 benefitted tomato growth in pots, while SynCom2 promoted tomato growth but also suppressed disease symptoms caused by *Fusarium*. Seed biopriming with both SynComs promoted tomato growth and protected plants against both *F. oxysporum* f. sp. *lycopersici* and *V. dahliae*. These findings highlight the critical role of microbial community composition in influencing tomato health. This study also demonstrates the potential of using microbiota deriving from suppressive compost to design targeted SynComs as tomato inoculants. Characterizing key bacterial traits can facilitate the development of defined, controllable microbial communities with consistent plant growth promotion and disease suppression capabilities in tomatoes.

SE14 001

INTERPLAY BETWEEN AMARYLLIDACEAE ALKALOIDS, THE BACTERIOME AND PHYTOPATHOGENS OF LYCORIS RADIATA

Zhou J.¹, Stringlis I.^{2,3}, Wen J.¹, Liu Y.¹, Xu S.^{1,4}, Wang R.^{1,4}

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Amaryllidaceae alkaloids (AAs) are unique secondary metabolites in Amaryllidaceae plants. Because of their various structures and activities, it is important to understand the functions of AAs in the interplay between plants and the beneficial and pathogenic microbiota. Here, we studied the interplay between AAs, the bacteriome and phytopathogens of *Lycoris radiata*, a traditional Chinese medicinal plant containing high amounts of AAs. Firstly, the relationship between AAs and bacterial composition in different tissues of *L. radiata* was studied. *In vitro* experiments revealed that AAs have varying levels of antimicrobial activity against endophytic bacteria and pathogenic fungi. Using bacterial synthetic communities with different compositions, we observed a positive feedback loop between bacteria insensitive to AAs and their ability to increase accumulation of AAs in *L. radiata*, especially in leaves. This may allow insensitive bacteria to outcompete sensitive ones for plant resources. Moreover, the accumulation of AAs enhanced by insensitive bacteria could benefit plants when challenged with fungal pathogens. Next, the influences of AAs on the composition of root associated bacteriome of *L. radiata* was studied. Analysis based on high throughput 16S rRNA gene amplicon sequencing showed that *Pantoea* species were highly enriched in and around the roots of *L. radiata*. Subsequent *in vitro* experiments verified that AAs could promote the swimming of *Pantoea* strains isolated from the rhizosphere, rhizoplane or endosphere of *L. radiata*. In turn, some *Pantoea* strains could increase the fresh weight, carotenoid accumulation and root viability of *L. radiata*, as well as could directly antagonize fungal pathogens. These indicated that AAs released by roots of *L. radiata* could recruit certain bacterial taxa from soil, which were able to benefit the plant growth and adaptation. In summary, this

study highlights the functions of alkaloids in plant-microbe interactions, opening new avenues for designing plant microbiomes that could contribute to sustainable agriculture.

SE14 002

THE SOIL BACTERIAL COMMUNITY REGULATES GERMINATION OF PLASMIDIOPHORA BRASSICAE RESTING SPORES RATHER THAN ROOT EXUDATES

Xiaorong Z., Sarenqimuge S., Von Tiedemann A.

University Goettingen, Goettingen, Germany

Clubroot, caused by *Plasmodiophora brassicae*, is a severe soil-borne disease that threatens the production of cruciferous crops worldwide. The disease is difficult to control and therefore effective sustainable control methods are required. An important step in the life cycle of *P. brassicae* is the germination of resting spores, which is required for root infection because dormant spores cannot infect plants. Previous studies reported that root exudates can trigger *P. brassicae* resting spore germination, thus enabling a targeted attack of *P. brassicae* on host plant roots. However, we found that native root exudates collected under sterile conditions from host or non-host plants cannot stimulate the germination of sterile spores, indicating that root exudates may not be direct stimulation factors. Instead, our studies demonstrate that soil bacteria are essential for triggering germination. Through 16S rRNA amplicon sequencing analysis, we found that certain carbon sources and nitrate can reshape the initial microbial community to an inducing community leading to the germination of *P. brassicae* resting spores. The stimulating communities significantly differed in composition and abundance of bacterial taxa compared to the non-stimulating ones. Several enriched bacterial taxa in stimulating community were significantly correlated with spore germination rates and may be involved as stimulation factors. Based on our findings, a multi-factorial 'pathobiome' model comprising abiotic and biotic factors is proposed to represent the putative plant-microbiome-pathogen interactions associated with breaking spore dormancy of *P. brassicae* in soil. This study presents novel views on *P. brassicae* pathogenicity and lays the foundation for novel sustainable control strategies of clubroot.

SE14 003

DECIPHERING THE EFFECTS OF AGRONOMICAL PRACTICES ON ASPERGILLUS INCIDENCE AND CARPOSPHERE'S MICROBIAL COMMUNITIES OF GRAPEVINE

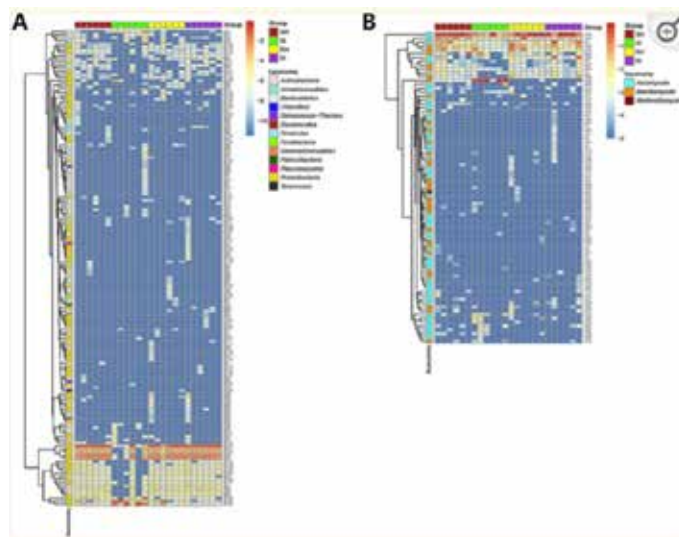
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Going through the new transitioning era of the "European Green Deal", the search for alternative, non-chemical, disease control methods is essential. *Aspergillus* bunch rot is considered one of the most important diseases of grapevines resulting in severe yield losses and, major qualitative

deterioration of grape products due to the production of mycotoxins. We investigated, in a two-year field study, the impact of agronomic practices like defoliation to enhance grape microclimate (DF), pruning method to reduce grape bunch density (LBD), and irrigation cut-off (NIR), at three developmental stages of grapevine (Pea size berry, Veraison, and Harvest), on (i) grape composition (titratable acidity, pH, and total soluble solids), (ii) on the frequency of occurrence of *Aspergillus* on grape berries and (iii) on the overall composition of grape carposphere microbiome. The density of *Aspergillus* on grape berries was significantly reduced by the applied management practices (DF, LBD, NIR). Amplicon sequencing analysis showed that both the phenological stage and the agronomic practices employed (particularly NIR and DF) imposed significant changes in the α -diversity and β -diversity of the grape carposphere bacterial and fungal communities. The NIR, LBD, and DF treatments which supported lower *Aspergillus* populations, network analysis revealed negative co-occurrence patterns between *Aspergillus* and several bacterial genera (*Streptococcus*, *Rhodococcus*, *Melittangium*) reported to have antifungal properties suggesting potential natural attenuation mechanisms for the control of *Aspergillus*. Overall, our study (i) showed that the application of halting of irrigation and thinning of leaves and grape bunches, reduce the occurrence of *Aspergillus* and hence the incidence of *Aspergillus* Bunch rot disease and (ii) identified preliminary evidence for interactions of *Aspergillus* with members of the epiphytic grape bacterial communities that might be involved in the suppression of *Aspergilli*, an observation which will be further pursued in following studies in the quest for the discovery of novel biological control agents.

aimed at developing strategies against wheat dwarf bunt. Collectively, this research advances the understanding of the microbial assembly of wheat seeds upon exposure to fungal pathogen infection.



Heatmaps of enriched bacterial and fungal genera by amplicon sequencing. (A) Heatmap of bacterial genera in all groups. (B) Heatmap of fungal genera in all groups.

SE14 004

MICROBIOME SIGNATURE OF ENDOPHYTES IN WHEAT SEED RESPONSE TO WHEAT DWARF BUNT CAUSED BY *TILLETIA CONTROVERSA* KÜHN

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Institute of Plant Protection, Chinese Academy of Agriculture Sciences, Beijing, China

Wheat dwarf bunt leads to the replacement of seeds with fungal galls containing millions of teliospores of the pathogen *Tilletia controversa* Kühn. This is the first study on the microbiome signature of endophytes in wheat seed response to wheat dwarf bunt caused by *Tilletia controversa* Kühn, and explore some antagonistic microbes used for biological control of the disease. Based on a combination of amplicon sequencing and isolation approaches, we analyzed the seed microbiome signatures of endophytes between resistant and susceptible cultivars after infection with *T. controversa*. We found 14 overlapping species by both methods; we detected 128 fungal species only by amplicon sequencing, 56 only by isolation, and 5 species by both methods. The results indicated that resistant uninfected cultivars hosted endophytic communities that were much more stable and beneficial to plant health than those in susceptible infected cultivars. The susceptible group showed higher diversity than the resistant group, the infected group showed more diversity than the uninfected group, and the microbial communities in seeds were related to infection or resistance to the pathogen. Some antagonistic microbes significantly suppressed the germination rate of the pathogen's teliospores, providing clues for future studies



SKALKOTAS

08.30-11.00 Concurrent Session **15**

Components of IRM Programs in an IPM Framework

Chair: **Silvana Paula-Moraes** (University of Florida, USA)

SE15 C01

BEYOND PEST CONTROL: TOPICS FOR THE IMPROVEMENT OF IPM AND IRM OF LEPIDOPTERAN PESTS ASSOCIATED WITH ROW CROPS

Silvana Paula-Moraes

UF/IFAS West Florida Research and Education Center

A challenge that remains for high input systems in row crops is the occurrence of pests that reach high population densities and demand management. In Lepidoptera, several species from the Superfamily Noctuoidea are critical pests worldwide. The capacity of economic impact is related to the damage capacity of the larval stage, favorable conditions for the development of several generations per year, and the ability to disperse and colonize new host plants in the succession of crops in the agricultural landscape. Insecticides are a common tactic in most intensive systems because of their fast action, ease of application, and relative economic viability, at least in the short term. In addition, the commercial adoption of transgenic plants expressing insecticidal proteins from *Bacillus thuringiensis*, commonly known as Bt crops, is now prevalent in corn and cotton in several countries, such as the U.S., Brazil, and South Africa. The reasons for rapid farmer acceptance can be explained by the high level of control provided by Bt technology for important lepidopteran pest species. However, there is a critical need to incorporate additional knowledge to promote environmentally sound IRM programs in an IPM framework. In this session, speakers will present and discuss aspects of pest ecology and toxicology, management tools such as biological control, grower behavior, dissemination of information through Extension programs as a viable approach for the management of economic lepidopteran pests, and the role of stakeholder support for the success of management programs.

showing the importance of the variability among years and within the season on susceptibility to commonly planted Bt traits in maize. Data will be presented to show declines in susceptibility to Cry toxins expressed in Bt maize in recent years in *H. zea* populations in the southern U.S. The challenges of implementing IRM practices for *H. zea* in maize will be discussed, as the durability of Bt traits targeting *H. zea* is compromised by low planting areas of structured non-Bt corn refuge.

SE15 C03

MANAGING THE CHANGING LEPIDOPTERA MAIZE PEST COMPLEX IN AFRICA: SHORT- AND LONG-TERM STRATEGIES

Johnnie van den Berg

North-west University, Potchefstroom, South Africa

The invasion of Africa by the fall armyworm (*Spodoptera frugiperda*) in 2016 changed the way in which lepidopteran maize pests are managed on the continent. *Spodoptera frugiperda* forms part of a complex of lepidopteran species which include *Busseola fusca*, *Chilo partellus* and *Sesamia calamistis*. Ninety-eight percent of farms in Africa are small-scale (<2 ha), and South Africa is the only country where large-scale systems dominate. Pest management approaches depend on the scale of these maize farming systems and although both systems employ IPM principles, the major differences are the amount and types of agricultural inputs used. Due to fall armyworm being a "new" pest on the continent, IPM programs are in the early stages of development. In small-scale (low-input) systems, yields are inherently low and agroecological methods are recommended for control. These include curative use of biopesticides, handpicking and egg crushing, as well as long-term approaches, e.g., habitat management, host plant resistance, and biological control. While biological and cultural control approaches provide long-term solutions, the use of pesticides and other inputs to improve plant vigor and yield also contribute to pest management, especially in low-yielding environments. The success observed with genetically modified Bt maize, which is only cultivated in large-scale systems in South Africa, may influence future decisions regarding its approval for cultivation in other African countries.

SE15 C02

RESISTANCE EVOLUTION TO BT MAIZE IN THE SOUTHERN UNITED STATES: THE CASE OF THE UBIQUITOUS *HELICOVERPA ZEA*

Francis P.F. Reay-Jones, G. David Buntin, Dominic D. Reisig, and Silvana V. Paula-Moraes

Transgenic maize hybrids that express insecticidal toxins from the bacterium *Bacillus thuringiensis* (Bt) can reduce injury from the corn earworm, *Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae), a ubiquitous pest of corn in the southern United States. While field trials have also shown that *H. zea* is generally not an economic pest of maize, intense selection pressure in Bt maize is driving resistance concerns in Bt cotton, where the insect (known as bollworm) is a major economic pest. This presentation will highlight ongoing research illustrating injury levels and yield of Bt and non-Bt maize in field trials in several southern U.S. states,



SE15 C04

EXTENSION AS AN IPM TOOL: THE LAND GRANT UNIVERSITY PROGRAM AND ITS ROLE IN PEST MANAGEMENT

Katelyn Kesheimer

Department of Entomology and Plant Pathology, College of Agriculture, Auburn University

Land Grant Universities in the United States are public universities with special funding to carry out a particular three-part mission. Teaching, Research, and Extension are all components of the 100+ universities with Land Grant designations. The role of Extension within this system is to provide non-formal education and learning activities to thousands of people throughout the country, including farmers and other residents of rural and urban communities. The impact of Extension programming on agriculture in the U.S. cannot be overstated. It does this through practical, research-based knowledge from the universities delivered to the public to create positive change. Extension entomology programs in the U.S. have taken this same approach to provide integrated pest management resources to growers based on local, unbiased data. This talk will explore the role of entomology in Extension, and discuss future prospects for Extension education.

SE15 C05

FARMERS PERSPECTIVES ON INFORMATION SOURCES AND IRM PRACTICES

Dominic Reisig

North Carolina State University, Plymouth, United States

Pesticide susceptibility is a common-pool resource for farmers. In the southern United States, farmers are required to plant 20% of their total maize acres to non-Bt hybrids to delay pest resistance. However, compliance is lacking due to various reasons. Conditional cooperation, where individuals cooperate based on others' actions, benefits common pool resources in forest systems. We hypothesized that farmers collaborating on drainage management would exhibit higher conditional cooperation, be more likely to plant non-Bt maize refuge, and respond to an emotional appeal. In 2023, we surveyed maize farmers in a Southern U.S. region, measuring conditional cooperation through face-to-face economic experiments. Half received information on non-Bt maize refuge planting, while the other half received this information alongside an emotional appeal. The emotional appeal group showed a 16% increase in refuge planting and a 20% rise in sufficient refuge planting. Belonging to a drainage management group affected game behavior significantly. Compared to moderate contributors, drainage management participants were three times more likely to be conditional cooperators and six times more likely to be altruists. These groups also planted more refuge, although they didn't respond to the emotional appeal. The appeal's impact was seen mainly in individualistic groups. Our findings hold potential implications for pesticide resistance management beyond Bt maize, suggesting the importance of cooperative frameworks in addressing resistance issues.

SE15 C06

SUPPORTING IPM/IRM FOR COST EFFECTIVE AND ENVIRONMENTALLY ACCEPTABLE PRACTICES IN U.S. COTTON PRODUCTION

Sally Taylor

Cotton Incorporated, Cary, NC, USA

Almost three decades following the introduction of transgenic cotton, Lepidopteran pests still challenge cotton grown in the United States (U.S.). Cotton producers and importers value research and Extension programs that provide practical solutions to current and future pest management issues. The mission of Cotton Incorporated's Agricultural and Environmental Research division is to help farmers increase yields, manage costs, and reduce their environmental footprint. One way we accomplish this mission is supporting regional collaborations among entomologists. This presentation briefly summarizes insect control collaborations across the U.S. cotton belt and highlights some of the basic and applied research programs that help to monitor and develop tools to manage these pests.



MC2

08.30-11.00 Concurrent Session **16**

Diseases in Tree Crops and Forests

Chair: **Georgios Vidalakis** (Microbiology & Plant Pathology, University of California Riverside, USA)

Evangelos Vellios (University of Thessaly, Greece)

SE16 001

PROTECTING VEGETATION AGAINST OZONE POLLUTION

Evgenios Agathokleous

School of Ecology and Applied Meteorology, Nanjing University of Information Science & Technology (NUIST), Nanjing 210044, China

Tropospheric ozone (O₃) is a widespread air pollutant, which causes phytotoxicities and suppresses tree productivity and crop yields, necessitating the development of strategies to remediate this problem. In this talk, the most recent and effective methods to protect plants against ozone phytotoxicities will be summarized, with emphasis on exogenous application of ethylenediurea, a synthetic antiozonant. The chemical mode of action will be examined in terms of chemical properties, leaf-level physiology, microbiome, and plant-microbe interactions. Ethylenediurea does not protect plants by adding nitrogen, neither is stomata control a primary mechanism explaining protection against ozone phytotoxicities. Instead, ethylenediurea has also ozone degrading properties, indicating a direct reaction with ozone. Ethylenediurea increases plant leaf area, photosynthetic rate, and growth, while it also protects against ozone-accelerated senescence and impairment of photosynthetic response to changes in light conditions. It also decreases foliar injuries, but changes plant-disease interactions, for instance worsening rust disease in some plants such as poplars. Moreover, ethylenediurea affects the microbiome, such as by modifying the abundance of microbes involved in (de)nitrification and N fixation processes. The mode of action remains unclear, and no universal mechanism may exist. However, as the mechanism appears to be species or genotype specific, in light of its remarkable effectiveness in protecting plants against ozone phytotoxicities, EDU could be used regardless its underlying mechanism. The presentation will conclude by drawing attention to the need of more holistic approaches to enhance plant health under air pollution and climate changes.

SE16 002

MYCOVIRUSES INFECTING THE RED-BAND NEEDLE BLIGHT FUNGUS OF PINE TREES AND CONIFERS

Ioly Kotta-Loizou

School of Life and Medical Sciences / Faculty of Natural Sciences, University of Hertfordshire, UK

Red-band needle blight, also known as pine needle blight or Dothistroma needle blight, is an economically important disease of conifers, particularly pine trees. Red-band needle blight is caused by the phytopathogenic fungus *Dothistroma septosporum*, which has a worldwide distribution. The aim of the study was to investigate the diversity of mycoviruses in *D. septosporum*, together with their role in fungal pathogenesis. To this end, a panel of *D. septosporum* field isolates from the United Kingdom were screened for the presence of mycoviruses using a small-scale double-stranded (ds) RNA extraction protocol. Mycoviruses were then purified, visualised, cloned, sequenced and classified. Virus-free isogenic lines were generated following treatment of virus-infected isolates with the protein synthesis inhibitor cycloheximide. Following mycovirus eradication, the growth and virulence of virus-infected and virus-free isogenic lines were compared in vitro and in planta. Screening for dsRNA revealed the presence of a novel mycovirus in *D. septosporum* belonging to the genus *Alphachrysovirus*, family *Chrysoviridae*, and named *Dothistroma septosporum chrysovirus* (DsCV) 1. The DsCV-1 genome comprised four dsRNA elements, of which three were monocistronic and encoded respectively an RNA-dependent RNA polymerase (RdRP), a capsid protein (CP) and a putative cysteine protease. The second largest dsRNA potentially encoded two hypothetical proteins, one small with no homology to known proteins and one large with homology to the alphachryso-P3 of other alphachrysoviruses. A direct comparison of the growth and virulence of DsCV-1-infected and -free isogenic lines illustrated that DsCV-1 infection reduced host growth and resulted in hypovirulence on pine saplings when fungal virulence was assessed by visual inspection, image analysis and quantitative polymerase chain reaction (PCR). These results illustrate the potential of DsCV-1 for biological control of red-band needle blight in the future.

SE16 003

FROM XYLELLA FASTIDIOSA TO ENTEROBACTERIACEAE: INVESTIGATING NEW BACTERIAL THREATS IN SOUTHERN ITALY

Carluccio G.¹, Vergine M.¹, Sabella E.¹, De Bellis L.^{1,2}, Luvisi A.¹

1. University Of Salento, Lecce, Italy, 2. National Biodiversity Future Center, Palermo, Italy

In Salento, Southern Italy, holm oaks (*Quercus ilex* L.) and olive trees (*Olea europaea* L.) are a consistent component of the landscape and economy. However, since 2013, the epidemic caused by *Xylella fastidiosa*, the causal agent of 'Olive Quick Decline Syndrome' has led to the destruction of the olive sector and the rural landscape. In this context, a phenomenon affecting holm oaks has recently been observed, consisting of a mass decline of oak groves. Research is therefore aimed at understanding the biotic causal agents of this decline and the plant-pathogen interaction to protect the forest heritage. Field surveys were conducted at various locations in the Salento region to assess the prevalence and severity of decline symptoms on oak trees and pathometric scale was used to assess the type and severity of decline observed. In addition, plant tissue samples consisting of twigs, bark fragments and soil, were collected to assess the presence of various pathogens through molecular methods. Analyses revealed, in addition to the occurrence of various pathogenic fungi, the presence of certain bacteria belonging to the *Enterobacteriaceae* linked to certain characteristic symptoms, which can be traced back to 'Acute Oak Decline' an emerging phenomenology, first described in England in 2014. This represents the first finding of such syndrome in the Central Mediterranean basin. Since the pathogenic potential of these bacteria is not fully known, further investigations are necessary to understand what the evolution of this pathology might be in Salento and how those bacteria affect holm oak.

SE16 004

IDENTIFICATION, PATHOGENICITY AND CHEMICAL CONTROL OF CHRYSOPORTHE STEM CANKER DISEASE OF EUCALYPTUS SPP IN MALAYSIA

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1. Forest Pathology Laboratory, Industrial Forest Research Centre, Restoration and Industrial Forest Division, Forest Department Sarawak, Kuching, Malaysia, 2. Laboratory of Forest Pathology and Tree Health, Department of Forestry Science and Biodiversity, Faculty of Forestry and Environment, Universiti Putra Malaysia, Serdang, Malaysia

Forest plantation of *Eucalyptus* and its hybrids is expanding rapidly in the Southeast Asia regions including Malaysia, and poised to replace Acacias as the main species planted. Stem canker disease was discovered infecting *Eucalyptus* species in five regions in Malaysia; Kelantan, Pahang, Sabah, Selangor and Sarawak. Molecular analysis revealed that *Chrysoporthe cubensis* and *Chrysoporthe deuterocubensis* were the causal

pathogens of stem canker disease for *Eucalyptus urograndis* and *Eucalyptus pellita*. Pathogenicity tests were conducted on healthy *Eucalyptus urograndis* and *Eucalyptus pellita*. The result demonstrated that all the isolates from both *Chrysoporthe* species were pathogenic to *E. urograndis* and *E. pellita*, with lesions appearing on inoculated stems and mortality observed in seedlings. A host range test was also conducted by using isolates against several forest plantation species including important dipterocarp tree species and non-dipterocarp tree species. *Neobalanocarpus heimii* seedlings showed a response when inoculated with both pathogens. Evaluation of five permitted fungicides through in-vitro screening and field experiments was then conducted against *C. deuterocubensis* (CHRY 18). Results revealed that prochloraz, manganese chloride, thiram, and mancozeb showed potential as effective chemical controls of *C. deuterocubensis* stem canker on *E. urograndis*. Initially, the presence of *Chrysoporthe* spp. in *Eucalyptus* plantations in Malaysia is poorly described although *Chrysoporthe* spp. has a significant economic importance to the sustainability of *Eucalyptus* plantations worldwide. This study provides new information on the status of *Chrysoporthe* canker disease, especially on the occurrence and effects on *Eucalyptus* plantation, by giving insights on the possible control and management of this key disease pathogen.

SE16 005

DIVERSE ORGANISMS CAUSE LEAF AND STEM DISEASES IN MACADAMIA NURSERY PLANTS

Khan J., Drenth A., Akinsanmi O.

The University of Queensland, Brisbane, Australia

Macadamia is an Australian native tree grown for its edible kernel in tropical and subtropical regions worldwide. Infected plants in the nursery may cause disease problems in commercial orchards, reducing productivity. We surveyed five major macadamia nurseries in Australia for leaf and stem diseases affecting nursery plants in different seasons. From over 1500 plant samples, we identified four leaf diseases, including yellow halo leaf blight, brown leaf blight, leaf anthracnose, and black leaf blight. In addition, graft dieback, stem canker and gall canker were detected. Multiple fungal species cause each disease. Leaf anthracnose was the most prevalent in mature leaves in the four seasons and was associated with eight *Colletotrichum* spp. However, only *C. siamense* and *C. gloeosporioides* sensu lato caused infection in pathogenicity assays. The highest diversity of 11 *Neopestalotiopsis* spp. and two *Pestalotiopsis* spp. were isolated from yellow halo leaf blight lesions. The occurrence of black leaf blight was limited to non-suberised leaves. *Alternaria alternata* species complex (*A. alternata* and *A. tenuissima*) is responsible for black leaf blight when plants are kept under wet conditions for a prolonged period. *Neofusicoccum parvum* and *Ne. luteum* were isolated from leaves with brown leaf blight symptoms and from stem tissues without symptoms. However, four Botryosphaeriaceae species and six *Diaporthe* spp. were associated with stem canker symptoms. *Diaporthe litchicola*



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and *D. australiana* were the most aggressive among the eight *Diaporthe* spp. that caused Phomopsis graft dieback. *Agrobacterium* sp. was isolated from root gall symptoms from one sample, which showed it is uncommon in macadamia nurseries in Australia.

SE16 006

EFFECT OF RAINFALL ON THE DISPERSAL OF SPORES OF *Venturia oleaginea*, THE CAUSAL AGENT OF OLIVE SCAB **Guerrero Páez F¹, Campos Figueras R¹, Romero Martín M¹, Alcázar P², Moral J¹**

1. University of Córdoba (ETSIAM. Agronomy Department. Excellence Unit María de Maeztu. UCOLIVO), Córdoba, Spain, 2. University of Córdoba (Botany, Ecology and Plant Physiology Department. Aerobiology), Córdoba, Spain

The olive tree has been the dominant woody crop in the Mediterranean Basin. Olive scab, caused by the obligate hemibiotrophic fungus *Venturia oleaginea*, is this crop's most critical aerial disease. The pathogen causes extensive defoliation, reducing the photosynthetic capacity and tree production. The pathogen is spread by asexual spores (conidia) dispersed by raindrops. Knowing the influence of rainfall and its characteristics (e.g., duration, intensity, etc.) on the dispersal of the pathogen is essential to identify the timing of disease control. Two experiments were conducted using a rain simulator (a Cornell infiltration gauge) to i) characterize the horizontal dispersal of pathogen conidia and ii) study the conidia vertical dispersal under the tree canopy. Experiments were conducted with highly susceptible olive plants (cv. Meski) with unequivocal and sporulated leaf lesions caused by *V. oleaginea*. In the first, moisture and dry plants were used as an inoculum source, and different slides with Melinex® tape were placed radially on them as conidial traps at 0.40, 0.60, 0.80, and 1 m. Thus, four 1-m plants were subjected to a 12.5 mm/h rainfall for 1 or 2 min. To study the vertical conidial dispersion, nine plants were subjected to rainfall for 0, 2, or 6 min. Immediately after rainfall, four slide trappers were placed under each plant. Experiments were conducted twice. The hydration of the plants was essential for spore release. In general, the density of captured conidia decreased exponentially with distance from the inoculum source, minimum at 1 m. Conidia were captured after 2 and 6 min-duration of rainfall under plant canopy, which points to vertical dispersion of the inoculum. These data suggest that the auto-infection process is dominant in the olive canopy, which agrees with the strong clustered distribution of the disease.

SE16 007

MOLECULAR BIOLOGY OF PHYTOPHTHORA IN FOREST DISEASE MANAGEMENT

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As pollution, climate change, and global pathogen movement intensify, so does the spread of *Phytophthora*, an important genus of plant pathogens, into native ecosystems. There are particular challenges associated with managing diseases in a forest environment. Our research focuses on finding solutions to control *Phytophthora agathidicida*, the causal agent of kauri dieback—a devastating disease affecting the iconic, ecologically, economically, and culturally important kauri trees in New Zealand. We have developed a tool to detect *Phytophthora* in the field and assess *Phytophthora* disease in leaves. We have furthermore discovered a signaling pathway that determines *Phytophthora agathidicida* sporulation and can lead to development of novel targets for disease management. These advancements have the potential to be applied not only to kauri dieback but also to other *Phytophthora* diseases and pathogens affecting trees more broadly.

SE16 008

TWIG CANKER AND SHOOT BLIGHT DISEASE OF ALMOND: UNDERSTANDING THE BIOLOGY OF *Diaporthe amygdali*

Francia C.¹, Beluzán F.¹, Novellón M.³, Vicent A.², Lázaro E.³, Berbegal M.¹, Abad-Campos P.¹, Armengol J.¹

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The fungus *Diaporthe amygdali* is the causal agent of twig canker and shoot blight disease of almond (*Prunus dulcis*) and peach (*Prunus persica*). Symptoms on almond trees in Mediterranean regions include necrotic lesions on shoots, leading to twig withering and defoliation. This pathogen produces pycnidia and a-conidia, primarily infecting almond tissues through various entry points. Studies on the biology of *D. amygdali* are scarce, especially in almond. This study aimed to fill gaps in understanding *D. amygdali* biology on this crop. Experiments were conducted with two almond strains of *D. amygdali* (PHAL 4 and PHAL 45) to assess mycelial growth, pycnidia development, conidia germination, host infection and lesion development under different temperature and wetness regimes. The results obtained in this study showed that mycelial growth of *D. amygdali* was observed from 5°C to 35°C, with the maximum growth rate at 30°C. The highest production of mature pycnidia was observed at 15°C, with a decrease in pycnidia production at higher temperatures. No mature pycnidia were observed below 15°C



or above 30°C. The time required for conidial germination was strongly related to incubation temperature, requiring 24 h to germinate at 10°C, 18 h to germinate at 15°C and less than 6 h for higher temperatures. Infection of the host was possible between 5 and 35°C, being higher with warmer temperatures and longer wetness periods. Lesion growth on almond shoots was highest between 20 and 25°C. These findings improve our knowledge of the pathogen's life cycle regarding different temperature and humidity conditions, and provide useful information for the development of decision support systems to improve disease management strategies in almond crops.

SE16 009

DIVERSITY AND CONTROL OF PHYTOPHTHORA SPECIES ASSOCIATED WITH WIDESPREAD DIEBACK AND MORTALITY OF WILD OLIVE TREES IN SARDINIA, ITALY

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Department of Agriculture - University Of Sassari, Sassari, Italy

Wild olive represents one of the most iconic woody plant species in the Mediterranean basin. It is a slow-growing evergreen tree characteristic of the sclerophyll communities, extremely tolerant to drought, salinity and diseases, thus commonly used as rootstock for grafting cultivated olive varieties. Since March 2022, dieback and mortality of wild olive trees has been observed in central Sardinia (Italy). Affected plants showed leaf chlorosis, wilting, defoliation of the whole crown, shoot blight and epicormic shoots, frequently associated with root rot and necrosis on the feeder roots, suggesting a possible involvement of soil-borne pathogens in the genus *Phytophthora*. Preliminary studies revealed the occurrence of *P. bilorbang* and *P. pseudocryptogea*, and their pathogenicity on wild olive seedlings was demonstrated. Subsequent, more in-dept research showed that the aetiology of these epidemics is more complex with several other *Phytophthora* species involved. Using a baiting technique, eleven *Phytophthora* species belonging to three distinct phylogenetic clades (2, 6 and 8), were isolated from 60% of the 91 rhizosphere soil samples collected around both symptomatic and asymptomatic wild olive trees, and two additional *Phillyrea latifolia* and *Myrtus communis* plants. The identified species include *P. asparagi*, *P. bilorbang*, *P. crassamura*, *P. cryptogea*, *P. inundata*, *P. kelmanii*, *P. oleae*, *P. oreophila*-like, *P. pseudocryptogea*, *P. sansomeana* and *P. syringae*. To confirm Koch's postulates, pathogenicity trials were conducted on 1-year-old wild olive potted trees using the soil-infestation method. Treatments with different fungicides, biostimulants and microbial formulations were undertaken in the field to evaluate their efficacy to reduce the impact of *Phytophthora* root infection. Preliminary results are discussed herein.



MC3

08.30-11.00 Concurrent Session **17**

NLR Guided Strategies for Durable Disease Resistance in Crop Plants

Chair: **Jiorgos Kourelis** (Department of Life Sciences, Imperial College London, UK)

Lida Derevnina (University of Cambridge, UK)

SE17 C01

PIKOBODIES: WHAT DOES IT TAKE TO BIOENGINEER NLR IMMUNE RECEPTOR-NANOBODY FUSIONS

Jiorgos Kourelis, Clemence Marchal, Andres Posbeyikian, Adeline Harant, Sophien Kamoun

Recurrent epidemics caused by plant pathogens pose a significant threat to global food security. Modifying natural components has been the only approach to retooling the plant immune system, but these approaches can be rapidly rendered ineffective by the emergence of new pathogen strains. Made-to-order synthetic plant immune receptors, however, offer a promising solution for tailored resistance to pathogen genotypes present in the field. Here we define four technical and conceptual advances which enabled successful engineering of NLR immune receptor-nanobody fusions, termed Pikobodies, for this purpose. These advances include 1) evolutionary analysis to identify domain boundaries, 2) structural modelling to predict integration boundaries, 3) rapid screening assays using transient immune and disease assays, and 4) further engineering to abolish autoactivity triggered by some nanobody integrations. We show that plant immune receptors can serve as effective scaffolds for nanobody fusions that bind fluorescent proteins (FPs), triggering immune responses and conferring resistance against plant viruses expressing FPs. Since nanobodies can be raised against most molecules, immune receptor-nanobody fusions have the potential to generate resistance against most plant pathogens and pests delivering effectors inside host cells.

previously identified SS15 – a SPRYSEC-type effector derived from the potato cyst nematode *Globodera rostochiensis* – as a potent suppressor of immunity in Solanaceous plants. SS15 targets the function of helper NLRs, namely NRC2 and NRC3, that are central nodes of a complex immune signalling network. Using biochemical and cellular approaches, we demonstrate that SS15 binds to and inhibits NRC2 oligomerization and plasma membrane association, which are critical for immune signalling. To overcome this suppression, we introduced mutations in the SS15-NRC2 binding interface and bioengineered an NRC2 variant that evades inhibition and restores NRC2 function in the presence of SS15. Our work exemplifies how a deeper mechanistic understanding of effector biology can provide valuable insights for developing novel strategies to generate disease resistant crops.

SE17 C03

EVOLUTION IN OVERDRIVE: NLR DIVERSITY GENERATION AND MAINTENANCE

Ksenia Krasileva

Plant And Microbial Biology, Uc Berkeley

Plants rely on innate immune receptors to sense diverse and rapidly evolving pathogens. Remarkable receptor diversity at the population level is essential for the success of this strategy. We have analyzed Nucleotide Binding Leucine Rich Repeat (NLR) innate immune receptor diversity in Arabidopsis and maize and identified several genomic and epigenomic features that correlate with intra-species diversity, predicted mutation rates and modes of selection. We identified that a subset of highly variable NLRs (hvNLRs) show high intra-species diversity, while their low variability paralogs (non-hvNLRs) are conserved between lines. At a population level, the diversity of hvNLRs provides a broad range of new pathogen recognition specificities and in well-annotated Arabidopsis reference accession correlates with proximity to TEs, high transcription and lower CG body methylation. hvNLRs also show elevated levels of both synonymous and non-synonymous nucleotide substitutions, and higher predicted mutation rates. Both hvNLR and non-hvNLRs have specific sites under positive selection while non-hvNLR paralogs are maintained by purifying selection. Our study provides insights into NLR diversity generation and maintenance allowing further research on diversity generation mechanisms in NLRs and NLR engineering.

SE17 C02

CYST NEMATODES COUNTERACT IMMUNITY BY INHIBITING ACTIVATION OF CENTRAL NODES OF A SOLANACEAE IMMUNE RECEPTOR NETWORK

Mauricio P. Contreras¹, **Hsuan Pai**¹, **Muniyandi Selvaraj**¹, **AmirAli Toghani**¹, **David M. Lawson**², **Yasin Tumbas**³, **Cian Duggan**³, **Enoch Lok Him Yuen**³, **Clare E.M. Stevenson**², **Adeline Harant**, **Abbas Maqbool**², **Chih-Hang Wu**⁴, **Tolga O. Bozkurt**³, **Sophien Kamoun**¹ and **Lida Derevnina**^{1†}

1. The Sainsbury Laboratory, University of East Anglia, Norwich, UK, 2. Department of Biochemistry and Metabolism, John Innes Centre, Norwich, UK, 3. Department of Life Sciences, Imperial College, London, UK, 4. Institute of Plant and Microbial Biology, Academia Sinica, Taipei, Taiwan, †Present address: Crop Science Centre, Department of Plant Sciences, University of Cambridge, Cambridge, UK

Plant pathogens deploy effectors that suppress plant immune responses and promote disease. Despite their important role in pathogenicity, our understanding of the mechanisms underlying effector function remain relatively limited. We



SE17 C04

LEVERAGING DANGEROUS MIX AUTOIMMUNITY TO INVESTIGATE HOST-MICROBE INTERACTIONS

Eunyoung Chae

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Plants have evolved an effective immune system in which receptors and other proteins participate to recognize invasive events in the cell. Immune components of an individual plant can be distinct from those of others in a given population, presenting a diverse array of immune repertoire. The diversity in the plant immune system, however, can result in an inadvertent activation of the system in hybrid plants, as a form of hybrid necrosis. Through extensive genetic analyses of hybrid necrosis, mismatched immune components have been discovered to result in autoimmune responses in pairs. These incompatible *DANGEROUS MIX* (DM) interactions often involve genes encoding nucleotide-binding domain leucine-rich (NLR) receptors. *DM* autoimmunity can be triggered by NLR-NLR receptor interactions as well as by interactions between an NLR and its non-NLR partner. These non-NLR partners provide an opportunity to probe cellular events that are being monitored by the intracellular NLR receptors. In this talk, the case study of *DM2h-DM3* autoimmunity will be further presented in detail. To investigate the molecular mechanism of how DM3, an enzyme with the alpha/beta hydrolase fold, activates DM2h, a TIR-NLR, we first determined structures of DM3 variants. The high-resolution cryo-EM structures of DM3 from Col-O and Hh-O revealed that the hybrid necrosis variant from Hh-O has structural differences in a dimer interface within the stable DM3 complex, consisting of six molecules of DM3 in the configuration of a trimer of dimers. Naturally occurring polymorphism found in DM3^{Hh-O} resulted in the loss of one helix and the creation of a disordered loop at the dimeric interface. With structure-guided mutagenesis, we confirmed functional validity of these structural changes using thermal shift assays and hypersensitive response assays in *Nicotiana benthamiana*. Mutations disrupting other interfaces did not result in changes in the DM3 function in terms of activating DM2h-mediated immune responses. We also confirmed that the immune function of DM3 is separable from the previously annotated PAP2 function as a prolyl aminopeptidase. Our results demonstrate that there exists a structural determinant in the DM3 complex critical for triggering DM2h-mediated immune responses. The role of DM3 in NLR signaling will be further discussed at XX International Plant Protection Congress 2024. With the studies of *DM* pairs, we gain further understanding on network properties of the plant immune system, wired to embrace numerous single points of failure.

SE17 C05

NRG1C ANTAGONIZES NRG1A-MEDIATED IMMUNITY BY COMPETITIVELY BINDING TO EDS1/SAG101

Junli Wang

Max Planck Institute for Plant Breeding Research, Cologne, Germany

Perception of pathogen effectors activates intracellular nucleotide-binding leucine-rich repeat (NLR) with N-terminal Toll/Interleukin-1 receptor (TIR) in planta and enables TIR-encoded NADase activities that are required for TIR-

NLR (TNL)-mediated immunity. The NADase activities of TIRs catalyze the production of small molecules which allosterically activate heterodimers of EDS1-PAD4 or EDS1-SAG101. Upon activation, EDS1-SAG101 directly interacts with downstream 'helper' NLR, N Requirement Gene 1 (NRG1A/1B), which has N-terminal coiled-coil (CC) domain, to execute host resistance and cell death via its Ca²⁺-permeable channel activity. NRG1C, a N-terminal truncated NRG1 is highly induced in TNLs triggered immunity and dampens NRG1A/1B mediated cell death in both *Arabidopsis thaliana* and *Nicotiana benthamiana*. However, the underlying mechanism is little known. In this current study, our gel filtration analysis shows that NRG1A interaction induces EDS1-SAG101 oligomerization. Moreover, we obtain the Crystal structure of EDS1-SAG101-NRG1C and the Cryo-EM structure of EDS1-SAG101-NRG1A^{WHD-LRR}. Structural evidence reveals that NRG1C outcompetes NRG1A for interaction with activated EDS1-SAG101 dimers due to its higher affinity for EDS1-SAG101. Our biochemical and structural analyses elucidate the details of EDS1-SAG101-NRG1A/1C interaction and shed light on how plants fine-tune TNLs triggered immune outputs.

SE17 C06

NLRSEEK: HIGH-THROUGHPUT DISCOVERY PIPELINE FOR FUNCTIONAL RESISTANCE GENES

Helen Brabham

2Blades, United Kingdom

Breeding crop species that are safe from pests and diseases is vital to build sustainable food systems crucial for food security. An effective and environmentally friendly method of disease control is to enhance the plant immune system by introducing functional resistance genes. A major class of plant immune receptors are nucleotide-binding, leucine-rich repeat receptors (NLRs), however identifying NLRs for use in elite crop varieties is time-consuming and resource-intensive. Through analyses of copy number variation and expression data, we identify high expression as an overlooked molecular signature of functional NLRs. Combining this signature with high-throughput crop transformation, we developed an approach that enables rapid identification and in planta validation of NLRs from non-domesticated germplasm. Screening 995 NLRs from 18 grass species identified 19 new resistance genes against wheat stem rust, a critical threat to wheat production. This pipeline facilitates rapid resistance gene discovery from diverse plant species to generate disease-resistant crops.

SE17 C07

RECEPTOR STRUCTURES AT THE PLANT-MICROBE INTERFACE FOR CROP IMPROVEMENT

Alexander Förderer

Root Biology And Symbiosis, Max-Planck-Institute of Molecular Plant Physiology, Potsdam, Germany

In the face of climate change, food security requires maximum plant productivity. To achieve this, crops must become more resistant to emerging pathogens and achieve high yields while conserving limited resources. A balanced interaction with microbes contributes significantly to crop health and is



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a promising target for manipulation. Plants have receptors that recognize harmful and beneficial microbes, leading to pathogen resistance or beneficial responses, respectively. Intracellular nucleotide-binding leucine-rich repeat receptors (NLRs) in plants recognize pathogenic effectors and trigger immune responses. Previously, we solved the cryo-electron microscopic (cryo-EM) structure of the wheat coiled-coil domain-containing NLR (CNL) Sr35 in complex with the effector AvrSr35 of the wheat stem rust pathogen. Direct binding of the effector to the leucine-rich repeats of Sr35 leads to the formation of a pentameric Sr35-AvrSr35 complex, also known as Sr35 resistosome, which functions strikingly similar to the pentameric ZAR1 resistosome of Arabidopsis. Relying on the structural conservation of these CNL resistosomes, structure-guided engineering of related Sr35 alleles from cultivated, susceptible cereals showed the great potential for knowledge-based resistance engineering in transient assays. My newly established group at the Max-Planck-Institute of Molecular Plant Physiology (Potsdam) capitalizes on this proof-of-principle from my postdoctoral work and extends structure-guided resistance engineering for crop applications via gene-editing and focuses on novel protein structural information from receptors at the plant-microbe interface for resistance breeding. Beyond improving crop resistance, our research also explores the potential of structure-guided engineering in symbiotic interactions of plants.



CONFERENCE I

08.30-11.00 Concurrent Session 18A

Fungicide Resistance: Diagnosis, Risk Assessment and Management, Integrated Pest Management

Chair: George Karaoglanidis (Aristotle University, Greece)

Dolores Fernandez Ortuno (University of Málaga, Spain)

SE18 001

THE SENSITIVITY OF EXSEROHILUM TURCICUM, THE CAUSAL PATHOGEN OF NORTHERN LEAF BLIGHT, TO DEMETHYLATION INHIBITOR FUNGICIDES IN SOUTH AFRICA

Van Butzelaar M., Berger D., Nsibo D.

Department of Plant and Soil Sciences, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa

Exserohilum turcicum, the causal agent of northern leaf blight (NLB) in maize, is the number one foliar pathogen of maize globally, causing up to 72% yield losses. Fungicide usage is a major management strategy of NLB and therefore, possible resistance to fungicides needs to be monitored to maintain their effectiveness. Here we aimed to determine the sensitivity and resistance mechanism of *E. turcicum* to demethylation inhibitor (DMI) fungicides, difenoconazole and propiconazole, in commercial and smallholder maize fields in South Africa. A total of 80 *E. turcicum* strains collected between 1970 and 2022 from Burundi, South Africa, Kenya and the United States of America were tested. This was an in vitro assay using 1 x 10⁴ conidia/ml against difenoconazole and propiconazole at 0, 0.00001, 0.0001, 0.001, 0.01, 0.1, 1, 10 and 100 µg/ml, followed by the sequencing of the EtCYP51 target mutation gene. The preliminary baseline EC₅₀ values for difenoconazole and propiconazole were 0.007 µg/ml and 0.1 µg/ml respectively, suggesting that strains were significantly more sensitive to difenoconazole than propiconazole (p = 0.0001). Although not statistically different, the recently collected strains from Eastern Cape, South Africa, a region with limited use of fungicides, showed reduced difenoconazole sensitivity of 0.01 µg/ml when compared to historical strains from the United States of America (EC₅₀ = 0.006 µg/ml at p > 0.05). The baseline sensitivity findings reveal that difenoconazole and propiconazole are still effective in managing NLB. The observed shift in the EC₅₀ values within and among fungicides are being investigated to determine any signatures of resistance. To our knowledge, this is the first baseline sensitivity study on *E. turcicum*-DMI fungicide interaction in South Africa. By gaining this knowledge, we will be able to create an opportunity in the agricultural sector to put more effective preventative- and practical measures in place against NLB, and potentially ensure less yield loss.

SE18 C01

EVOLUTION OF RESISTANCE IN FUSARIUM FUJIKUROI TO FUNGICIDES

Chuanqing Zhang, Chenyang Ge, Chenxin Mao

Zhejiang Agriculture and Forest University, Hangzhou, Zhejiang, China

Rice bakanae disease (RBD) caused by *Fusarium fujikuroi* is a

typical seed-borne fungal disease, which is widely distributed around the world. Seed treatments through fungicides are the main even only effective management strategy for management of RBD. Prochloraz (FRAC group 3, G1) a C14-demethylase inhibitor (DMI), was adopted as most extensive agent for RBD since 2010. Unfortunately, is a plant pathogen with high risk of resistance development to fungicides. In this study, we monitored the resistance development of *F. fujikuroi* to prochloraz and alternative fungicides in chronological order. Since 2010, isolates of *F. fujikuroi* were collected from different regions and their sensitivity to fungicides were determined continuously. The fitness of resistance isolates was assessed and molecular mechanisms of resistance was elucidated and confirmed. *F. fujikuroi* has developed serious resistance to prochloraz and failure of seed treatment with prochloraz is frequently reported in China. In 2022, the resistance frequency is as high as 79.5%. The point mutation (TCT→ACT) at position 312 on the FfCYP51B protein (S312T) and overexpression of *FfCyp51a* and *FfCyp51b* together lead to the prochloraz-resistance. Interestingly, prochloraz resistance in *F. fujikuroi* caused by cooperation between FfCyp51B with Cyp51A shows cross-resistance only to prothioconazole but not to other DMIs such as tebuconazole, and difenoconazole. The respective deletion of *FfCyp51A*, *Cyp51B*, and *Cyp51C* suggested sophisticated interactions within the three *Cyp51* genes in determining sensitivity to DMI fungicides in *F. fujikuroi*. Phenamacril (FRAC group 47, B6) is a *Fusarium*-specific fungicide with novel mode of action and is applied in RBD to replace prochloraz. Its resistance developed rapidly and was firstly detected in 2016 caused by the S219P/L point mutation in Myosin-5. The resistance frequency increased to 47 % in 2018 and a point mutation (AAG→ACG) at amino acid 218 of myosin-5 (K218T) was dominate than S219P/L. In recent years, researchers tried to adopt succinate dehydrogenase inhibitor (SDHI) fungicide to management RBD. However, natural resistance to the SDHI fungicide in *F. fujikuroi* was generally observed. Penflufen, for example, sensitivities of *F. fujikuroi* population surprisingly exhibited a bimodal distribution, caused by the presence of about 30% natural resistance sub-population.

SE18 002

BIOLOGICAL ACTIVITY OF DIFENOCONAZOLE AGAINST EXSEROHILUM TURCICUM, THE CAUSAL PATHOGEN OF NORTHERN LEAF BLIGHT OF MAIZE AND FUNGAL BASELINE SENSITIVITY

Nsibo D., Small B., van Butzelaar M.

Department of Plant and Soil Sciences, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa

Exserohilum turcicum, the causal agent of northern leaf blight (NLB) in maize, is the number one foliar pathogen of maize

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globally, causing up to 72% yield losses. Difenoconazole, a demethylation inhibitor (DMI) fungicide, is increasingly being used to manage NLB globally. Therefore, this study aimed to investigate the effect of difenoconazole on the biology of *E. turcicum* and determine any possible gain in resistance to the fungicide. A total of 80 *E. turcicum* strains collected between 1970 and 2022 were tested in an in vitro assay at a conidial concentration of 1×10^4 conidia/ml against difenoconazole at 0, 0.00001, 0.0001, 0.001, 0.01, 0.1, 1, 10 and 100 µg/ml. This was followed by sequencing the *EtCYP51* gene region to identify any target mutations attributed to resistance. Five strains were used to determine the effects of difenoconazole on the biology of *E. turcicum* both in vitro and in planta. The baseline sensitivity EC_{50} values ranged between 0.0009 and 0.01 µg/ml with an average of 0.007 µg/ml. In planta assays, the control efficacy of 14-93%, lesion area of 36191-46396 pixels, and disease index of 4-22 when the fungus was managed preventatively when compared to a control efficacy of 26-45%, lesion area of 45687 - 99658 pixels, and disease index of 14-28 when the fungus was managed curatively. Furthermore, mycelium electrolyte leakage increased over time and ranged between 7 and 13 µS/cm after 120 min. Overall, these findings suggest that difenoconazole still exhibits strong biological activity against *E. turcicum* and is more effective in a preventative than a curative manner. These results are crucial for effective management of *E. turcicum*. To maintain the efficacy of difenoconazole, it is important to monitor the shifts in the sensitivity of *E. turcicum* populations to difenoconazole.

SE18 003

METABOLOMICS REVEALS INSIGHTS INTO THE PATHOGENICITY AND RESISTANCE OF COLLETOTRICHUM ACUTATUM SPECIES COMPLEX TO FUNGICIDES

Giannakaris S.¹, Vasiou E.¹, Papadopoulou E.¹, Aliferis K.^{1,2}

¹. Agricultural University of Athens, Laboratory of Pesticide Science, Iera Odos 75, 118 55, Athens, Greece, ². McGill University, Department of Plant Science, 2111 Lakeshore Road, H9X 3V9, Montreal, Canada

Olive anthracnose caused by the *Colletotrichum acutatum* species complex, has recently emerged as a major threat for the olive tree cultivation, mainly due to its devastating impact on yield quantity and quality. Regardless of plant protection measures, the pathogen still causes severe losses. This is mainly due to the lack of knowledge on the genetic background of the pathogen's morphotypes and its linkage to traits such as pathogenicity and resistance to fungicides. In order to dissect the metabolism of selected *C. acutatum* isolates belonging to various morphotypes, gas chromatography-mass spectrometry (GC/MS) untargeted metabolomics was employed. Furthermore, their pathogenicity was assessed by performing artificial infections on unripe and ripe fruits of the cultivars *Koroneiki* and *Kalamon*, and their resistance to selected active ingredients (a.i.) was assessed *in vitro* (Figure 1). Major differences were discovered among the strains regarding their pathogenicity and sensitivity to the applied a.i. leading to their separation into two distinct groups. Strains of Group 2 caused typical symptoms on both cultivars, while being highly sensitive to selective a.i. In contrast, strains of Group 1 induced symptoms only in wounded ripe fruits of one cultivar, while exhibited resistance to the a.i. Metabolomics confirmed these observations by uncovering differences among the metabolic profiles of the two groups, and highlighted metabolites highly correlated to the traits being investigated, such as, among

others branched chain and aromatic amino acids, L-proline and L-OH proline, various carboxylic acids, GABA, and α,α-trehalose. This study highlighted the diverse nature of the *C. acutatum* species complex. Among the metabolites-biomarkers of effect, several were strongly correlated to the pathogenicity and sensitivity to fungicides, information that could be further exploited in the development of alternative and effective plant protection agents and/or programs.

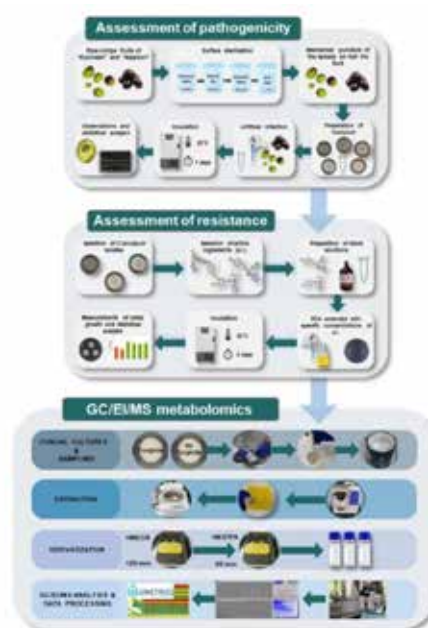


Figure1. Pipeline of the executed research

SE18 C02

FUNGICIDE SENSITIVITY in PYRENOPHORA TERES f. TERES ESTONIAN POPULATION

Regina Pütsepp, Riinu Kiiker, Liis Andresen, Andres Mäe

Department of Plant Protection, Centre of Estonian Rural Research and Knowledge (METK), Estonia

Net blotch caused by *Pyrenophora teres* f. *teres* (Ptt) is the most common foliar disease, causing substantial loss of spring barley productivity in Northern European countries as well as in Estonia. Climatic conditions are favorable for the development of the disease and pathogen spread, susceptible hosts are grown, and primary inoculum is not fully diminished. Optimal disease control with fungicides can give a substantial barley yield increase in a high disease pressure. Therefore, the objectives of this research were to assess *Ptt* populations in Estonia for (i) QoI resistance by detecting the correlated mutations in Cyt b, (ii) SDHI resistance by analysing mutations in SDH subunits, (iii) DMI resistance by identifying molecular changes in *Cyp51A* and *Cyp51B* gene and *Cyp51A* promoter region, and (iiii) potential sexual recombination by analysing mating type prevalence. Sensitivity towards DMI, SDHI, and QoI fungicides was estimated on the basis of pure cultured isolates aiming at ten isolates per locality. The resistance tests were carried out using the microtiter plate assay according to the FRAC protocol. The entire collection of *Ptt* was analysed for resistance mutations in fungicide target proteins. This is the first research study of Estonian *Ptt* field population characterizing several molecular changes in fungicide target proteins (CYP51A, CYP51B, Cyt b, and SDH subunits C and D) and *Cyp51A* gene promoter, which have potential impact



on DMI, QoI, and SDHI fungicide sensitivity. The data presented confirm the ongoing evolution of fungicide sensitivity in the *Ptt* population in Estonia. To prolong the effective lifetime of fungicides active ingredients should be applied in alternation as well as in mixtures where available, preferably including other modes of action.

SE18 C03

MONITORING AND MOLECULAR MECHANISMS OF FUNGICIDE RESISTANCE OF BOTRYTIS CINEREA STRAINS ISOLATED FROM STRAWBERRIES IN KOREA

Son D.¹, Choi S.¹, Liu H.¹, Park S.¹, Jang H.¹, Chang T.², Sang H.¹

¹. Chonnam National University, South Korea, ². Kyungpook National University, South Korea

Strawberries have one of the highest production values among crops in Korea. However, due to the high disease pressure of gray mold caused by *Botrytis cinerea*, different classes of fungicides have been applied to control the disease. During 2022-2023, a monitoring study on fungicide resistance in *B. cinerea* strains isolated from 90 strawberry farms in Korea was conducted. The majority of strains (about 83-90%) exhibited resistance to Methyl Benzimidazole Carbamates (MBC) and Quinone outside Inhibitor (QoI) fungicides, while about 58-70% of strains showed resistance to succinate dehydrogenase inhibitors (SDHI) fungicides. To investigate the molecular mechanisms of fungicide resistance, the genes targeted by fungicides, β -tubulin (*TUB2*), *CytB*, and succinate dehydrogenase complexes (*SdhB*, *SdhC*, and *SdhD*), were sequenced in the strains. MBC-resistant strains contained two types of mutations (E198A or E198V) in *TUB2*, and QoI-resistant strains contained a G143A mutation in *CytB*. Interestingly, a mutation, P225F in *SdhB*, was dominant in Korean strains, potentially due to a fungicide spray program with a specific chemical group of SDHI fungicide. In addition, the involvement of the mutations in fungicide resistance was confirmed using a genetic transformation system and AlphaFold model-based docking. Based on results of target gene mutation(s), a diagnostic method for detecting fungicide resistance using Taq-Man qPCR analysis was developed. This method was applied to disease field samples, successfully detecting the mutation. Our comprehensive results and developed method will contribute to the reduction of gray mold disease by enabling the selection of effective fungicides.

SE18 C04

PREVALENCE, MECHANISMS OF RESISTANCE, AND MANAGEMENT OF DMI AND MBC FUNGICIDE RESISTANCE IN MONILINIA FRUCTICOLA FROM SOUTHEASTERN US PEACH ORCHARDS

Schnabel G., Gura W.

Clemson University, United States

Sterol demethylation inhibitor (DMI) fungicides continue to be essential components for the control of brown rot of peach caused by *Monilinia fructicola*. A total of 238 *M. fructicola* isolates were collected from various commercial and two experimental orchards and sensitivity to propiconazole was determined based on a discriminatory dose of 0.3 μ g/ml. Results indicated 16.2%, 89.2% and 72.4% of isolates from

Alabama, Georgia, and South Carolina, respectively, were resistant to propiconazole. All resistant isolates contained the genetic element 'Mona' upstream MfCYP51, but it was absent from most sensitive isolates. Results show over the last 20 years resistance levels have not changed and no evidence of other resistance genotypes was found. A discriminatory dose of 1 μ g/ml thiophanate-methyl was used to distinguish sensitive (S) and moderately sensitive (S-LR) isolates from low resistant phenotypes, while 50 and 500 μ g/ml thiophanate-methyl concentrations were used to determine high resistant (HR) phenotypes. Results indicated 55.7%, 63.5%, and 75.9% of isolates from Alabama, Georgia, and South Carolina, respectively, were S to thiophanate-methyl; 44.3%, 36.5%, and 21.4% were S-LR; no isolates were LR; and only 3 isolates (1.3%) from South Carolina were HR. Only HR isolates revealed the E198A mutation. The high label rate of a commercial product containing thiophanate-methyl controlled brown rot caused by S and S-LR isolates in detached fruit studies but was ineffective against HR isolates. Our study suggests that current fungicide resistance management programs have been effective and suppressed development of new DMI resistant genotypes. The study also shows that thiophanate-methyl may again be useful to peach growers in the southeastern US for brown rot and fungicide resistance management.

SE18 C05

SELECTION FOR FENHEXAMID RESISTANCE IN BOTRYTIS CINEREA RELATING TO SPRAY DOSAGE, TIMING, AND MIXTURE

1 Mengjum Hu, 1 Stephen Boushell

¹University of Maryland College Park, College Park, United States

Spray tactics such as dosage, timing, and mixing partner have all been shown to affect fungicide resistance selection. However, the effects of those factors have often been examined individually, which could lead to inconsistent results between experimental settings. In this study, the high-risk fungicide fenhexamid was used to determine the combined effects of fungicide dose, mixture, and application timing together on resistance selection in *Botrytis cinerea* across varied frequencies of resistance. Our detached fruit assays showed that the low doses of fenhexamid, low-risk fungicide Captan, and application post-infection slowed resistance selection, while not comprising the control efficacy. In addition, even a low frequency of resistant *B. cinerea* population can lead to a significant reduction of disease control efficacy. In planta assays were then conducted to validate those spray tactics using *Vitis vinifera* 'Cabernet Sauvignon' grown in a greenhouse for two years. In agreement with the detached fruit assays, the greenhouse trials indicate that the lower dose of the at-risk fungicide, fenhexamid, most effectively managed fenhexamid resistance. Further, no major differences between the labeled and lowered doses of fenhexamid were observed in disease control. In addition, a mixture with the multi-site fungicide Captan generally resulted in a net-positive effect on both resistance management and disease control. Taken together, our findings largely agree with the use of the lowest possible fungicide dose and low-risk fungicides, but may challenge the previous assumption that preventative sprays benefit resistance management.



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BANQUET

11.30-14.00 Concurrent Session **19**

Plant Health Research Coordination: An International Endeavor

Chair: **Giovani Baldissera** (*Euphresco, France*)

SE19 C01

PLANT HEALTH RESEARCH COORDINATION: AN INTERNATIONAL ENDEAVOUR

B. Giovani

Euphresco, 21 Bd Richard Lenoir, 75011 Paris, France

It is not possible to avoid all the challenges to plant health posed by global trade, increasing travel activities and climate change. However, it is possible to optimise strategies to address these challenges with effective coordination and cooperation. Research plays a key role in underpinning phytosanitary activities, ranging from pest risk analysis, regulation, surveillance, taxonomy, diagnostics and mitigation measures. Research also helps to maintain and develop scientific expertise and infrastructure that support plant health. The Euphresco network for phytosanitary research coordination and funding started in 2006 to support phytosanitary research programme owners and programme managers to develop and take advantage of synergies amongst national research programmes and activities. The success of Euphresco as a primarily European network for phytosanitary research coordination has set the ground for discussions on the development of initiative(s) to address the needs of other regions of the world and global phytosanitary research coordination. The presentation will showcase the role of Euphresco as a platform for phytosanitary research coordination and collaboration and how the network has evolved since 2006.

SE19 C02

PLANT HEALTH RESEARCH PRIORITIES FOR THE MEDITERRANEAN REGION

A.M. D'Onghia

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Mediterranean agriculture, forests and other environments are seriously threatened by numerous quarantine and emerging pests, and their negative impacts are expected to increase due to the acceleration of global trade and to climate change that respectively favour the movement of these organisms over long distances and facilitate their adaptation to new environments. To address these threats, plant health challenges require rethinking the organization and coordination of research in all countries for: reducing fragmentation of actions; converging national programs; optimizing funding; and building critical mass. In this context, the Compendium on "Plant Health Research Priorities for the Mediterranean Region", published in 2021 to celebrate the International Year of Plant Health, was the first joint initiative of Euphresco and CIHEAM Bari launched to improve the coordination of research on plant health in the Mediterranean region and to enhance stakeholder cooperation in the area.

The compendium reports the information and views from national experts from the Balkan-Mediterranean, Eastern Mediterranean, Maghreb, and Western Mediterranean sub-regions on priorities of pests, research, infrastructures and capacity. The Compendium was updated in 2022 with a Supplement containing research needs for priority pests of common interest to the Mediterranean countries, which should be addressed in the short-term. Among these priorities, the following topics were selected for the Euphresco round of transnational collaboration: 1. Epidemiological studies on the potential vectors of *Xylella fastidiosa*; 2. Validation of DNA extraction from different matrices/hosts of *X. fastidiosa*; 3. Development/validation of diagnostic tests to identify exotic/severe strains of citrus tristeza virus (CTV); 4. Research topic on tomato brown rugose fruit virus (TBRFV). The Mediterranean model will inspire the consultations that will be conducted in Euphresco III project on a global scale to gather information useful for developing the strategic research agenda and identifying topics of common interest for transnational collaboration.

SE19 C03

TOOLS AND STRUCTURES FOR EARLY WARNING AND BETTER PREDICTION OF RISKS

M. Carvajal-Yepes

The Alliance of Bioversity International & the International Center for Tropical Agriculture, Palmira, Colombia

Early warning systems involves the timely detection and communication of potential or emerging threats posed by pests and diseases. This can be achieved through the use of various tools developed in the last decades. For instance, incorporating molecular analysis aids in identifying pathogens at a genetic level, in order to understand their potential impact. Satellite imagery, drones and other remote sensing technologies enable high-resolution data collection for monitoring crop health and detecting early signs of diseases. Furthermore, data analytics and machine learning are deployed to predict pest and disease outbreaks. While tools are crucial, the structures facilitating collaborations for sharing information, capacity and coordinating efforts in disease monitoring and management, are equally or more important. In this talk, we will review examples of tools, structures, and the role of the CGIAR centers in facilitating the identification of potential signs of pest and disease outbreaks at an early stage. This allows for prompt and target intervention to mitigate or prevent significant damage to crops.



SE19 C04

IN SITU DETECTION AND IDENTIFICATION OF PESTS AS AN OPPORTUNITY TO LINK FIELD AND LABORATORY ACTIVITY

N. La Porta

Fondazione Edmund Mach, Via Mach 1, 38098 San Michele all'Adige, Italy

Early detection of plant diseases is a crucial factor to prevent or limit the spread of a pest that could cause significant economic loss. Testing in the laboratory can be laborious, time consuming, expensive, and usually requires specific technical expertise. Moreover, in developing countries, it is often difficult to find laboratories equipped for this kind of analysis. Therefore, in the past years, efforts have been made for the development of fast, specific, sensitive, and cost-effective tests that can be successfully used directly in the field by non-expert personnel using simple equipment. Nucleic acid-based methods have proven to be a good choice for the development of detection tools in several sectors, such as human/animal health, food safety, and water analysis, and their application in plant pathogen detection is becoming more and more common. The portable laboratory features inexpensive instruments that can be carried as hand luggage and uses standard molecular biology methods, protocols and reagents that tolerate adverse environmental conditions. *In situ* diagnostic methods have a high potential for early detection of pests for agriculture and forestry, as they allow molecular detection of plant pests accessible to anyone, anywhere, and at any time. *In situ* methods should not replace laboratory testing, which remains essential for activities such as identification and classification of new pests or for the study of plant defence mechanisms. Instead, these new methods can provide a useful, fast, and efficient preliminary *in situ* screening that can strengthen our approach against plant pest.

SE19 C05

MODERN TOOLS AND APPROACHES TO TACKLE PESTS IN THE MEDITERRANEAN CROP PRODUCTION SYSTEMS

N.T. Papadopoulos

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Laying on three continents, the coastal Mediterranean areas and countries in general, constitute a unique social, physical and agricultural environment. Same or similar crops are cultivated, and often identical pest problems are encountered. Several alien invasive species arrive first in the Mediterranean countries towards invading Europe. In addition, climate change has already a dramatic impact on crop production and management of pests in the Mediterranean food production systems. Recent technological advances render available a range of tools, services and approaches that can be adopted for pest management purposes. These include among others, electronic sampling and pest monitoring systems (e.g. e-traps), precise mapping and interactive spatial depiction of pest populations and crop infestation rates, bioclimatic and population growth modelling, novel forecasting approaches, mapping and tracing of pest management interventions, real time sharing of large amount of data. The presentation will elaborate on management of current and imminent threats

of Mediterranean crops by pests, placing special emphasis on alien invasive species. Use and adoption of modern technology to tackle these pests is discussed considering the never-ending need for high quality and precision data from the field. Examples of modern approaches to address invasive fruit flies in Europe are provided by the recently concluded and the ongoing Horizon 2020 funded projects FF-IPM and REACT respectively. A list of areas for developing common approaches and strengthening of the collaboration among plant protection agencies across Mediterranean countries will be discussed.

SE19 C06

HOW RESEARCH CAN SUPPORT POLICY-MAKING: FUTURE PERSPECTIVES

M. Ciampitti

Servizio Fitosanitario DG Agricoltura, Piazza Città di Lombardia 1, 20124 Milano, Italy

Research can play a crucial role in supporting policy-making on plant health and plant protection. Plant protection authorities have the task to adopt effective measures for the protection of the territory and its plants, ensure safe trade, and mitigate the impacts of climate change on the health of crops and forests; policy-makers are supposed to develop integrated decision making through scientists and economists working within an interdisciplinary framework. The World Trade Organization emphasizes that phytosanitary measures must be anchored in science, based on a risk assessment, and follow international standards, guidelines, and recommendations developed by the International Plant Protection Convention. In this framework, collaboration between policy-makers and researchers and between the different research teams, should be promoted and encouraged, at national level, but especially at international level as quarantine and emerging plant pests move without respecting borders. International research collaboration across nations, institutions and disciplines leads to higher quality of science, efficient use of resources, better outcomes and wider research results dissemination. However, these benefits of collaboration only occur where there is mutual interest and alignment of goals (including a "vision"), effective leadership, facilitation of processes and structures, support for collaboration, and ultimately funding – for both research and collaboration. In addition, the need to develop a balanced portfolio of research work, ranging from strategic to applied research, is essential in creating synergistic collaboration. The IPPC community is striving to set by 2030 a voluntary mechanism for global phytosanitary research coordination, to accelerate development of science to support all regulatory phytosanitary activities. A network of organisations that fund research projects and coordinate national research in the phytosanitary area, like Euphresco, may provide useful perspectives on policy and structures for this new mechanism. Through research coordination and a close relationship between researchers and policymakers, the competent authorities will then be able to develop more effective policies and regulations to mitigate plant pest risks and ensure sustainable food production.



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SE19 C07

FUTURE CHALLENGES

Laura Mugnai

SE19 C08

HOW TO PROTECT PLANT HEALTH IN THE NEXT 20 YEARS

Dimitris Tsitsigiannis

SE19 C09

RESEARCH TO ADDRESS FUTURE CHALLENGES AND TO STRENGTHEN PLANT PROTECTION

Anna Maria D'Onghia

SE19 C10

FROM RESEARCH TO REALITY: CPN BRIDGES THE GAP IN PLANT HEALTH SOLUTIONS

D. Mueller

Iowa State University, 2205 Adv Tch Res Bd, 2213 Pammel Dr, Ames, IA 50011-1101, United States of America

The Crop Protection Network (CPN) spearheads a multinational alliance, uniting university and provincial extension specialists with public and private experts. Fueled by unbiased research, the CPN helps farmers and agricultural professionals make informed decisions protecting alfalfa, cotton, corn, small grains, and soybeans. Beyond state and international boundaries, the CPN amplifies the reach of agricultural extension, serving as an important conduit for research on plant health. This network actively shares impactful content, from practical guidelines to new discoveries, equipping stakeholders with the latest tools to tackle crop protection challenges effectively. At its heart, the CPN boasts a network of more than 320 specialists spanning 40 U.S. states and Ontario, Canada. This collaborative platform fosters interdisciplinary teamwork, forging research alliances and seamlessly sharing scientific knowledge. Grounded in unbiased research, the CPN harnesses collective expertise to tackle complex problems in plant health, ultimately driving more effective and sustainable agricultural practices.

SE19 C11

THE ROLE OF THE ARAB SOCIETY OF PLANT PROTECTION IN ENHANCING PLANT HEALTH RESEARCH COORDINATION IN THE ARAB REGION

S.G. Kumari, K.M. Makkouk and I. Al-Jboory

Arab Society for Plant Protection, Beirut, Lebanon

The strategic objectives in plant health research and development in any country or region are achieved through (i) enhancement of understanding and knowledge of plant health issues and solutions, (ii) development of evidence-based risk assessment policy approaches to empower delivery partners and practitioners to make solid decisions and take necessary action, (iii) develop and adopt innovations and new technologies in support of plant health policy objectives, and (iv) create high-quality plant health research capabilities. The Arab Society for Plant Protection (ASPP), can play an influential role in achieving some of the above-

mentioned objectives by enhancing collaboration among scientific groups from different Arab and Mediterranean countries through a variety of activities such as (i) organizing regional conferences that bring together scientists from different Arab countries and the rest of the world to shed light on significantly important plant health issues of common concern and publicize best pest management practices, (ii) organizing regional workshops addressing specific plant health issues that require joint effort, (iii) encourage and facilitate distribution of up-to-date knowledge related to crop health management, and (iv) enhance technical skills through specialized courses. In addition, ASPP can facilitate the formation and implementation of joint regional research projects that focus on plant health management. Furthermore, ASPP can play an essential role in identifying plant health challenges facing the Arab region and reflecting on future solutions that are needed to minimize yield losses caused by pests to enhance food security for future generations in the Arab region.

SE19 C12

THE INTERNATIONAL ASSOCIATION FOR THE PLANT PROTECTION SCIENCES

G. Norton, E. A. Heinrichs, and M. Tamo

The University of Queensland, PO Box 5486 Stafford Heights, Brisbane, Queensland 4053, Australia

The origin of IAPPS dates back to the first International Plant Protection Congress (IPPC), held in Belgium in 1946. Subsequently, ad hoc committees were formed to plan future IPPCs, until the late 1990s, when IAPPS was formally established (54 years later) at the 2000 IPPC meeting in Jerusalem. IAPPS has overseen subsequent IPPCs, generally, at four-year intervals.

Currently, the IAPPS Board consists of:

- an executive committee including a President, Vice President, Secretary General, Treasurer
- fourteen International Regional coordinators, covering the major continents
- representatives that include the Association of International R&D Centres for Agriculture, the Biopesticide Industry, Elsevier Crop Protection (IAPPS official journal), and the next IPPC Chair

The mission in IAPPS – ‘to promote the development of an integrated approach to plant protection research and its practical application’ – involves the following:

- oversight of the IPPC and encouraging the information exchange that occurs
- involvement in regional seminars and workshops
- providing news items, newsletters, and online educational and training materials
- support the development of national and regional plant protection societies

Therefore, over the past two decades, IAPPS has extended its activities well beyond its involvement in the IPPC. During the years prior to Covid, this increasingly involved IAPPS members, together with participants from other agencies, organising regional seminars and workshops, largely focussed on emerging pest problems. In the past year or so, there has been a revival of these activities. However, the major development has been the increased role of IAPPS in digital communication. For instance, IAPPS now provides:



- the popular, Global Plant Protection News blog
- the monthly IAPPS Newsletter, posted on the IAPPS website (www.plantprotection.org), and subsequently published in the hard copy of the Crop Protection Journal
- online access to Crop Protection (the official IAPPS journal) for IAPPS members
- access to up-to-date summaries of recent articles in Crop Protection

An education and training section has recently been added to the IAPPS website, managed by an IAPPS editorial committee. Digital publications include a series of plant protection 'stories': a multi-author review of digital identification tools for plant biosecurity; and online pathway keys for identifying insect pests and natural enemies of rice in Asia & West Africa. Where possible, material on the IAPPS website is published both as down-loadable, pdf documents as well as online (html) publications. This has the advantage of allowing users to view the article in their local language, using the Google Translate facility embedded in the IAPPS website. *The IAPPS Board is always interested in exploring new ways in which we might work collaboratively with other agencies to achieve common goals more effectively.*



SKALKOTAS

11.30-14.00 Concurrent Session **20A**

Biopesticides and Biofertilizers

Chair: Sotiris Tjamos (Agricultural University of Athens, Greece)

Panagiotis Sarri (Microbiology, Department of Biology University of Crete, Greece)

SE20 C01

BACILLUS VELEZENSIS K165: A MULTITASKING BIOCONTROL AGENT

Eirini G. Poulaki¹, Litsa Ampntelnour², Vasilis Dimitrakas¹, Ioannis A. Stringlis¹, Sotiris Tjamos¹

1. Phytopathology lab, Agricultural University of Athens, Greece, 2. School of Life Sciences University of Warwick, England

The biocontrol agent *Bacillus velezensis* K165 (formerly known as *Paenibacillus alvei* K165) was initially isolated in the 1990s from the rhizosphere of tomato plants cultivated in soil solarized fields. Over the years, it has demonstrated remarkable efficacy in protecting plants against a range of soil-borne pathogens. Its genome harbors gene clusters responsible for producing secondary metabolites, including antibiotics such as fengycin. Untargeted metabolomic analysis has further unveiled its ability to produce compounds that trigger plant defense mechanisms, aligning with its capacity to induce systemic resistance (ISR). Experimental investigations have revealed that K165's ISR-inducing activity relies on the FLS2 receptor, WRKY 70, SID2, EDS5, and NPR1, key components of salicylic acid-dependent defense pathways. Moreover, comprehensive analyses integrating histone acetyltransferases mutant screening, chromatin immunoprecipitation assays, and transcriptomic profiling have elucidated the pivotal role of histone acetylation in K165's biocontrol activity. This epigenetic modification not only contributes to K165's immediate biocontrol efficacy but also protects next generations of plants against pathogens like *V. dahliae*. More recent insights have highlighted the importance of K165's volatile organic compounds (VOCs) in conferring protection against *V. dahliae* in *Arabidopsis* plants. RNAseq analysis has shown the specific impact of K165 VOCs on the chitin perception mechanism in plants and the synthesis of lignin, further elucidating the molecular basis of its biocontrol activity. Overall, these findings underscore the multifaceted mechanisms by which K165 operates as a potent biocontrol agent, offering promising avenues for enhancing plant resilience against soil-borne pathogens. Continued research into the intricate interplay between K165 and plant defense mechanisms holds significant potential for developing sustainable strategies to mitigate crop diseases and bolster agricultural productivity.

SE20 C02

HOW TO COMBINE TRICHODERMA SPP. WITH SUSTAINABLE PEAT ALTERNATIVES

Jane Deboe and Bart Vandecasteele

Trichoderma spp. are widely used as biological control agents (BCA), as they target multiple plant pathogens both directly, through their antagonistic and mycoparasitic activity, and indirectly, by triggering plant defence mechanism. In horticulture, peat is still a major constituent of growing media. However, there are environmental concerns regarding peat extraction and utilization and there is an urgent need to find sustainable alternatives for peat in horticulture. A promising avenue is the use of residual biomass, such as composts, plant fibers, nature conservation management residues and biochars. However, these peat alternatives are highly variable due to different feedstocks and production methods and there is few knowledge about which characteristics predict their BCA carrying capacity. We tested the BCA carrying capacity of materials in incubation tests with commercial *Trichoderma*-based BCA using plating, qPCR analysis and metabarcoding. We measured several chemical and biological characteristics of the peat alternatives and tried to find which characteristics predict their BCA carrying capacity. In addition, the biocontrol activity of the BCA in growing media with peat alternatives was tested in strawberry greenhouse trials. Promising peat alternatives showing high BCA colonization capacity are plant fibers such as miscanthus, composts such as green compost, nature conservation management residues such as acidified soft rush and biochars produced from woody biomass at low temperatures. A negative correlation was found between BCA carrying capacity and the initial fungal biomass (as measured by PLFA) of compost and management residues. For biochars, multivariate linear regression showed that their BCA carrier capacity can be predicted by four chemical parameters: the carrier capacity is higher at high water-soluble carbon, low pH-H₂O, low inorganic carbon and low total phosphorus content. Strawberry experiments in the greenhouse revealed an important role of the rhizosphere microbiome for the BCA to colonize the peat alternative growing media.



SE20 C03

MICROBE-MICROBE INTERACTIONS IN THE RHIZOSPHERE INFLUENCE BIOCONTROL ACTIVITY OF *LYSOBACTER CAPSICI* AZ78 BY MODULATING ITS METABOLOME AND TRANSCRIPTOME

Francesca Brescia^{1,2}, Martina Marchetti-Deschmann³, Marc Ongena⁴, Marco Moretto², Michele Perazzolli^{1,2}, Ilaria Pertot^{1,2}, **Gerardo Puopolo^{1,2}**

1. Center of Agriculture, Food, Environment, University of Trento, San Michele all'Adige, Italy, 2. Research and Innovation Centre, Fondazione Edmund Mach, San Michele all'Adige, Italy, 3. Institute of Chemical Technologies and Analytics, TU Wien (Vienna University of Technology), Vienna, Austria, 4. Microbial Processes and Interactions Laboratory, Terra Teaching and Research Center, Gembloux Agro-Bio Tech, University of Liège, Gembloux, Belgium

Lysobacter spp. are plant-beneficial rhizobacteria frequently associated with soil suppressiveness. However, their application in soils does not reproduce this phenomenon. To understand if microbial interactions may influence *Lysobacter* spp. biocontrol activity, we investigated two and three-way interactions between *L. capsici* AZ78 (AZ78), *Bacillus amyloliquefaciens* subsp. *plantarum* S499 (S499) and *Pseudomonas protegens* Pf-5 (Pf-5) on a growth medium mimicking the tomato rhizosphere environment. S499 negatively modulated AZ78 inhibitory activity against *Pythium ultimum*. This effect was related to a substantial impact on AZ78 transcriptome. In particular, S499 caused the downregulation of genes responsible for the biosynthesis of the Heat Stable Antifungal Factor (HSAF). Metabolome analysis confirmed the absence of HSAF in AZ78. Using mutants, we found that S499 may influence AZ78 metabolome and transcriptome by releasing bacilysin and difficidin. Differently, Pf-5 positively affected AZ78 inhibitory activity, modulated the expression of a lower number of genes, and stimulated the release of antifungal compounds. In the three-way interaction, Pf-5 reduced the negative effect of S499 on AZ78 by stimulating AZ78 release WAP-8294A, an antibiotic active against Gram-positive bacteria. Overall, our results first hint at how microbe-microbe interactions in the rhizosphere might influence *Lysobacter* spp. 's ability to contribute to soil suppressiveness.

SE20 C04

TOMATO-BENEFICIAL FUNGUS-BIOTIC STRESSOR INTERACTION: UNTANGLING THE COMPLEX NETWORK OF MOLECULAR MECHANISMS BY -OMICS STUDIES

Silvia Proietti¹, Gaia Salvatore Falconieri¹, Alberto Pascale², Laura Bertini¹, Sheridan Lois Woo^{3,4,5}, Michelina Ruocco⁵, Francesco Loreto^{3,6}, **Carla Caruso¹**

1. Department of Ecological and Biological Sciences, University of Tuscia, Viterbo, Italy, 2. Plant-Microbe Interactions, Department of Biology, Science for Life, Utrecht University, Utrecht, Netherlands, 3. BAT Center—Interuniversity Center for Studies on Bioinspired Agro-Environmental Technology, University of Naples Federico II, Naples, Italy, 4. Department of Pharmacy, University of Naples Federico II, Naples, Italy, 5. Institute for Sustainable Plant Protection (IPSP-CNR), Portici (NA), Italy, 6. Department of Biology, Via Cinthia, University of Naples Federico II, Naples, Italy

Plant roots can exploit beneficial associations with soil-inhabiting microbes, promoting plant growth and immune capacity. *Beauveria bassiana*, an endophytic fungus, has a wide range of plant hosts, including tomato (*Solanum lycopersicum* L.), an economically very important species grown all over the world. *B. bassiana* is extensively recognized for its positive effect in growth promotion and pathogen/pest bio-control. However, little is known about the molecular mechanisms that regulate plant-*B. bassiana* interactions. In order to obtain a comprehensive snapshot of the molecular events underlying the tomato-*B. bassiana* relationship, plant proteome reprogramming caused over time by fungal invasion was thoroughly investigated. The proteomics data revealed perturbation of critical biochemical pathways by *B. bassiana*, related with both primary and secondary metabolism, and plant growth. Plant hormone analysis revealed up-regulation of a wide range of growth-related hormones, including gibberellins, and defense-related hormones such as jasmonate. *B. bassiana*-colonized plants also became more resistant against *Botrytis cinerea*, likely because of improved functioning of their antioxidant machinery. The tripartite interaction *B. bassiana*-tomato-*B. cinerea* was further explored by proteomics and hormone analysis, confirming the protective role of the beneficial fungus at molecular level. Finally, we demonstrated that *B. bassiana* synergistically interacts with a second beneficial fungus, *Trichoderma afroharzianum*, strengthening its biocontrol activity.

SE20 C05

SYNERGISM BETWEEN BIORATONALS AND STRATEGIC LOW DOSES OF DMI FUNGICIDES AGAINST IMPORTANT FRUIT CROP DISEASES

Schnabel G., Wesche J.

Clemson University, United States

Biological control agents (BCAs) *Bacillus subtilis* formulated as Theia® and *Pseudomonas chlororaphis* formulated as Howler® are registered for control of fungal and bacterial diseases of fruit crops. Combinations of Theia® and Howler® with strategic concentrations of demethylation inhibitor (DMI) fungicides were investigated to explore potential synergisms. *B. subtilis* bacteria were cultured in nutrient broth and combined with technical grades and two formulations of propiconazole (Emulsifiable Concentrate [EC] and Wettable Powder [WP]) and metconazole (EC and Water Dispersible Granule [WDG]) at 0, 10, 50, 100, and 150 µg/ml active ingredient. After co-cultivation, optical density (OD₆₀₀) and colony forming units (cfu values) were evaluated. In contrast to EC formulations, the WP or WDG formulations at 10 or 50 µg/ml of both DMIs did not affect vegetative cell growth. The mixture of Theia® and each formulated DMI at 50 µg/ml active ingredient resulted in a significant reduction of *Monilinia fructicola* lesion development on apple, *Colletotrichum siamense* lesion development on cherry, and *Botrytis cinerea* lesion development on cherry. The combination of Theia® with EC formulations showed weaker disease reduction. Theia® + non-EC formulated propiconazole reduced brown rot disease incidence of apple more effectively than Theia® + EC formulated propiconazole. Similar results were found for combinations of DMI fungicides with Howler®. Howler® was also found to reduce and often prevent DMI fungicide-induced expression of the 14-alpha demethylase (*MfCYP51*)

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gene, encoding the target enzyme for DMI fungicides. Our results indicated bactericidal effects of DMI fungicides and the potential for synergism in combination with a DMI fungicide. A novel mechanism of action of Howler® fungicide was discovered that may be exploited for management of DMI fungicide resistance based on target gene overexpression.

SE20 C06

BIOLOGICALS IN MODERN AGRICULTURAL PRACTICES

Stefan Tresch

BASF SE, Germany

There is an ever-increasing demand of growers and farmers to produce high-quality, environmentally conscious, sustainable products. By offering a wide range of tools to protect crops against pests and diseases, we can overcome these challenges through effective, sustainable, and biological control. Biological control is the use of living organisms and substances derived from nature to manage pests and diseases, improve crop protection management and support plants throughout their life cycle. Biological products provide benefits for grower like no residues left after their use or can be an alternative tool for resistance management. Many of the products are very target specific and with less or non-effects towards other beneficial insects. The broader use of some biological products can be restricted due to their inconsistent performance under variable conditions or limited due to e.g. short shelf life. How biological products can be combined in modern integrated pest management programs will be shown. Furthermore, it will be discussed what new technologies can broaden our toolbox to combat pests and diseases and what technical challenges have to be solved for practical use on grower level.

characterization of the nodule microbiome from grain pulses by culture-dependent methods and the selection of plant growth-promoting bacteria to be used in agronomic practices in a sustainable agriculture context. To this end, the genetic diversity of indigenous rhizobia nodulating grain legumes in Greek soils were studied. Case studies with selected indigenous rhizobial strains used as inoculants in greenhouse and open field trials will be presented. Noteworthy, inoculation of common bean with an indigenous rhizobial strain was superior to a commercially supplied rhizobial strain in terms of nodulation and BNF capacity. In addition, NRE bacteria were evaluated for their ability to inhibit the in vitro growth of phytopathogenic bacteria and fungi. Some of them were able to reduce the symptoms of grey mould caused by *Botrytic cinerea* in tomato and eggplant. In conclusion, beneficial microorganisms are among the best approaches for sustainable agricultural production and a feasible solution to sustain the twin problems of global food security and environmental stability.

Acknowledgements: This research was funded by the Hellenic Foundation for Research and Innovation (HFRI) under the HFRI PhD Fellowship grant (Fellowship Number: 957), the European Union's Horizon 2020 research and innovation programs under Grant Agreements: i) No. 727973, project "TRUE- TRAnsition paths to sustainable legume-based systems in Europe", ii) No 727929, project "TOMRES—A novel and integrated approach to increase multiple combined stress tolerance in plants using tomato as a model" and the EUROLEGUME research project of the 7th Research Framework Programme of the European Union (No 613781). This publication is financed by the Project "Strengthening and optimizing the operation of MODY services and academic and research units of the Hellenic Mediterranean University", funded by the Public Investment Program of the Greek Ministry of Education and Religious Affairs".

SE20 C07

ENHANCING LEGUME AND VEGETABLE GROWTH AND HEALTH: HARNESSING MICROBIAL BIOSTIMULANTS

Anastasia Tampakaki

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The agricultural sector faces a dual challenge: increasing productivity to support a growing global population while improving resource efficiency and reducing environmental impact on ecosystems and human health. Fertilizers and pesticides are vital tools in agriculture, essential for growers to enhance yields and maintain consistent productivity. Biostimulants, a class of fertilizing products, attract significant attention for their capacity to improve crop characteristics. Among them, microbial biostimulants stand out for their dual role in promoting nutritional processes and protecting plants against phytopathogenic organisms. A good sustainable agricultural practice to achieve optimal biofertilization is the selection of efficient, competitive, and well-adapted bacterial strains in different edaphoclimatic zones. Root nodules of legumes are suitable for the isolation of rhizobia, responsible for nodule formation and non-rhizobial endophytic (NRE) bacteria with plant growth promoting and phytoprotecting properties. The main objective of this study was the



MC2

11.30-14.00 Concurrent Session **21**

Selected Highlights in Plant Protection

Chair: Maria Lodovica Gullino (*Coltivato, Italy*)

SE21 C01

ADVANCES IN BIOLOGICAL CONTROL

Monica Hoftte

SE21 C02

ADVANCES IN PEST MANAGEMENT OR ECOSYSTEM SERVICES

Ramon Albajes

SE21 C03

BIOSECURITY ISSUES, WITH SPECIAL EMPHASIS IN THE ENTOMOLOGICAL SECTOR

Mariangela Ciampitti

SE21 C04

A CAREER ON BOTH SIDES OF THE ATLANTIC: MEMOIRS OF A MOLECULAR PLANT PATHOLOGIST

Nickolas J. Panopoulos

Professor Emeritus, Department of Environmental Science, Policy and Management, University of California, Berkeley, California 946 19, Department of Biology, University of Crete, 710 03 Heraklion, Greece, Hellenic Agricultural Academy, Agricultural University of Athens, 118 55 Athens, Greece

My career in plant pathology evolved over more than four decades, starting with the bachelor's degree from the Agricultural University of Athens and specializing in Phytopathology. After a brief fellowship period at NCSR-Demokritos I sought to obtain a Ph. D. in the Department of Plant Pathology, University of California, Berkeley. My academic research career evolved sequentially at Berkeley and at the Department of Biology, University of Crete and Institute of Molecular Biology and Biotechnology of FORTH in Heraklion, Greece. My research orientation was shaped by an irresistible curiosity to understand basic plant-pathogen interaction mechanisms. This curiosity was implanted by inspired teachers and mentors, from within and outside the plant pathology field. To these people I extend my sincere thanks. I focused mainly on phytopathogenic bacteria as research models, mainly due to the development of emerging tools of recombinant DNA following the 1960-1970s decade and their broad adoption in the life sciences in general. As a result, phytopathological research experienced dramatic shifts in the following decades, focusing mainly on molecular (genome-level) analysis, initially of viral and bacterial genomes, and subsequently of fungal and plant genomes, constantly drawing ideas, knowledge and tools from related and unrelated scientific fields in hopes that basic research will eventually mature into practical applications in plant protection. This hope is best expressed symbolically by the inscription on the west pediment of Hilgard Hall, the building which housed the Department of Plant Pathology at Berkeley

at that time: «To Rescue for Human Society the Native Values of Rural Life». I will refer to selected examples of relevant studies. In my professional path I had the opportunity to train many high caliber students and to collaborate with post-doctoral associates, visiting scientists, technicians and faculty colleagues while at Berkeley and in Greece. To all these people and to my family I extend my sincere thanks.

SE21 C05

EFFECT OF CLIMATE CHANGE ON PLANT DISEASES AND THEIR MANAGEMENT

Maria Lodovica Gullino^{1,2}, Massimo Pugliese³, Giovanna Gilardi³, Angelo Garibaldi³

1. International Agricultural Festival Coltivato, Torino, Italy, 2. AgriNewTech s.r.l, Torino, Italy, 3. University of Torino - Agroinnova, Italy

Climate change is having a significant impact on global agriculture as a whole. Increases in carbon dioxide (CO₂) and temperatures are expected to induce complex effects on plant diseases. Different approaches have been followed to study the impact of climate change on plant diseases, such as modeling and experimental trials in simulated conditions. Phytotrons allow to simulate future scenarios, combining temperature and carbon dioxide and evaluating their effect on foliar and soil-borne diseases. Vegetable crops like lettuce, rocket, basil, beet and zucchini, as well as grapes and other plants were grown in phytotrons under six different simulated climatic conditions: standard CO₂ concentration for the area (400 -450 ppm) with standard (ranging from 18 to 22/24° C) and elevated temperature (5-10 °C higher than standard), elevated CO₂ (800 -850 ppm) with standard and elevated temperature. An increase of powdery mildew on zucchini, of *Alternaria* leaf spot on rocket salad, of black spot and downy mildew on basil, of *Allophoma tropica* on lettuce, of *Phoma* leaf spot on leaf beet and on rust of pelargonium was observed when both CO₂ level and temperature increased. Variable effects were instead observed when individual climate parameters were taken into consideration. Additionally, the influence of climate change on mycotoxin production and disease management strategies has been considered. In the case of *Alternaria* spp., tenuazonic acid production improved on cauliflower and rocket when both CO₂ level and temperature increased. The biosynthesis of roridin E increased by the rise in carbon dioxide. Regarding the efficacy on disease management practices, azoxystrobin and *Ampelomyces quisqualis* were found to be the most effective against *Podosphaera xanthii* on zucchini at high CO₂ and temperature conditions. The efficacy of mancozeb and azoxystrobin improved at high CO₂ against *Phoma* leaf spot on beet. Results are discussed also in relation to possible mitigation measure.



MC3

11.30-14.00 Concurrent Session 22

Management of Biological Invasions in a Changing World

Chair: Nikos T. Papadopoulos (University of Thessaly, Greece)

SE22 C01

INTRODUCTION TO MANAGEMENT OF BIOLOGICAL INVASION

Nick T. Papadopoulos

native insect pests, and discusses the constraints and needs for contingency planning, including a proactive approach to develop the SIT package for other potential major invasive insect pests.

SE22 C02

AREA-WIDE MANAGEMENT OF FRUIT FLIES USING THE STERILE INSECT TECHNIQUE TO RESPOND TO INVASIVE SPECIES

Rui Pereira

Insect Pest Control Subprogramme, Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture, Vienna, Austria

Traditional control methods of fruit flies are mostly based on the uncoordinated application of insecticides using an orchard-by-orchard approach. This approach is not effective or sustainable in view of fruit fly movement and is also damaging to the human health and the environment. Also the rapid expansion of international travel and trade are facilitating the spread of non-native species, while climate change is creating new ecological niches enabling the establishment of pests in previously inhospitable regions. A fruit fly area-wide integrated pest management (AW-IPM) strategy aims to prevent the entire targeted population of Exceeding an economic threshold. This requires the treatment of all habitats to avoid leaving significant infestations from where migrant insects can re-establish in areas of concern. The AW-IPM requires coordination among all stakeholders, long-term commitment, usually multiyear planning, and a centralized organization dedicated exclusively to its implementation. This entails collection of necessary baseline data including species present, temporal, and spatial dynamics of the target populations over several years, host status and sequence. Among the control tactics available, the sterile insect technique (SIT) can contribute to the AW-IPM approach in situations where the number of key pests is low, and when integrated with other control methods. Among the methods available for integration, depending on the characteristics of the infested area, are sanitation, bait sprays, mass-trapping, and male annihilation technique. The SIT is a method of pest control involving area-wide inundate releases of sterile insects to reduce reproduction in a field target population of the same species in an area. It is one of the most environment-friendly methods available and can be implemented using four different strategies, i.e. suppression, containment, eradication, and prevention. With the markets growing and becoming sensitive to environmental concerns regarding pesticide residues, the SIT is being used more and more on fruit fly suppression replacing conventional control methods. This chapter compares some eradication campaigns in the pre-SIT era with those integrating the SIT to eradicate, contain or prevent the establishment of non-

SE22 C03

THE INVASION OF THE FALL ARMYWORM SPODOPTERA FRUGIPERDA (LEPIDOPTERA: NOCTUIDAE) IN EUROPE: THE SITUATION IN GREECE

Spyridon Antonatos¹, Ioanna Lytra¹, Vasiliki Evangelou¹, Iro Georgopoulou¹, Evangelia Tselou¹, Danai Dimopoulou¹, Panagiotis Milonas¹, Arampatzis Christos² and Dimitrios P. Papachristos¹

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The fall armyworm *Spodoptera frugiperda* (Lepidoptera: Noctuidae), is one of the most serious pests of a wide range of plants including maize, soybean, cotton and rice. It is native to tropical and subtropical regions of the American continent. In 2016 the insect was detected in West and Central Africa where severe outbreaks occurred. Thereafter, it rapidly spread throughout the continent and by May 2017 was present in almost all sub-Saharan African countries. In 2018 and 2019 the pest was recorded in several Asian countries and in 2020 in Australian mainland. In July 2020 its presence was confirmed in the Canarian islands of Spain while during 2022 and 2023 the insect detected in Turkey, Cyprus, Madeira Island of Portugal and lastly in Romania. In autumn of 2023 an outbreak of the pest occurred in Greece. Male adults of the species were captured in pheromone traps in seven regional units of south and central part of Greece in the regions of Crete, Peloponnese, central Greece and North Aegean. In most areas the first catches occurred in September and the number of trapped adults decreased in October. During November a significant increase was observed and finally in December, the catches decreased again to zero catches in early January. However, in the regional unit of Lasithi in Crete adults were continuously captured in traps during winter. All the positive traps were placed in fields with Solanaceae plants, mainly tomatoes and eggplants. Despite the captures of adult males in the traps in most cases no infestation was observed in the crops that were inspected. Intensive trapping efforts are in place in the infected areas and in neighboring ones for monitoring the development of the outbreak.

The study was funded by the Hellenic Ministry of Rural Development and Food under the national plant pest surveillance program.



SE22 C04

THE OZONE SOLUTION: TACKLING THE INVASIVE BOX TREE MOTH

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The Box Tree Moth (*Cydalima perspectalis*) poses an enormous threat to boxwood (*Buxus* spp.) populations worldwide, so effective control methods need to be researched. This study investigates the efficacy of gaseous ozone as a novel solution to control the invasive box tree moth. In controlled experiments, we evaluate the effects of ozone treatments on suppressing population growth and mitigating the damage caused by the larvae. Our results are promising and highlight the considerable potential of ozone treatments in controlling the spread of this invasive pest while minimising the environmental impact. Specifically, fourth instar larvae and pupae were subjected to a gaseous ozone treatment of 10 to 60 minutes (0.7 – 4.3 ppm). The effective treatments were observed in larvae, which showed efficacy rates of over 32% after 30 and 60 minutes of ozone exposure. When pupae were treated, efficacy rates of 61% were observed after 30 minutes and 100% after 60 minutes of ozone exposure. No adverse effects of ozone on the box trees were observed throughout the study. In addition to the findings on the effectiveness of ozone treatments, this study also emphasises the potential of ozone as a novel technology for pest control. Ozone offers several advantages as a pest control agent, including its ability to penetrate hard-to-reach areas. By harnessing the oxidative properties of ozone, we can effectively control pests while minimising the use of conventional chemical pesticides. In addition, ozone treatments leave no residue on treated areas, reducing concerns about chemical build-up and minimising the risk of environmental pollution. This makes ozone a compelling option for integrated pest management programmes looking for sustainable and environmentally friendly solutions to pest control challenges.

SE22 C05

POSTHARVEST TREATMENTS TO ADDRESS INVASIVE PESTS OF FRESH FRUIT COMMODITIES

Lisa Neven

USDA-ARS, Wapato, Washington, United States

The presence or potential presence of a pest in a shipment of fruit destined for an export market in a country where the pest does not currently exist can trigger the implementation of regulatory actions by the importing country. The application of a stand-alone postharvest quarantine treatment to prevent the accidental introduction of an invasive species to another country has remained a mainstay of meeting import requirements. There are numerous types of treatments that have been accepted either regionally or internationally as a means to mitigate the risk of accidental introductions. These treatments can be classified as physical, including temperature extremes, fumigation, irradiation, atmospheric extremes, systems approach, and combination treatments. This presentation will cover the types of treatments used to control postharvest pests in fresh fruits, advantages and disadvantages of each, and their level of use and international acceptance.

SE22 C06

THE RETURN OF THE KHAPRA BEETLE: ENEMY AT THE GATES

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The khapra beetle, *Trogoderma granarium* Everts (Coleoptera: Dermestidae) is a quarantine species over a large geographical areas, and it is included in the 100 worst invasive species globally. *Trogoderma granarium* can develop in more than 100 different types of dry products, but it is particularly important for grains and related amylaceous commodities, while its further spread has been considered as a major threat for global food security. The diapausing larvae of this species can easily survive for several years, and are particularly tolerant to most of the commonly used chemical control methods, such as contact insecticides and fumigants, but also to non-chemical methods, such as extreme temperatures and controlled atmospheres. Moreover, *T. granarium* does not respond well in a wide range of traps and attractants, which means that its presence may remain undetected for long periods of time. Although this species has been successfully eradicated in some parts of the world, its interceptions have been increasing and are expected to increase further, due to climate change and the intensification of international trade, especially in the case of durable agricultural commodities. Considering that durable products, such as wheat, rice and maize, are critical food security indicators, the detection and control of *T. granarium* through harmonized strategies is of outmost importance.

SE22 C07

A HOLISTIC APPROACH IN MANAGING BIOLOGICAL INVASIONS IN EUROPE: THE FRUIT FLY PARADIGM

Nikos T. Papadopoulos and Marc de Meyer and the FF-IPM partners

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Climate change, human mobility and trade globalization facilitate dispersion and establishment of harmful organisms into new areas. Europe is one of the hot-spots for biological invasions with large numbers of interceptions and detection recorded every year that lead to successful establishment and spread of several pests. Fruit flies (Diptera: Tephritidae) is one of the most important groups of invasive pests in Europe. Approximately 1/3 of all interceptions in fruits and vegetables count for fruit fly species, and currently there are reports on detections and outbreaks of major invasive pest, such as *Bactrocera dorsalis* in few EU countries. To support the management of fruit fly invasion in Europe, the recently concluded Horizon 2020 funded project FF-IPM (GA 818184) generated new biological knowledge for the three target species (*B. dorsalis*, *Bactrocera zonata* and *Ceratitis capitata*) and generated a range of interceptions and detection tools and approaches, including electronic traps,

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multientry identification tools for larvae and adults and a list of molecular ID tools. Novel bioclimatic and population growth models, as well as analysis of fresh fruit trading were used to develop a list of dynamic predictive tools and a Pan European alert system for invasive fruit flies. Novel detection tools and strategies have been developed as well. Tackling the range expansion of *C. capitata* to central Europe and the continental northern parts of the Mediterranean countries, the FF-IPM project develop biological control tools for off season management. Last but not least a novel in silico coordinated management approach to address the pest off and on season has been developed and pilot tested giving promising results. The current paper summarizes the main findings of the FF-IPM project and discusses the fruit fly paradigm as a model for managing invasive pests of agricultural importance in Europe and elsewhere.

SE22 C08

INVASIVE MEDFLY PROPAGULES IN MOSAIC LANDSCAPES: BEHAVIOR, DEVELOPMENT, ESTABLISHMENT AND DETECTION

Slawomir A. Lux

inSilico-IPM, Poland

The behaviour and survival of invasive propagules or dispersed remnant populations in mosaic landscapes are the least understood processes in invasion biology. The same is true of the role played by scattered, ephemeral food sources that occur in semi-wild or suburban environments. Occasional detections of medfly in areas recognized as fly-free raise a theoretical and regulatory controversy that can be reduced to the question: “*how long can local populations survive in the cryptic ultra-low-density phase and remain undetected by the surveillance network*”. Due to their local occurrence and ultra-low densities, these phenomena elude empirical research. To gain necessary insight, a series of stochastic simulations were carried out, emulating the behavior, development and fate of individual members of the residual / initial medfly populations. For this purpose, the PESTonFARM model was used, parameterized according to current knowledge about medfly biology and its behavior in the field conditions. The simulations were carried out for a hypothetical area with known sporadic Medfly presence, taking into account the key features of local topography, historical weather patterns, host phenology and a surveillance system. The projections generated by the model support the hypothesis about the possibility of the prolonged existence of cryptic populations fluctuating at very low densities, which may slip out of the surveillance grids and remain undetected for longer periods of time. Analysis of various scenarios emulated by the model has revealed a number of specific environmental factors that, acting simultaneously, are able to maintain fly populations at low densities. The application of the model also allowed estimating the chances of detection for various scenarios of terrain topography, host and weather patterns. The theoretical and practical implications of the proposed approach and methodology, its key merits and limitations, were also discussed.

SE22 C09

RECENT ADVANCE IN BIOCONTROL OF KEY INVASIVE HORTICULTURAL AND FIELD CROP PESTS IN SUB-SAHARAN AFRICA: A VITAL TACTIC FOR THEIR AGROECOLOGICAL MANAGEMENT UNDER CHANGING CLIMATE

Samira A. Mohamed, Subramanian Sevgan, Francis Obala, Henri Tonnang, Komi Mensah Agboka, Thomas Dubois, Abdelmutalab Azrag, Beatrice Muriithi, Benignus Ngowi, Shepard Ndlela, Elfatih Abdelrahman, Sunday Ekesi

Africa is most vulnerable to and most affected by climate change, with far reaching consequences, including increasing the frequency of insect pests' invasion. This is further exacerbated by the continent's poor quarantine infrastructure and undertrained phytosanitary personnel. In the last few decades, the continent was invaded by several devastating pests, such as Diamond Back Moth (DBM), *Plutella xylostella*; fruit flies (*Bactrocera dorsalis*, *B. zonata*); South American tomato leaf miner, *Phthorimaea* (=Tuta) *absoluta*, and fall armyworm, *Spodoptera frugiperda*. The establishment and wide spread of these pests resulted in massive yield losses of their respective host plants with implication on food security across the continent. In a desperate attempt to manage these pests, growers' resort to excessive use of insecticides, with catastrophic impact on “One Health”. Here we aim to highlight efforts undertaken to tackle these pests using a nature-based approach, with focus on use of parasitoids. The efficiency of native and introduced parasitoids (*Diadegma semiclausum*, *Cotesia vestalis* (both targeting DBM), *Fopius arisanus* and *Diachasmimorpha longicaudata* (both targeting *B. dorsalis*); *Dolichogenideia gelechiidivoris* (Targeting *P. absoluta*); and *Cotesia icipe*, *Telenomus remus*, *Cotesia marginiventris*, *Chelonus insularis* (targeting FAW) were evaluated under laboratory and field conditions and their interaction with native parasitoid determined. Also, performance of the parasitoid under current and future change scenario was assessed using different modeling approach. The introduced parasitoids species demonstrated high efficiency against their respective target pests and overall, they were able to outcompete their respective indigenous counterpart. The suitable area for establishment for the two-fruit fly parasitoids species are essentially confined to Sub-Saharan Africa, South America and Southeast Asia with projected expansion, especially in southern Africa. The released parasitoids were able to spread up to 8 km in one season, with the model showed that *F. arisanus* spread to approximately 229.97 km² across Africa. The introduced parasitoid species as well as some of indigenous parasitoids in case of FAW, led to substantial suppression of their respective target pest and should be an integral component of agroecological management approach of these pests in the face of climate change.



SE22 001

COMBINED METABOLOME AND TRANSCRIPTOME ANALYSIS REVEAL THE MECHANISM OF HIGH TEMPERATURE TOLERANCE ON *SPODOPTERA FRUGIPERDA* UNDER GLOBAL CLIMATE CHANGE

Yan X.^{1,2}, Li Z.^{1,2}, Zhao Z.^{1,2}

1. College of plant Protection, China Agricultural University, Beijing, China, 2. Sanya Institute, China Agricultural University, Hainan, China

The rapid invasion in agricultural pest and severe climate change pose immense challenges to global food security. *Spodoptera frugiperda* (J. E. Smith), the initially detected in Yunnan province, China on early 2019 and was listed as the first category of crop pests and diseases by Ministry of Agriculture and Rural Affairs of China on 2020. As a highly adaptable polyphagous migratory pest, *S. frugiperda* possesses a certain high temperature tolerance, which can propagate in harsh temperature environment, but the mechanism of high temperature tolerance through transcriptional and metabolic regulation to date is poorly understood. Here, through bioassays of multiple temperature treatments (30°C, 32°C, 34°C, 36°C and 38°C) for three tissues and multi-omics combinations, we found several physiological indexes decreased with the increase of temperature, however, the survival rate was still high at 38°C. Several chitin family genes were significantly differentially expressed and enriched to amino sugar and nucleotide sugar metabolism of KEGG pathway and chitin binding of GO pathway. Metabolites are also associated with the biosynthesis of keratin, xylocarane and wax, carbohydrate conversion and amino acid biosynthesis. The results of the combined analysis of transcriptomics and metabolomics showed correlation of high quality, which was consistent with the results of separate analysis. After RNAi (RNA interference), the relative expression of filtered genes was decreased and knockout of the gene using CRISPR/Cas9 (Clustered Regularly Interspaced Short Palindromic Repeats, Associated Proteins 9) technology significantly increased the sensitivity of the knockout homozygous strain to high temperature. Our results provided new insights for understanding the high temperature tolerance field control and of *S. frugiperda*.

SE22 002

FIGHTING A GOOD FIGHT AGAINST AN INVASIVE PATHOGEN THROUGH THE BOXWOOD BLIGHT INSIGHT GROUP

Daughtrey M., Luster D., Calabro J., Gray J., Hall C., Weiland J., Baysal-Gurel F., Gouker F., Kong P., Crouch J., LaMondia J., Pscheidt J., Santamaria L., Shishkoff N., Snover-Clift K., Hong C.

Virginia Tech, Virginia Beach, United States

Boxwood blight caused by *Calonectria pseudonaviculata* was first reported in the United Kingdom and New Zealand in the mid-1990s and officially confirmed in the United States in 2011. It has since spread to thirty U.S. states. The aim of our collaboration has been to protect boxwood, an iconic landscape plant, major evergreen crop and important forest tree species from the blight. To effectively fight against this emerging disease, scientists from twelve labs in seven U.S. states teamed up with domestic and foreign stakeholders

and collaborators in 2020 to establish an international consortium – the Boxwood Blight Insight Group (BBIG). As of August 2023, the BBIG team has published 49 refereed journal articles on a range of important topics from diagnostics to pathogen biology, boxwood resistance and microbiome, plus mitigation strategies. Its major educational products included: an International Boxwood Seminar series consisting of eight well-attended webinars with speakers from seven countries and attendees from five continents; quarterly BBIG Newsletters and Monthly digests; On-Demand sessions in 2021 and 2022 at Cultivate - the largest annual green industry educational event hosted by AmericanHort; as well as over 130 outreach email notifications on the latest research results and educational opportunities to the 400-member global BBIG community. Additionally, the team has organized one Idea Café and two symposia at the annual meetings of the American Phytopathological Society and hosted five international boxwood blight epidemiology meetings. More recently, we launched an BBIG Boxwood Seminar series. Information from meetings, newsletters, research publications, webinar recordings, diagnostic tools etc. are available at the BBIG website. These activities have fast-tracked the U.S. and the world onto a more sustainable path toward healthy boxwood production and plantings. BBIG was recognized with the 2022 USDA National Institute of Food and Agriculture Partnership Award for Program Improvement through Global Engagement.



CONFERENCE I

11.30-14.00 Concurrent Session **18B**

Fungicide Resistance: Diagnosis, Risk Assessment and Management, Integrated Pest Management

Chair: George Karaoglanidis (Aristotle University, Greece)

Dolores Fernandez Ortuno (University of Málaga, Spain)

SE18 C06

USE OF BACILLUS AMYLOLIQUEFACIENS QST713 AND CLONOSTACHYS ROSEA IK726 AS BIOCONTROL AGENTS TO CONTROL MULTIDRUG RESISTANT STRAINS OF BOTRYTIS CINEREA

Mpalafas A.¹, Sofianos G.¹, Tzelepis G.², Karaoglanidis G.¹

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Botrytis cinerea is classified as a high-risk pathogen for fungicide resistance development. Accumulation of target-site mutations and overexpression of efflux transporters can lead to Multiple (MLR) and Multidrug (MDR) resistance, respectively. Apart from contributing to fungicide resistance, little is known as to whether efflux pumps can also bind to secondary metabolites of potential biocontrol agents (BCAs), thus rendering MDR strains more resistant to BCAs. In this study, we tested the ability of *Bacillus amyloliquefaciens* QST713 (Ba QST713) and *Clonostachys rosea* IK726 (Cr IK726) to control the MDR strains both *in vitro* and *in planta*. *B. cinerea* strains with different types of multiple and multidrug resistance (MLR, MDR1, MDR1h) and sensitive strains were used. Ba QST713 was evaluated *in vitro* with the method of dual cultures in solid substrate and Cr IK726 with the development of the pathogen in the crude extract of the BCA. The *in planta* evaluation of the BCAs was carried out by applying them on bean plants, 24 h pre inoculation. Fludioxonil was used as a chemical reference treatment. The results concerning the *in vitro* experiments showed that both BCAs could effectively inhibit the growth of the strains. Ba QST713 caused an inhibition zone around the pathogen and reduced its growth by up to 60%, while, Cr IK726 inhibited partially or even completely the growth of some strains. In *in planta* experiments, MDR strains were more susceptible to Ba QST713 than sensitive ones, highlighting a potential fitness cost of them. In conclusion, the BCAs can reduce disease incidence and severity caused by the resistant strains and in some cases, they provide similar or higher efficacy compared to fludioxonil. The use of BCAs shows high potential and is a promising way to manage MLR/MDR strains.

Acknowledgments: The project was supported by H.F.R.I. under the "2nd Call for H.F.R.I. Research Projects to support Faculty Members & Researchers" (Project Number: 2959)

SE18 C07

OVERVIEW OF THE FUNGICIDE SENSITIVITY STATUS OF BARLEY PATHOGENS IN EUROPE

Gerd Stammer, Anna Glaab, Mascha Hoffmeister, Dieter Strobel

BASF SE, Limburgerhof, Germany

Ramularia leaf spot caused by *Ramularia collo-cygni* has become one of the most important diseases in barley in Europe. *R. collo-cygni* is classified as a high-risk pathogen by the Fungicide Resistance Action Committee (FRAC) and resistance evolution is known for this pathogen. Quinone outside inhibitors (QoIs) have been highly effective for *Ramularia*-control, but resistance developed soon after market introduction with a rapid spread over Europe. Efficacy of other fungicides, such as succinate dehydrogenase inhibitors (SDHIs) and demethylation inhibitors (DMIs) – is also affected by acquired resistance. Mainly target site mutations are responsible for the reduction of sensitivity of *R. collo-cygni* towards SDHIs and DMIs, but the mechanisms are complex, and compounds of both classes still contribute to disease control. Net blotch is caused by *Pyrenophora teres*, which has developed SDHI-resistance by various target site mutations with different levels of adaptation. QoIs are still effective, since this pathogen did not develop broadly the G143A mutation, which causes strong QoI resistance. Most adapted isolates carried the F129L mutation, which are still controlled in the field by some QoIs, such as pyraclostrobin. A single *P. teres* isolate carrying the G143A has been identified and it could be shown that this was caused by a gene transfer from *Pyrenophora tritici-repentis*. Leaf scald, caused by *Rhynchosporium secalis*, can be effectively controlled by DMIs, QoIs and SDHIs. There have been single cases for QoI resistance, caused by the G143A mutation, since more than a decade, but such strains did not distribute further for unknown reasons and are rarely found. SDHI adapted isolates, caused by the N85S mutation the SDH-C gene, have also been rarely detected. Monitoring studies need to be continued to identify resistance cases and their spread and to understand their relevance for field control.

SE18 C08

STATUS OF RESISTANCE DEVELOPMENT OF IMPORTANT POTATO PATHOGENS IN EUROPE

Juergen Derpmann, Andreas Mehl

Bayer AG, CropScience Division, Monheim, Germany

Potato production is threatened mainly by two foliar plant diseases in case weather conditions are favorable: Early Blight, caused by *Alternaria solani*, and Late Blight, caused by *Phytophthora infestans*. To control these pathogens fungicides are widely used. In the recent past, *P. infestans*



strains and *A. solani* strains adapted to selection pressures from commonly used fungicides from various modes of action were reported. To monitor the spread of such phenotypes, samples were collected in important potato growing regions in Europe in 2023 as well as previous years. Additionally, molecular tools were applied to identify the mutations responsible for detected phenotypes. In both pathogens isolates were detected showing accumulation of mutations leading to resistance to multiple modes of action, which have implications for resistance management strategies. Cross-resistance studies were conducted to demonstrate the impact of detected mutations and mutation-combinations on selected compounds from the group of QoIs and SDHIs as well as OSBPIs and CAAs. Full sensitivity was confirmed for DMIs and the newly elucidated mode of action of Vacuolar-type Proton ATPase of Fluopicolide. Lastly, greenhouse experiments were conducted to elucidate the impact of the detected mutations and mutation-combinations on commercially available solutions. It's implications on fungicide resistance management strategies are discussed.

SE18 C09

UNRAVELING THE MOLECULAR BASIS OF DMI RESISTANCE IN *CERCOSPORA BETICOLA*

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Cercospora leaf spot (CLS), caused by the fungal pathogen *Cercospora beticola*, is the most destructive disease of sugar beet worldwide. Resistance to the sterol demethylation inhibitor (DMI) fungicide tetraconazole has been previously correlated to synonymous and non-synonymous mutations in *CbCyp51*. We have extended these analyses to the DMI fungicides prothioconazole, difenoconazole, and mefentrifluconazole in addition to tetraconazole to confirm whether the synonymous and non-synonymous mutations at amino acid positions 144 and 170 are associated with resistance to these fungicides. Nearly half of the 593 isolates collected in this study were resistant to all four DMIs. Another 20% were resistant to tetraconazole and prothioconazole but retained sensitivity to difenoconazole and mefentrifluconazole. We found five *CbCyp51* haplotypes and associated them with phenotypes to the four DMIs. One mutation, L144F, is found in two codon variants TTC and TTT, where TTC is associated with resistance and TTT is associated with sensitivity even though both codons encode phenylalanine. Additionally, resistance is also strongly associated with the synonymous mutation E170. Quantitative PCR did not identify obvious trends in *CbCyp51* expression between haplotypes. However, mutant strains where the native *CbCyp51* gene is swapped with a different haplotype has confirmed the role of these mutations with DMI resistance. These results will be presented.

SE18 C10

RNA INTERFERENCE, A NOVEL STRATEGY TO BE INCORPORATED INTO INTEGRATED PEST MANAGEMENT PROGRAMS FOR THE CUCURBIT POWDERY MILDEW CONTROL

Dolores Fernández-Ortuño^{1,2}, Nisrine Bakhat^{1,2}, Laura Ruiz-Jiménez^{1,2}, Leonardo Velasco³, Alejandro Perez-Garcia^{1,2}

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Powdery mildew fungi are plant-pathogenic ascomycetes that are among the most common and important plant fungal pathogens causing diseases in a broad range of angiosperms, including both dicots and monocots. Major crops, such as various vegetable crops are susceptible to powdery mildews, but cucurbits are arguably the group mostly affected. In Spain, the disease is caused by the biotrophic fungus *Podosphaera xanthii*. At present, the management of this disease is based on the use of resistant varieties and the application of fungicides; however, the rapid development of new races of the pathogen, the resistance generated to the most frequently employed mildewcides and the European Green Deal ("farm to fork" strategy) make necessary the development of alternative control tools and molecules with novel fungicide activity to be integrated into the IPM programs. In this study, the potential of the RNA interference (RNAi) technology to design "dsRNAs fungicides" to be used as "bio-fungicides" in crop protection was tested. For that, essential genes involved in *P. xanthii* fungal development and disease symptoms such as key proteins necessary for fungal respiration (FRAC group 7), cytoskeleton (FRAC group 1) and methionine biosynthetic pathway (FRAC group 9) were targeted. These oligonucleotides were tested alone and in combination with commercial fungicides at low doses such as boscalid, cyprodinil, fluopyram and thiophanate-methyl. In addition, the use of nanotechnology to improve the efficacy of RNAi for the control of cucurbit powdery mildew was also explored. For that, the encapsulation of the dsRNAs molecules in carbon dots (CDs) nanoparticles was carried out in greenhouse experiments to test their efficacy. The results obtained could make a major contribution towards integrated pest management and sustainable agriculture of this important disease worldwide.

This research work is part of the R+D+i project PID2022-1362400B-C21, funded by MCIN/AEI/10.13039/501100011033/FEDER, UE.



SE18 C11

CHARACTERIZATION OF A NOVEL MFS TRANSPORTER IN BOTRYTIS CINEREA THAT CONFERS PARTIAL RESISTANCE TO BOSCALID AND FLUOPYRAM

Georgios Sofianos¹, Georgios Tzelepis², Edoardo Piombo², Mukesh Dube^{2y}, Magnus Karlsson², Georgios Karaoglanidis¹

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Botrytis cinerea is a notorious pathogen causing pre- and post-harvest spoilage in many economically important crops. Excessive application of site-specific fungicides to control the pathogen has led to the selection of strains possessing target site alterations associated with resistance to these fungicides and/or strains overexpressing efflux transporters associated with multidrug resistance (MDR). MDR in *B. cinerea* has been correlated with overexpression of *atrB* and *mfsM2*, encoding an ATP-binding cassette (ABC) and a major facilitator superfamily (MFS) transporter, respectively. However, it remains unknown whether other transporters may also contribute to MDR phenotypes. In the current study, the transcriptome of a *B. cinerea* multidrug resistant (MDR) field isolate, was analyzed upon exposure to the fungicide fludioxonil, and compared to the BO5.10 reference isolate. The transcriptome of this field isolate displayed significant differences as compared to BO5.10, including genes involved in sugar membrane transport, toxin production and virulence. Among the induced genes in the field isolate, even before exposure to the studied fungicide, were several putatively encoding ABC and MFS transmembrane transporters. Overexpression of a highly induced MFS transporter gene, namely *mfs3*, in the BO5.10 isolate, led to an increased tolerance to the fungicides fluopyram and boscalid, indicating an involvement in efflux transport of these compounds. Overall, the data from this study suggest a potential role of a newly characterized gene (*mfs3*) in boscalid and fluopyram detoxification and gives insights towards better understanding the molecular mechanisms involved in MDR, contributing to the development of more efficient control strategies against this pathogen.

The research project was supported by the Hellenic Foundation for Research and Innovation (H.F.R.I.) under the "2nd Call for H.F.R.I. Research Projects to support Faculty Members & Researchers" (Project Number: 2959).

SE18 C12

TOWARD UNDERSTANDING MECHANISMS OF FUNGICIDE RESISTANCE IN DOLLAR SPOT (CAUSAL FUNGUS: CLARIREEDIA JACKSONII, FORMERLY SCLEROTINIA HOMOEOCARPA) ON GOLF COURSE

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Dollar spot, caused by the fungus *Clarireedia jacksonii* (formerly *Sclerotinia homoeocarpa*), poses a significant threat to turfgrass, particularly in the field of turfgrass management such as golf courses. The economic impact

of dollar spot arises from its ability to cause direct losses through turf damage and the associated expenses related to disease management. While effective strategies for disease control and integrated pest management are employed, ongoing research into breeding resistant turfgrass varieties is essential for mitigating the economic consequences of dollar spot. However, the emergence of fungicide resistance in the causal agent, *C. jacksonii*, is a major concern. Dollar spot has shown resistance to four fungicide classes (benzimidazole, dicarboximide, demethylation inhibitor, and succinate dehydrogenase inhibitor). A recent publication highlighted reduced in vitro sensitivity to fluazinam, though no field resistance has been observed yet. The presence of cross- and multiple-resistance to fungicides and multi-drug resistance complicates disease management. Fungicide resistance in dollar spot is fueled by various mechanisms that enable the pathogen to withstand exposure to fungicides. Key mechanisms include target site modification, efflux pumps, and metabolic detoxification, but a comprehensive understanding of resistance mechanisms is still lacking. Turf management practitioners need to be aware of these mechanisms to implement effective fungicide resistance management strategies. This involves practices such as rotating fungicides with different modes of action, utilizing mixtures of fungicides, and integrating cultural approaches to minimize selection pressure. Regular monitoring for signs of resistance and adjusting management practices accordingly is imperative for achieving sustainable disease control.

SE18 C13

FOUR POINT MUTATIONS IN VACUOLAR H⁺-ATPASE SUBUNIT A CONFER FLUOPICOLIDE RESISTANCE IN PHYTOPHTHORA CAPSICI

Tan Dai^{1,2}, Fan Zhang¹, Sicong Zhang¹, Pengfei Liu¹, Can Zhang¹, Qin Peng², Jianqiang Miao², Xili Liu¹

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Fluopicolide, which is an excellent oomycete inhibitor, is classified as delocalization of spectrin-like proteins inhibitors by FRAC. In the current study, a *Pca-actin* knockout had no effect on the sensitivity of *Phytophthora capsici* to fluopicolide. The vacuolar H⁺-ATPase subunit a (PcVHA-a) was identified using a BSA-seq and DARTS assay. Four kinds of point mutations (G767E, N771Y, N846S, K847R) in PcVHA-a that cause fluopicolide resistance in *P. capsici* were confirmed using site-directed mutagenesis. The results of molecular docking, ATPase activity assays, and DARTS experiments provided further evidence of an interaction between fluopicolide and PcVHA-a. Sequence analysis and a molecular docking assay proved the specificity of fluopicolide to oomycetes or fish. These findings suggest that PcVHA-a is the target of fluopicolide, and H⁺-ATPase could be used as a novel target for the development of new fungicides. In addition, a unique combination of BSA-seq and DARTS was used in this study to quickly determine the target protein of the fungicides, providing a crucial means for the exploration of the target proteins of drugs.



BANQUET

18.00-20.30 Concurrent Session 23

Plant health phytiatry

Chair: Eris Tjamos (*Hellenic Society of Phytiatry, Greece*)

Amanda Hodges (*Entomology and Nematology Department, University of Florida, USA*) **Jeffrey Bradshaw** (*Entomology / Doctor of Plant Health, University of Nebraska, USA*)

SE23 C01

SEEDING THE FUTURE: EDUCATION OF PRACTITIONERS IN PLANT HEALTH

Dr. Jeffrey Bradshaw

Director, Doctor of Plant Health Program, University of Nebraska-Lincoln

The interdisciplinary Doctor of Plant Health (DPH) program was established by the University of Nebraska – Lincoln in 2009 with its first graduates in 2013. Since then, ~ 3 students per year of self-supported students have graduated to be employed in many areas of plant health. Through their interdisciplinary education and experiential learning activities, DPH graduates have become leaders in industry, universities, non-profit organizations, and private companies. These leaders have been instrumental in diffusing innovations throughout the varied landscapes in which plant health is important: e.g., from urban landscapes to large-scale, intensive agriculture, and from informal education to applied research. A diverse array of organizations have facilitated training of students during their DPH residency period. Case-study examples of our alumni will be presented and clearly show the value of advanced, professional degrees for co-creating solutions for the world's plant health problems.

SE23 C02

DPM-INTERNS AND OPPORTUNITIES FOR INTERDISCIPLINARY PLANT HEALTH SOLUTIONS

Dr. Amanda Hodges

DPM Director, Entomology and Nematology Department, University of Florida, Gainesville

The interdisciplinary Doctor of Plant Medicine (DPM) program was established by the University of Florida in 1999. Over 110 graduates have been employed in industry, regulatory plant health, and academic positions. This comprehensive training provided prepares student interns and graduates to serve as innovative problem-solvers within any plant health context. DPM students have completed internships in several countries, including Ecuador, Greece, Switzerland, and Brazil. Currently, the DPM program has an ongoing collaborative partnership with several institutions in Ecuador for student interns and also non-profit connections in Tanzania and the South Sudan. Throughout the U.S., DPM student interns have been placed in numerous states and U.S. territories, including the U.S. Virgin Islands. Agency hosts for student interns have varied. DPM student interns are ideally suited for programs interested in adapting to support graduate level internships. Employers of DPM graduates benefit from the practical and interdisciplinary approach of solving plant health problems.

SE23 C03

PLANT DOCTOR: AN EXPERT OF AGROENVIRONMENTAL SYSTEMS NEEDED FOR ONE HEALTH

Dr. Dimitris Tsaltas

Department of Agricultural Sciences, Biotechnology and Food Science, Cyprus University of Technology, Lemesos, Cyprus

One Health is an amalgamated, unifying approach that aims to sustainably balance and optimize the health of people, animals and plants. The health of all ecosystems are nowadays interconnected and only a total health perspective can support stable wellbeing of all living organisms. Mainstreaming 'One Health' means that we can more effectively predict, prevent, prepare, detect, and respond to global health threats. Plants are the base of the health pyramid or at least a corner of a tetrahedron as they interact with all living organisms (animals, humans and microbes). Plants are in direct contact with our environment (aerial and soil). Therefore, the understanding that plant health is of paramount importance in our planetary boundaries is very important. It requires our scientific and professional attention as plant scientists in general and as plant doctors specifically. Crop protection is a cornerstone of One Health and as in all ecosystemic services, a global, inclusive concept is required to understand that plants health is not easily achievable. Incorporating plant health into One Health discussions implies a stronger emphasis on ecological health through the trade-off between food security and planetary boundaries. As with other aspects of One Health, its value for plant health is to create more inclusive approaches evaluating plant protection interventions that address agricultural needs, but also realize co-benefits with ecosystem, animal, and human health. Pesticide and fertilizer use, heavy metals, mycotoxins, food security and food safety are affecting significantly our multi-system health. Plant health can affect humans even indirectly, when yields drop and food scarcity prevails. Famine and malnutrition are humankind's worst nightmares. It is then that cure of plant problems need deep and extensive knowledges understanding the biological system as well as the socioeconomic context of food and feed security and safety.

SE23 C04

ONE HEALTH: A NEW PERSPECTIVE TO HIGHLIGHT THE SUCCESSES OF PLANT HEALTH AND THE NEED FOR A NEW QUALIFICATION IN THIS FIELD

Dr. Ramon Albajes

Agrotecnio CERCA Center, University of Lleida, Lleida, Spain

FAO promotes a One Health approach as part of the transformation of the agri-food system for the health of people, animals, plants and the environment. One Health



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is a transdisciplinary approach used to address complex concerns related to human, animal, plant and ecosystem health. A health perspective can highlight the net benefits of progress in plant protection techniques (PPT): on the one hand, PPT reduces crop yield losses and thus helps increase agricultural productivity and global food security by mitigating malnutrition that still affects at least 10% of the world's population. On the other hand, the more efficient use of natural resources by the PPT and the reduction of environmental pollution favors the sustainability of the agri-food system by minimizing the unintentional negative impacts of phytosanitary practices on people, animals and ecosystems. Without a doubt, the consideration of the 'One Health' perspective helps to assess the advances in plant health in a cost-benefit analysis and justifies the need to intervene to control crop pests within the landscape framework. The vision of plant health simply as an element of the agri-food system already entails a certain complexity, which derives from the multiple interactions of biotic and abiotic factors, the increasing invasion and establishment of exotic organisms and the consequent emerging pests and social concerns on food safety and environmental pollution. If we add the 'One Health' perspective, the complexity of the scientific base and practice increases and demands a new holistic framework for R&D&TT and training. Based on my experience for more than 40 years of university training in Spain, I fully support the initiatives aimed at the creation of a degree and master's degree in Plant Health. Introducing Phytriatic/Plant Health as a novel degree in the current university curricula would provide a better prospective to fulfill the key role of Plant Health in the One Health approach. Contrary to that, the trend of recent changes in agronomy curricula in Europe has gone in the opposite direction and plant health has rarely been promoted in many European university degrees.

SE23 C05

PHYTIATRICS AND ENVIRONMENT AS A MASTER OF SCIENCE: A VANGUARD IN CROP PROTECTION

Christos G. Athanassiou*

Laboratory of Entomology and Agricultural Zoology, Department of Agriculture, Crop Production and Rural Environment, University of Thessaly, Greece

In 2018, a novel Master's course program has been launched at the Department of Agriculture, Crop Production and Rural Environment, University of Thessaly in Greece: Phytiatrics and Environment. Until today, more than 40 students graduated from that course, with the vast majority of them working professionally in the field of crop protection. This Master of Science is practically one of the first of its kind in phytiatrics in Europe, and apparently the first of its kind in Greece. In this context, emphasis is given in the field of diagnostics, but also in the area of modern protocols in integrated pest, disease and weed management, with multiple activities towards this direction. This program is indicative of the feasibility in organizing and operating similar actions in Universities in Greece, and proves to be all the more modern in a dynamically changing environment in the science of plant health and the agro-food chain.

SE23 C06

PLANT HEALTH ERASMUS MUNDUS JOINT MASTER DEGREE AT SIX UNIVERSITIES IN EUROPE INTERNATIONAL, APPLIED, SYSTEM-AND RESEARCH ORIENTED

Susanne Weigand, Josep Armengol, Alberto Pozzebon

Georg-August University Göttingen, Göttingen, Germany

Highly qualified experts in the field of crop production and plant health are urgently needed, if the objective of producing twice as much food with half the ecological footprint by 2050, is to be achieved. However, in recent years research and teaching in plant health have taken a sharp turn towards basic approaches. This has resulted in significant advances in fundamental techniques, but has been associated with a loss of the systems-related knowledge and expertise needed to deal with the complexity of pest and disease problems encountered in practice. The displacement of agricultural sciences as systems science by basic disciplines ignores the fact that multidisciplinary and multifaceted approaches are required to ensure further advancement to address the challenges of agriculture. The "Erasmus Mundus Master's Degree in Plant Health in Sustainable Cropping Systems" was first selected by the European commission for funding in 2015, when the consortium received a total funding of 2.8 Mio Euro over a period of five years to provide full scholarships for highly qualified students and has been extended twice for another 4 and 6 years until 2030. The Master PlantHealth is designed by five leading European Higher Education Institutions, the Universities of Göttingen (Germany), Universitat Politècnica de València (Spain), Università di Padova, (Italy), L'Institut Agro and AgroParisTech, (France). It offers a high-level academic program based on the most up-to-date research and cooperates with federal research institutions, legal authorities and the agrochemical industry at different levels to educate a new generation of young academics, who will take the responsibility in future research and innovation in crop health management. The number of applications has been continuously increasing coming from > 80 countries. Graduates very successfully apply for PhD positions at renowned universities worldwide and find numerous employment opportunities in companies; research centers; organizations and research institutes both in Europe or their home countries.



SKALKOTAS

18.00-20.50 Concurrent Session 208

Biopesticides and Biofertilizers

Chair: Sotiris Tjamos (Agricultural University of Athens, Greece)

Panagiotis Sarris (Microbiology, Department of Biology University of Crete, Greece)

SE20 001

DEVELOPMENT OF RNA-BASED FUNGICIDES AGAINST FUSARIUM DISEASES OF CEREALS

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1. Centre for Horticultural Science, Queensland Alliance for Agriculture and Food Innovation, Brisbane, Australia, 2. School of Agriculture and Food Sustainability, The University of Queensland, Brisbane, Australia

Fusarium Head Blight (FHB) is a fungal disease affecting grain crops caused by *Fusarium graminearum*. It can lead to 20% - 100% of grain yield loss and produce mycotoxins during infection that are harmful to humans and animals. Chemicals such as Demethylation Inhibitors (DMI) are commonly used to reduce the incidence and impact of FHB. However, the overuse of these fungicides can cause potential harm to soil-beneficial microorganisms, affect soil structure and fertility, and lead to fungicide resistance. Biological controls are being investigated as an alternate control method, with RNA interference (RNAi) technology showing immense potential as a tool in agriculture to overcome pests and diseases. Our investigation delves into RNAi's potential as an exogenously applied solution for mitigating FHB in wheat and barley. Utilising confocal microscopy, we have substantiated the environmental absorption of exogenous dsRNA into germ tubes of *Fusarium graminearum*, albeit with varying efficiency. Building upon these findings, we devised an in vitro screening method employing target-specific dsRNA to explore fungal inhibition. Encouragingly, our in vitro assays reveal noteworthy fungal inhibition, suggesting scalability. In planta seedling assays using the *F. graminearum* – wheat/barley pathosystems were also tested by spray application of dsRNA. Intriguingly, our in planta experiments demonstrated significant inhibition of the *F. graminearum* infection in barley, contrasting with modest effects in wheat, potentially due to disparate uptake efficiency. Northern blots were used to detect dsRNA movement in these plant systems within the leaf and heads, to answer this question. These results underscore the potential of utilising target-specific dsRNA as a biocontrol strategy against *F. graminearum*. However, further research is imperative to ascertain its efficacy against FHB in mature plants. This approach could be pivotal in curbing FHB's impact while circumventing the drawbacks associated with conventional fungicides.

SE20 002

BURKHOLDERIA SP. SSG – A SAFE AND POWERFUL BROAD-SPECTRUM BIOCONTROL AGENT AND BIOFERTILIZER

Kong P., Hong C.

Virginia Tech, VIRGINIA BEACH, United States

Burkholderia sp. SSG was an endophyte isolated from boxwood blight-reverted leaves of *Buxus sempervirens* 'Suffruticosa'. The objective of present study was to determine its potential as a biocontrol agent and biofertilizer, including health and environmental risk. SSG was evaluated on eight crops against a variety of diseases caused by fungi, oomycetes, bacteria and a virus, and also on boxwood for growth promotion under controlled environments and field production settings. It was also assessed for health/environmental risks using four standard methods - restriction fragment length polymorphism analysis, PCR with specific primers for the Burkholderia cepacia complex (Bcc) epidemic strain marker, onion maceration assay and genome sequencing. SSG provided excellent control of boxwood blight caused by *Calonectria pseudonaviculata* (Cps). Specifically, SSG at 10^7 cfu/mL lysed all conidia in broth culture. SSG at 10^8 cfu/mL reduced blight of container-grown boxwood by >98% when applied 1 day before or 3 hours after plants were challenged with Cps under controlled environments. Its blight control efficacy decreased with decreasing bacterial concentration and increasing lead time. When applied monthly onto field-grown boxwood plants in western North Carolina, SSG provided a similar level of blight control as the fungicide standard – Concert II applied at 3-week intervals. SSG also was effective against a variety of other diseases caused by *Alternaria tenuissima*, *Botrytis cinerea*, *Colletotrichum fructicola*, *C. gloeosporioides*, *Pseudonectria rouselliana*, *Phytophthora capsici*, *P. nicotianae*, *P. ramorum*, *Xanthomonas campestris*, and tomato spotted wilt virus on gravel pads or in a containment facility. Additionally, SSG promoted boxwood growth by 60 to 317%, cultivar dependent. SSG was identified as a new member of Bcc with distinct characters from known clinical strains as determined by all four analyses. SSG is a powerful broad-spectrum biocontrol agent and biofertilizer, and it is safe for field applications.

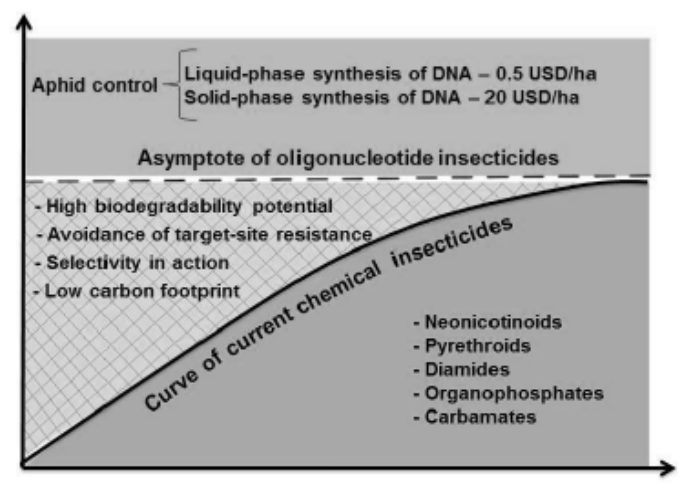
SE20 003

OLIGONUCLEOTIDE INSECTICIDES: LOW-COST INNOVATION FOR ECO-FRIENDLY APHID CONTROL AND AN ASYMPTOTE FOR CURRENT CHEMICAL INSECTICIDES

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Most technological innovations start out very expensive and don't work well, but eventually start working well and become very cheap. Contact unmodified antisense DNA (CUAD) biotechnology has come a long way for 16 years and today, for the control of aphids, this innovation has reached a price of less than a dollar per hectare, which creates unprecedented prospects for its large-scale application in agriculture. The CUAD platform has a number of characteristics that distinguish it from all known classes of insecticides: unmodified antisense DNA as the active substance, ribosomal RNAs as the target, DNA containment as the mechanism of action. We have shown that a low concentration (0.1 mg/L) of the oligonucleotide insecticide Schip-11 (5'-TGTGTTCTCGTTA-3'; complementary to ITS2 of polycistronic rRNA transcript) can lead to $76.1 \pm 7.7\%$ mortality of waxy grey pine needle aphid *Schizolachnus pineti* in the experimental group vs. 7.7 ± 0.1 in water-treated control group ($p < 0.01$) on the 12th day of the experiment. At a consumption rate of 200 L per hectare, the price of the required amount of oligonucleotide insecticide will be about 0.5 USD when using liquid-phase DNA synthesis and 20 USD when using solid-phase DNA synthesis. Oligonucleotide insecticides have unique characteristics: low carbon footprint, high safety for non-target organisms, rapid biodegradability, avoidance of target-site resistance. This next-generation class of insecticides creates opportunities that have never existed before in crop protection. Using flexible CUAD platform, any farmer, botanical garden or pest control company is capable of creating its own unique arsenal of insecticides, well-tailored for a particular population of insect pest. Aphids as key pests of important temperate crops that shape food security, can be controlled with oligonucleotide insecticides already today, ensuring prosperity for agricultural countries that implement this innovation.



SE20 004

POTENTIAL EFFECTS OF AN ORGANIC BIOSTIMULANT ON ZYMV TRANSMISSION AND INFECTION IN CUCURBITA PEPO L.

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Biostimulants are substances that promote plant growth and efficiency of nutrient uptake by improving plant and/or rhizosphere characteristics. The effect of a biostimulant containing alfalfa, brown algae, and molasses extracts on the interaction among zucchini plants (*Cucurbita pepo* L.), zucchini yellow mosaic virus (ZYMV) and aphid vectors was evaluated. ZYMV is an aphid-transmissible potyvirus that affects several crops and weeds worldwide. Several crop traits were compared between healthy and ZYMV-infected plants which were treated or untreated with biostimulant. Biostimulant-treated plants had increased growth with higher values of vegetative parameters (number of leaves and dry biomass weight) in healthy conditions and of reproductive parameters (number of flowers and fruits) in infected conditions. After treatment, the ZYMV titre decreased over time even if no differences in symptom severity was observed compared to the untreated control plants. Real-time PCR was used to quantify the expression of several plant defence genes, and the pathogenesis related gene 1 (PR1), involved in systemic acquired resistance, and the peroxidase gene (POD), belonging to oxidative stress pathway, were upregulated in ZYMV-infected plants treated with the biostimulant. The effect of biostimulant on the settling preference and life traits of the aphid vector *Myzus persicae* was also investigated. Aphid choice test experiments using detached zucchini leaves showed that fewer specimens settled on biostimulant-treated plants, with and without ZYMV. Biostimulant treatment led to reduced aphid survival and decreased offspring production, regardless of the infection condition of the plants. The potential role on aphid choice of volatile organic compounds (VOCs) emitted by treated and untreated plants was also assessed in olfactometer experiments. Taken together, these findings suggest that biostimulant treatment can reduce disease risk in *C. pepo* crops by potentially contributing to the concurrent control of ZYMV and its vector.

SE20 005

FUSARIUM OXYSPORUM FO12 STIMULATES DICOT PLANTS GROWN ON CALCAREOUS AND NON-CALCAREOUS SOILS

Sevillano-Caño J.¹, Agustí-Brisach C.¹, García-Mina J.², Zamarreño A.², Barrón V.¹, Sánchez-Rodríguez A.¹

1. Department of Agronomy (DAUCO, Unit of Excellence María de Maeztu 2020-2024), Campus of Agri-Food International Excellence at Rabanales (ceiA3), University of Córdoba, Córdoba, Spain, 2CIPAV TimacAGRO International-Roullier Group, Navarra, Spain

Fungal endophytes can establish mutually beneficial relationships with plants and significantly contribute to



plants' resilience against both biotic and abiotic stresses. This unique trait positions them as promising candidates for applications in plant biostimulation. The non-pathogenic *Fusarium oxysporum* FO12, an effective biological control agent against Verticillium wilt of olive, was found to promote plant growth in different plant species in short-term hydroponic or sterile calcareous soil experiments. However, a lack of knowledge on the effect of this fungal strain when plants are grown on natural soils and the effects of the inoculation methods were identified. For these reasons, we evaluated two FO12 application methods (seed dressing and soil treatment) to assess its impact on plant growth, nutrient uptake, and plant hormone production in sunflower plants under diverse natural soil conditions (calcareous and non-calcareous soils) throughout the entirety of a crop cycle. To this end, pot experiments under controlled conditions were developed. Plant analysis involved height, leaf chlorophyll content, phenological stages, and plant hormone content, culminating in biomass and mineral uptake assessments. Our findings indicate that both application methods of FO12 boosted plant height, and accelerated development regardless of soil conditions. Moreover, plant reproductive biomass was increased by 27% and 31% in non-calcareous and calcareous soils, respectively. Additionally, phosphorus, potassium, and zinc uptake were enhanced by 46%, 36%, and 12%, mainly through the soil treatment and the phytohormone profiles were modified, albeit with variations observed depending on the soil conditions. These results highlight the potential of FO12 as a plant biostimulant under distinct natural soil conditions, reflecting its efficacy in more realistic agricultural environments. However, our results suggest that the growth-promoting effects of FO12 may be correlated with soil fertility, and the two different inoculation methods may operate at different levels within the plant-soil system.

Funding: This research was cofunded by Spain's Ministry of Economy and Competitiveness (Project AGL 2017-87074-C2-2-R), by the Spanish Ministry of Science and Innovation (Projects PID2020-118503RB-C22 and PID2021-1236450A-I00 'BIOLIVE'). The authors acknowledge financial support from the Spanish State Research Agency through the Severo Ochoa and María de Maeztu Program for Centers and Units of Excellence in R&D (Ref. CEX2019-000968-M).

SE20 006

CONTRASTING EFFECTS OF SEED TREATMENT WITH FUNGAL CELL WALLS ON TOMATO AND ARABIDOPSIS

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The application of plant-beneficial fungi offers promising avenues for enhancing crop growth and resistance to biotic and abiotic stresses. We investigated the effect of different fungal cell components on plant immune components of different plant species. Here, we investigated the potential of fungal cell walls, particularly those of, in conferring plant-beneficial traits. We applied lyophilised cell wall extracts of the model basidiomycete fungus *Schizophyllum commune* as seed coatings on tomato and Arabidopsis seeds and evaluated their impact on plant growth and resistance to various pathogens. Our results revealed species-specific responses. In tomato, seed treatment with *S. commune*

cell walls promoted seedling growth and induced systemic resistance against *Botrytis cinerea* accompanied with enhanced expression of key defence genes *PI550* and *PI440*. However, in Arabidopsis seedling growth was reduced and susceptibility to *Botrytis* was enhanced. Further analysis demonstrated that seed pre-treatment with *S. commune* cell walls reduced reactive oxygen species (ROS) levels in Arabidopsis leaves and suppressed MAMP-triggered ROS bursts in roots. However, MAMP-triggered ROS bursts were stronger in tomato after seed pre-treatment with *S. commune* cell walls. Chitin and β -glucans, two of the most abundant components of the *S. commune* cell wall, were implicated as key players in differentially altering plant resistance and ROS bursts in different seed coated plants. In conclusion, our findings suggest that lyophilised mycelium of *S. commune* holds promise as a seed coating for promoting plant growth and resistance to pathogens. However, the bioactive effects of these materials are plant species-dependent and may arise from additive interactions of cell wall components. Understanding the mechanisms underlying these effects will facilitate the development of alternative plant protection products with consistent efficacy across different crops and field conditions.

SE20 007

BIOCONTROL WITH PHAGES

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Bacteriophages are viruses that infect and replicate within bacteria. They are the most abundant and diverse entities in the biosphere, playing an important role in regulating bacterial populations in various environments. Phages have attracted significant attention for their potential applications in medicine, particularly in the field of phage therapy, but also represent a fascinating area of research with prominent applications in agriculture. The aim of our study was to isolate and characterize lytic bacteriophages infecting the phytopathogens (i) *Pseudomonas syringae* pv. *tomato* (*Pst*), the causal agent of the bacterial speck of Tomato, and (ii) *Xanthomonas campestris* pv. *campestris* (*Xcc*), causing black rot, one of the most devastating diseases of Brassicas. Using soils collected from various fields in Northern Greece, we isolated upon enrichment, the lytic *Pseudomonas* phage *dad_01* and the lytic *Xanthomonas* phages *dad_01* to *dad_07*. The *Pseudomonas* phage *dad_01* was specific to *Pst* strains, stable in a wide pH range and also stable to temperatures up to 40 °C. The *Pst* resistance frequency to this phage was estimated to 6.23×10^{-5} . All *Xanthomonas* phages were specific to *Xcc* strains and stable in a wide pH range. Yet, they displayed different degrees of thermotolerance and also differed in their killing efficacy as revealed by Efficiency of plating (EOP) and planktonic killing assays (PKA). The *Xcc*



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resistance frequency to the isolated *Xanthomonas* phages was ranging from 2.11×10^{-4} (for the phage dad_02) to 7.53×10^{-5} (for the phage dad_03). Importantly, a cocktail of the isolated *Xanthomonas* phages reduced the average resistance frequency by 14.77-fold suggesting that phage cocktails may be more appropriate for biocontrol in the field. Bioassay conducted in tomato and broccoli revealed that the newly isolated phages were capable of drastically reducing disease severity suggesting that phages hold promise as biocontrol agents.

SE20 008

EXPLORING THE MECHANISMS UNDERLYING THE ACTION OF TRAMESAN, AN EXO-POLISACCHARIDE FROM *TRAMETES VERSICOLOR*, AS PLANT RESISTANCE INDUCER IN *ARABIDOPSIS THALIANA*

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Environmental Biology Department, Sapienza Università di Roma, Rome, Italy

Crop pathogens causes huge loss in food production reducing the yield and quality of agricultural products. To counteract plant pests, chemical fungicides are widely used. However, the intensive use of agrochemicals leads to the emergence of resistant pathogens strains. Moreover, pesticides pose several risks for the environment and human health. The European regulation 128/2009 governing the use of pesticides has recently removed several products from the market, promoting the employment of non-synthetic (natural) compounds in agriculture. In this context, Tramesan, an exopolysaccharide derived from the basidiomycete *Trametes versicolor*, is emerging as a natural, user-friendly, low-cost alternative to protect crops. In fact, in addition to strongly inhibits aflatoxin biosynthesis by the mycotoxigenic fungus *Aspergillus flavus*, it has been previously demonstrated that Tramesan enhances the resistance of wheat against Septoria Leaf Blotch Complex (SLBC), through the induction of plant defence responses. However, the mechanism by which Tramesan elicits plant defences remains unclear. Here, we explored the mechanisms underlying the action of Tramesan in the model plant *Arabidopsis thaliana*. In particular, we performed infection assays with *Botrytis cinerea* in *Arabidopsis* plants sprayed with Tramesan and evaluated the induction of defense responses such as, the expression of defense genes and the H_2O_2 production. Moreover, we investigated the possible antifungal effect of Tramesan on *B. cinerea* by analysing *in vitro* the growth of the fungus and the germination of spores. The results suggest that Tramesan, other than slightly inhibiting the growth of *Botrytis cinerea*, has the ability to induce *Arabidopsis* defense responses to the fungus. In conclusion, our work provides a starting point for further molecular analysis that will allow us to clarify the mode of action of Tramesan and therefore to develop sustainable strategies for crop protection.

SE20 009

METABOLOMICS: THE TOOL OF CHOICE FOR THE DISCOVERY OF THE MECHANISM(S) OF ACTION OF BIOSTIMULANTS AND BIOPESTICIDES

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The recent discovery and development of biostimulants and biopesticides have provided solutions to challenges that the agri-food sector is facing. They can play significant role in modern agriculture, offering improved, sustainable and environmentally friendly alternatives to traditional pesticides and fertilizers. Nonetheless, knowledge on their mechanism(s) of action (MoA) is incomplete, thus, cutting-edge tools need to be employed. One such tool is metabolomics, which can offer global overview of biostimulant and biopesticide effect on the metabolism of target organisms, and subsequently can provide insights into the underlying mechanisms. Several analytical platforms can be employed in order to detect, identify, and quantify metabolites present in biological samples, such as gas chromatography-mass spectrometry (GC/MS), liquid chromatography-MS (LC/MS), and nuclear magnetic resonance (NMR) spectroscopy analyzers. These platforms can be employed individually or in combination to maximize metabolome coverage. Metabolomics is applied in the analyses of the metabolism of the organisms in question at predefined time intervals, in order to discover the fluctuations in metabolite levels. Metabolomics provide insights into the metabolic status of organisms following treatments and could result in the dissection of the MoA of bioactive agents being applied. However, interpreting results necessitates a thorough understanding of study context, including experimental design, sample type and objectives. Concluding on the plant growth promoting capacities, nitrogen fixation, and plants' priming could greatly assist the development of biostimulants and the understanding of their bioactivity. Additionally, information on the effect of biopesticides on the metabolism of target organisms could provide valuable information on their MoA. Principles and examples of metabolomics application in biostimulant and biopesticide research are presented. Biostimulants and biopesticides represent promising means for promoting sustainable agriculture, addressing the challenges of feeding a growing global population, while safeguarding the environment for future generations.



SE20 010

ANTAGONISTIC MICROORGANISMS AND BIOSTIMULANTS FOR MANAGING FUSARIUM WILT OF TOMATO UNDER GREENHOUSE

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Fusarium wilt of tomato, also known as Fusarium wilt disease, is a fungal infection caused by *Fusarium oxysporum* f. sp. *lycopersici* (Fol) and represents one of the most destructive diseases affecting tomato worldwide. Biocontrol agents, physical methods, genetic resistance and grafting are the most tested measures, as alternative to chemical methods, for managing Fol. Biostimulants are substances or microorganisms that can enhance plant growth and stress tolerance. While primarily used to promote plant health and productivity, some biostimulants also show potential against plant diseases. Seven trials have been carried out on potted tomato plants cv Cuor di Bue in greenhouse to evaluate the efficacy of antagonistic strains of *Fusarium* spp., *Trichoderma* spp. and *Pseudomonas* sp., obtained from suppressive soils, substrates and compost, and of microbial and non microbial biostimulants against Fol. The microorganisms used were selected for their demonstrated effectiveness in controlling other soilborne pathogens on different crops. Biocontrol agents and biostimulants were applied with different methods, in nursery and at transplanting, and were compared to a commercial formulation of *Trichoderma* spp. and to tiophanate-methyl. Tomato plants were grown using a commercial peat substrate infested with 1 g/L of Fol race 1 fungal biomass. Several antagonistic *Fusaria* strains, particularly 233/1 RB, MSA35, and FC21, a microbial biostimulant containing mycorrhiza and two plant-based biostimulants significantly reduced by 40-70% fusarium wilt severity. This study suggests the potential effectiveness of antagonistic microorganisms and biostimulants in controlling Fol. Furthermore, it emphasizes the significance of optimizing application techniques to ensure effective management of Fusarium wilt disease.

SE20 011

EFFICACY OF NOVEL BIOFUNGICIDES IN CONTROLLING POWDERY MILDEW IN VINEYARDS

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Powdery mildew is a significant grape disease that affect the yield of grapes. Biological fungicides are an alternative that enables the use of chemicals in agriculture to be reduced. This study aimed to evaluate the efficacy of novel biological fungicide products, two composts compared to five biological commercial fungicides and one chemical fungicide in controlling powdery mildew in vineyards. Field trials were conducted from 2022 to 2023 on grapes of two Greek *Vitis vinifera* varieties, Moschato (white) and Agiorgitiko (red).

Fotofor®, a novel plant immunity inducer, developed by Vioryl S.A., containing a complex of vitamin B and an amino acid, KLEMAFORT®, developed by Vioryl S.A., a non-Saccharomyces yeast formulated in liquid suspension and two composts by Ikorganic 1) onion crop residues and green prunings b) onion crop residues, mushroom litter substrates and green prunings. Disease severity was assessed on leaves and the AUDPC (Area Under Disease Progress Curve) was calculated to represent the epidemic progress. At the most effective products, estimation of four phytohormones, salicylic acid (SA), methyl jasmonate (MeJA), indole acetic acid (IAA), abscisic acid (ABA) was determined by high-performance liquid chromatography-diode array detector (HPLC/DAD). Also, the evaluation of three genes PR1, SOD, and PAL was succeeding with the use of RT-qPCR. Fotofor®, exhibited superior efficacy across all experiments against powdery mildew more than 85%, following compost onion/mushroom, Klemafort®, Vacciplant®, Romeo®, Compost onion, Fytosave®, Sulphur 96DP, Botrybel®, Mevalone®. Application of Fotofor®, Compost onion/mushroom and Klemafort® to grapes significantly enhanced all hormone activities and only the Fotofor® show significant expression of genes SOD, PR1 and PAL compared to control. The results indicate promising novel fungicides with different mode of action, underscoring its potential significance in modern agriculture.

SE20 012

ADOPTION OF USE OF BIOLOGICAL NEMATOCIDES

Westerdahl B.

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A study was conducted to monitor the adoption of biological nematocides in California, USA. Data was collected from databases developed and maintained for 32 years by the California Department of Pesticide Regulation (https://www.cdpr.ca.gov/database_menu.htm). Six biological nematocides have registered active ingredients: *Myrothecium verrucariae* strain AARC-0255 (M, Ditera DF), *Quillaja* (Q, Nema-Q, Nemomex), *Purpureocillium lilacinum* strain 251 (P, Melocon WG), Abamectin (A, Divanem), *Burkholderia* sp. strain A396 (B, Majestene), and Azadirachtin (29 different brand names). Yearly data was collected for date of application, county, crop, active ingredient, product name, weight of product applied, weight of chemical applied, and area treated. With the exception of Azadirachtin (widely used as an insecticide as well as a nematocide), the data can be interpreted as indicating usage patterns for registered nematocides. For example, in 2021, M was applied 353 times in 13 counties on 14 different crops for a total of 11,115 kg of product applied to 1,505 ha; Q was applied 79 times in 6 counties on 5 different crops for a total of 5,087 kg of product applied to 727 ha; P was applied 373 times in 12 counties on 22 different crops for a total of 25,627 kg of product applied to a total of 1,103 ha; A was applied 132 times to turfgrass in 13 counties for a total of 247 kg of product applied to a total of 76 ha; and B was applied 41 times in 3 counties on 2 different crops for a total of 7,525 kg of product applied to a total of 631 ha. Biological nematocides have seen a steady increase in use. For example, in 1997, D was applied only 33 times in 3 counties on 5 different crops for a total of 1,809 kg of product applied to a total of 42 ha.



Concurrent Sessions

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SE20 013

COMING OF AGE: BIOSTIMULANTS AND BIOCONTROLS IN AGRICULTURE TO COMBAT THE CLIMATE CRISIS AND SECURE FOOD SUPPLY, AN R&D PERSPECTIVE

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MC2

18.00-20.30 Concurrent Session **24**

Emergent Forest Diseases in a Changing and Interconnected World

Chair: Matteo Garbelotto (Department of ESPM, UC Berkeley, USA)

SE24 C01

NOVEL LARGE SCALE TREE MORTALITY IN CALIFORNIA IS CAUSED BY LATENT PATHOGENS TRIGGERED BY CLIMATE CHANGE

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Until recently, disease caused by latent pathogens, i.e. pathogens that alternate between endophytic, pathogenic and saprobic phases, has been reported only in localized situations of acute plant stress. Surprisingly, since 2015, latent pathogens have been repeatedly identified as causes of the widespread mortality of trees and shrubs throughout Northern California. Isolations and pathogenicity tests revealed that disease of each of eight tree/shrub species was associated with latent pathogens in the genera *Botryosphaeria*, *Diplodia*, *Neofusicoccum*, *Dothiorella*, *Diaporthe* and *Pseudosydowia*. California is experiencing a megadrought: we hypothesized that drought may be a driver of the emergence of novel latent pathogens. By maintaining soil moisture just above the wilting point in a set of potted plants, we were able to show that disease severity increased significantly in plants that were drought-stressed and inoculated with latent pathogens. Finally, by using a model "host x pathogen" combination, we addressed whether disease severity in potted plants may be positively associated with drought, warmer temperatures, fungal strain and wounding, alone and together. Results showed that disease increased at warmer temperatures, while drought increased disease only at the warmer temperature. Presence of latent pathogens was significant in all treatments, while wounding on its own was significant only in the hot and dry treatment. Results further suggest that warmer temperatures do not increase disease because of their effect on fungal strains, but because of their effect on plant physiology. Our data support a major role of latent pathogens in causing large scale tree mortality in association with climate change.

SE24 C02

THOUSAND CANKERS DISEASE THREATENS JUGLANS SPECIES ACROSS EUROPE: MONITORING AND CONTROL EFFORTS FOR PROTECTING EU UNCONTAMINATED AREAS

Salvatore Moricca, Alessandra Benigno, Edoardo Falciani, Matteo Bracalini, Tiziana Panzavolta

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Thousand Cankers Disease (TCD) is a disease complex lethal to black walnut (*Juglans nigra*), caused by the fungus *Geosmithia morbida* (GM) and its obligate vector, the Walnut

Twig Beetle (WTB) *Pityophthorus juglandis*. First reported in the western United States about 15 years ago, the disease has now spread to the central and eastern North America, where it is threatening black walnut within its native range. As of 2015, TCD has been reported in Europe, presumably introduced here with black walnut propagation material. In fact, starting from the 90s, several black walnut plantations were established in various European countries with the financial contribution of the EU. The disease, which has now spread epidemically in central-northern Italy, was recently reported (in 2023) in France. Since TCD seriously threatens artificial plantations and amenity trees, both the fungus and its vector are quarantined organisms in Europe. The damage that the exotic TCD could cause in Europe to the equally exotic black walnut is negligible compared to the greater risk that it may massively attack the native English walnut, putting the cultivation of this species and its economy in crisis. This risk is more real than ever because the close interconnectivity has already resulted in host jumps in many mixed plantations. Efforts to try to mitigate the impact of TCD in Europe must operate along 4 lines: 1) enact strict regulations on the import and movement of *Juglans* commodities within the EU territory; 2) develop accurate and sensitive diagnostic tools (conventional/molecular); 3) constant monitoring of the territory for the detection of initial disease foci, by using advanced diagnostic tools and vector-specific traps; and 4) carefully investigate the epidemiology of TCD, to delve deeper into the joint influence of climate and biotic interactions on the reproductive success of both the fungus and the beetle.

SE24 C03

DISEASE MANAGEMENT IN PINUS RADIATA PLANTATIONS IN CHILE

Dr. Rodrigo Ahumada N.

Forestal Arauco, Concepción, Chile

Commercial plantations in Chile are comprised of exotic *Pinus* and *Eucalyptus* species, which make up approximately 2.4 million ha. *Pinus radiata* is the main species with about 1.4 million ha and *Eucalyptus* spp. with close to 860,000 ha. Currently, this is the foundation of the forest industry in the country and the second most important economic activity in Chile after cooper. Both species had been considered as relatively pest and disease free in Chile, despite their susceptibility to several agents, elsewhere in the world. Globalization has given rise to a very substantial increase in trade between countries and continents, greatly increasing the use of wooden containers. This pattern has increased the risk of introducing new pests and pathogens. During the last 30 years almost every year a new insect or disease has been detected in the country and that will continue in the future. Since 1985 when pine shoot moth was detected several new pests and diseases have been detected and have



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caused damage in nursery and plantations. Among the most important species affecting *P. radiata* are; *F. circinatum* present only at nursery level, *D. sapinea* especially important after forest fire damage or periods of severe drought, *P. pinifolia* focused on humid areas, bark beetles damaging the establishment of plantations and export products. To prevent the introduction of new pests and pathogens that may affect forest productivity and cause severe damage to the ecology and economy of the country, quarantine measures to reduce the chance of accidental introductions of pathogens and insects are very important.

SE24 C04

THE PANGLOBAL PLANT PATHOGEN PHYTOPHTHORA CINNAMOMI THREATENS MEDITERRANEAN FOREST ECOSYSTEMS

Bruno Scanu

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The oomycete *Phytophthora cinnamomi* is one the most notorious and invasive pathogen infecting more than 5000 woody plant species worldwide. Native to southeast Asia, it has clonally spread across all continents causing serious economic, environmental, and social impacts in horticultural and forest ecosystems. Over the last decades *P. cinnamomi* has been associated with widespread dieback and mortality of evergreen oak and chestnut trees in the Mediterranean basin, boosted by temperature increases and extreme weather events such as flooding and drought. Scarce public awareness, sensible human impact on forest areas, and the new EU regulation on the use of fungicides and fertilizers further increase the risk for Fagaceae ecosystems. This presentation will provide a risk map of *P. cinnamomi* based on its habitat and distribution model in Mediterranean ecosystems under different climate scenarios as well as evidence of the efficacy of treatments with phosphonates in suppressing pathogen infection in oak and chestnut trees. In addition, the development, validation, implementation, and dissemination of an Integrated Pest Management (IPM) protocol, tailored to these specific target ecosystems, will be presented. This includes the subsequent application of a biofumigant, microbial formulations and biostimulants as well as prevention strategies in declining evergreen oak and ink diseased chestnut stands across three Mediterranean countries.

SE24 C05

EARLY DETECTION, MONITORING, AND ECOLOGICAL MODELING OF FOREST PATHOGENS SUPPORT DEVELOPMENT OF PROACTIVE FOREST HEALTH MANAGEMENT STRATEGIES

Bennett P.I.¹, Hanna J.W.¹, Kim M.-S.², Stewart J.E.³, Klopfenstein N.B.¹, Garbelotto M.⁴

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Forest pathogens cause severe, long-term ecological, economic, and cultural impacts, while also reducing global carbon sequestration in diverse forest ecosystems. Early detection and monitoring of forest pathogens are essential for sustainable forest health management and global biosecurity. Environmental sampling (e.g., aerial spore trapping, soils) for native and invasive forest pathogens coupled with molecular detection/identification enables efficient, long-term monitoring and assessments of forest disease threats. Recent technological developments, such as taxon-specific, real-time PCR and loop-mediated isothermal amplification, allow for rapid, sensitive, and specific molecular detection/identification of forest pathogens. The integration of geospatial data with inoculum abundance data from various spore-trapping efforts supports development of aerial spore dispersal models. Data integration also supports bioclimatic modeling using methods such as maximum entropy to predict potential distributions of suitable habitat for forest pathogens and their hosts under contemporary and projected future climates. Forest stand-management decisions such as shifting species composition, planting of resistant/tolerant species, use of improved seed sources, thinning/harvest practices, and the application of preventative chemical or biological treatments, are also supported by information from these approaches. Furthermore, the utility of these monitoring/modeling approaches has been demonstrated with several forest pathogens that are native to North America and invasive in Europe, including *Nothophaeocryptopus gaeumannii*, which causes Swiss needle cast of Douglas-fir (*Pseudotsuga menziesii*), and *Heterobasidion irregulare*, which causes root disease on a wide variety of pine hosts (*Pinus* spp.). Other case studies include *Armillaria solidipes*, which causes root disease on diverse conifers and hardwoods, and the invasive *Cronartium ribicola*, which causes white pine blister rust on five-needle pines. These tools and techniques provide information to guide forest land managers as they make decisions about how to mitigate the impacts of native and invasive forest pathogens, while enhancing forest health, productivity, sustainability, and climate resilience.



MC3

18.00-20.30 Concurrent Session 25

Emerging Pests with Relation Plant Biosecurity and Food Safety

Chair: Abraham Gamliel (*Aro Volcani Institute, Israel*)

James P. Stack (*Kansas State University, USA*)

SE25 C01

PLANT HEALTH IN THE EVOLVING ONE HEALTH CONCEPT

James P. Stack

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The age-old understanding of the close connections among human health, animal health, and the environment has received renewed interest over the past few decades resulting in the establishment of national and global organizations to promote its merits. Numerous conferences have convened to address the emergence from animal reservoirs of pathogens causing serious human diseases. Absent from these organizations and conferences, has been the inclusion of plant health as an important component of One Health. A fair question might be, is there sufficient justification for a more inclusive One Health concept that includes plant health? The short answer is yes. There are multiple examples of, 1) plant pathogens causing diseases in humans and animals, 2) plants as reservoirs of human pathogens, and 3) plants and plant products that serve as vectors of human and animal pathogens. Due to the increasing prevalence and severity of human fungal disease coupled with the increasing incidence of fungicide resistance in human fungal pathogens, the World Health Organization established a fungal priorities list in 2022. Fungicide resistance in human pathogenic fungi has been directly linked to plant-based agriculture. Fungicide-resistant *Fusarium* infections in humans are rising dramatically and include several species of plant pathogenic fungi – true trans-kingdom pathogens. When inoculated onto plants, *Fusarium* isolates from human infections caused disease in those plants. Grains were shown to vector animal viruses including, African Swine Fever in transoceanic simulations. In addition, increasing environmental stressors, including climate change, land use change and population growth, will challenge our ability to provide the nutrition necessary to keep that growing population healthy. The newly expanded definition of One Health in the U.S. states “...human, animal, plant, and the environment...”. The more prominent feature of plant health in the One Health Concept is long overdue; the rationale and justification for that statement will be discussed.

SE25 C02

RISK ASSESSMENT, PEST ALERTS AND EMERGENCE: WHAT DETERMINES THE TIMELINES?

Michael Jeger

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Plant health regulation requires that risk assessments are made based on scientific evidence to enable risk management decisions. Evidence arises from two main sources:

authenticated new pest reports, findings or alerts made by national, regional, and international bodies; and accounts of emerging pest outbreaks and disease epidemics published in the peer-reviewed scientific literature. The information obtained from the scientific literature is stronger when based on quantitative pest surveys, surveillance, or other monitoring methods. A scientometric study of new reports in the CABI Distribution Data Base and scientific articles published in 2021, restricted to plant pathogens, showed little overlap in the results, although *Candidatus Liberibacter asiaticus* (and vectors), citrus tristeza virus, and *Fusarium oxysporum* f.sp. cubense were well represented in both sources of information. A similar outcome was found in a similar study made for 2022 publications, but with a different set of overlapping pathogens. The reasons for this lack of overlap are that new pest reports will usually precede major pest outbreaks which may in turn lead to significant crop losses. The question arises what the relationship between these two sources of information is and what is the timeline from a new pest report to a major outbreak? To answer this question, we used information on the years EU Emergency Measures were put in place since 2000 (as a proxy for a major outbreak occurring) and compared this with data on new reports from the EPPO Reporting Service. Examples will be given of where the timeline was long (*Diabrotica virgifera virgifera*), short (*Phytophthora ramorum*, *Xylella fastidiosa*, tomato brown fruit rugose virus), or where emergency measures preceded the new report (*Spodoptera frugiperda*). Similar comparison will be made for other continents and conclusions drawn on the time needed to determine trends in the emergence of plant pests and the effectiveness of regulation.

SE25 C03

A RISK MANAGEMENT FRAMEWORK FOR PLANT BIOSECURITY

Abraham Gamliel

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Plant and food biosecurity targets the agricultural industry and the food supply chain by accidental or deliberate introduction of a plant pathogen or insect pest, hence, damaging crops, food and feed. It is therefore a challenge for the stability of a society, and its economy, since, agriculture and the food industry are a primary sector of any nation's economy. There are many windows for threats from the introduction of pest or disease agents. Therefore, a risk management program, an interdisciplinary set of actions before and following the introduction of harmful organisms, should be carefully prepared and effectively executed. The risk management framework comprises a sequence of activities with structured

responsibilities among regulatory authorities, scientists, extension professionals and farmers. Risk management includes the activities for pest risk assessment, preparedness, detection, diagnosis, procedures of containment eradication and management, and finally a recovery plan. The risk management steps should be coordinated within a country and at a regional level in order to ensure that a disease or insect pest outbreak is successfully managed and does not establish in the area or spread to neighboring countries. A risk management framework is the key for rapid and effective counteracting measures, aiming at nullifying the impact of an outbreak from a nonnative pathogen or pest. The potential impact of such a pathogen or pest may be devastating, especially if targets are the major staple food crop in a country. Risk management is also relevant for an accidental or deliberately introduced pathogen since the significance of both is in many cases similar. The activities which comprise the risk management framework should, above all, be structured and coordinated with responsibilities and hierarchical authorities. The margins for error in the process are narrow, since the time and space to correct failure in the management of an outbreak are minimal. While risk management in the event of an intentionally caused outbreak is very similar to unintentional outbreaks, additional preventive measures directed at the motivation of perpetrators and their prosecution should be taken.

SE25 C04

DEVELOPING AN EFFECTIVE BIOSECURITY SYSTEM TO ADDRESS INTRODUCTION OF EMERGING PESTS

Simon McKirdy

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Global trade and travel have dramatically increased the risks associated with the introduction of plant pests and pathogens into new geographic areas in short periods of time. An effective biosecurity system needs to include effective policies and measures for prevention, surveillance, mitigation, and recovery. Ideally, those policies and measures are integrated across sectors within countries and coordinated with primary trading partners, as well as, with second order trading countries. Among the many challenges to plant biosecurity, are the disruptions to trade networks and supply chains caused by regional conflicts that cause sudden network realignments to avert food shortages and minimize economic impacts. These realignments often result in new network pathways that are established without adequate risk and pathway analyses to support prevention measures, thus, increasing the likelihood of the introduction of plant pests and/or pathogens into new areas. Current trade and biosecurity policies are outdated relative to the magnitude of trade and the tremendous advances in detection and diagnostics technologies. To protect plant health and safeguard trade, we need a more comprehensive biosecurity strategy that places equal emphasis on prevention and surveillance. Building an effective biosecurity system to address the introduction of emerging pests and pathogens is a bottom-up process requiring a solid foundation in biosecurity principles.

SE25 C05

A PLANT DISEASE COMPLEX BETWEEN A PLANT PARASITIC NEMATODE, FREE LIVING NEMATODES AND A FUNGUS - REEVALUATING PRATYLENCHUS CAPSICI DISEASE ETIOLOGY

Sigal Brown Miyara¹, Abraham Gamliel², Patricia Bucki¹, Marina Benichis² and Xue Qing³

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The new nematode species *Pratylenchus capsici* which was recently identified in Israel is continuing being the major cause of a devastating damage observed on pepper crop resulting in stunted growth and significant yield reduction. In efforts to develop efficient nematode integrated management, *Pratylenchus capsici* disease etiology was reevaluated through straightforward experimental strategies. Molecular phylogeography analysis suggests that contemporary gene flow is prevented among different agricultural farms, while population dispersal from weeds to pepper occurs on a relatively small scale and might support nematode survival in between seasons. Host range studies illustrate *P. capsici* demonstrate a polyphagous nature as indicated by highly reproduction on additional vegetables as eggplants, tomatoes and cucumbers. Metabarcoding analysis of soil microbial community using ITS and 16S marker gene for fungal and bacterial identification respectively, of *P. capsici* infested roots, indicated that *Olpidium* species are widely presented in *Pratylenchus* introduced root-lesion, and might be a faithful companion associated with roots infected by *P. capsici*. Moreover, pepper roots staining revealed a dense appearance of *Olpidium* life phases as multinucleate thallus, zoosporangium, resting spores and newly encystment of zoospores on pepper roots as well as free living nematodes attracted into lesion region. Bayesian phylogeny analyses placed the recovered *Olpidium* in two unresolved shallow clades, more closed to *Olpidium virulentus* rather than the monophyletic clade of *Olpidium brassicae* and *Olpidium bornovanus*. Therefore, we assign our recovered species as *O. virulentus*. In order to understand better the nature of *Pratylenchus*-*Olpidium* co-occurrence, application of fungicidal and nematicidal treatments to infected plants was carried out. Results indicates *P. capsici* penetration doesn't effect on *Olpidium* occurrence although *Olpidium* appear to support better *Pratylenchus* disease occurrence on Pepper roots. Over our results, facilitate with developing innovative management strategies through tailoring them within the agricultural practices according to *P. capsici* etiology and characteristics



SE25 C06

EMERGING STRAINS AND THEIR MULTI-TROPHIC INTERACTIONS THREATENING FOOD SAFETY AND BIOSECURITY

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The emergence of new plant bacterial pathogens represents a threat to public health and the stability of food supply chains, as these pathogens exhibit aggressive characteristics with the potential for substantial economic losses. In our lab, we are studying the emergence of these plant bacterial pathogens and their interactions with foodborne human pathogens. In a recent study, we identified a highly pathogenic strain of soft rot-causing bacteria, *Dickeya fangzhongdai*, affecting a crucial staple crop, potato. Genome analyses revealed significant heterogeneity in pathogenicity determinants compared to the type strain, with ANI and dDDH values of 97.1% and 73.3%, respectively. Noteworthy differences in motility were observed, characterized by pronounced aggressiveness that resulted in the maceration of entire taro corms. This was confirmed by transmission electron microscopy (TEM), which revealed peritrichous flagella, in contrast to the type strain DSM10197T that exhibited no flagella. In another aspect of our research, exploring interactions between foodborne pathogens (*Salmonella enterica* and Shiga toxin-producing *Escherichia coli*) and soft rot bacteria unveiled a dual role. Soft rot bacteria not only damage crops but also provide a niche for foodborne pathogens to proliferate inside hosts. We also discovered a *Pectobacterium* strain harboring a complete plasmid of a pathogenic *Salmonella enterica*. This plasmid exhibited an almost 100% ANI with other plasmids of *Salmonella enterica* and carried antibiotic-resistant genes (ARG). These studies underscore the escalating challenges associated with emerging bacterial pathogens and their profound implications for food safety and biosecurity. It is imperative that the global community remains vigilant and proactive in addressing these emerging threats.

SE25 001

THE INDISPENSABLE ROLE OF SCIENCE IN THE CANADIAN FOOD INSPECTION AGENCY'S PLANT PROTECTION PROGRAM

Nanang D

Science Branch, Canadian Food Inspection Agency, Canada

Healthy plants are integral to food security, biodiversity and economic prosperity. The Canadian Food Inspection Agency (CFIA) is Canada's National Plant Protection Organization under the International Plant Protection Convention. Accordingly, the CFIA is responsible for administering a plant protection program that aims to safeguard Canada's plant resource base against the entry, establishment and spread of economically and environmentally significant plant pests while facilitating safe trade. Science plays an indispensable role in the successful execution of this program. Risk assessment, risk intelligence, data analytics, research, surveillance and

diagnostic testing – all form the cornerstones of the CFIA's plant protection program, which provide a strong foundation to support the CFIA's policies, actions and decisions. This presentation will illustrate how the CFIA employs innovative technologies and methodologies to enhance early detection, rapid response and effective management of plant health risks. Furthermore, the presentation will underscore how the CFIA leverages partnerships and collaborations to further its mission to mitigate potential threats in a rapidly changing environment.

SE25 002

POPULATION DYNAMICS OF THE PATHOGEN CAUSING LATE BLIGHT OF POTATO AND TOMATO IN CANADA: HISTORICAL PERSPECTIVES AND CURRENT TRENDS

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Late blight of potato and tomato, caused by *Phytophthora infestans*, is an important disease of solanaceous crops worldwide. In Canada, several pathogen population displacement events have occurred in recent decades. In the mid 1990s, the A1 mating type, metalaxyl-sensitive (MS) US-1 genotype was largely displaced by the A2 mating type, metalaxyl-insensitive (MI) US-8 genotype in much of the country. An exception would be the west coast of Canada, where the A1, MI US-11 genotype became predominant. The original US-1 population was completely displaced in 2-3 years. A second displacement event occurred in 2009, when genotypes US-22 (A2, MS), US-23 (A1, MS) and US-24 (A1, MS) replaced US-8 as the predominant genotype in most potato-producing regions, and also caused severe disease in tomato crops. Novel pathogen genotypes have been transported great distances via potato seed and tomato transplants to become established in new crop production regions. Pathogen spread of shorter distances also occurs via dissemination of sporangia from infected crops to adjacent fields in rain and storm events. In recent years, the US-23 genotype has predominated in most potato- and tomato-producing regions of Canada. An exception would be the west coast of the country where a greater diversity in pathogen genotypes is found in commercial crops, home and community gardens, implying that sexual recombination generating novel genotypes is occurring. The monitoring of pathogen populations of *P. infestans* must continue to alert the industry to new threats and provide up to date management options for late blight. Education of home and community gardeners and the promotion of late blight-resistant tomato varieties have become important tools in the management of late blight in commercial crops.



Concurrent Sessions

Wednesday, 3 July

SE25 003

ADDRESSING BIOHAZARDS OF PRIMARY PRODUCTION IN AGRICULTURE

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Pathogens constitute biohazards for the primary production of crops and livestock, where they can cause major economic damage to farmers, the agricultural industry, society, and international trade. Introductions of pathogens mostly have natural causes, but they may be also be accidental or intentional. Agroterrorism is the intentional introduction of plant or animal pathogens into agricultural production systems with the intention to cause socioeconomic harm and generate public fear. The risk for acts of agroterrorism is low, but not negligible. New concerns about threats arise from the rapid advancements in biotechnology and emerging technologies. In this study, FORSA, an analytical framework for risk and vulnerability analysis, was used to review how to prepare for and mitigate the possible effects of natural, accidental or intentional biohazards in agricultural production. The overall goal with such an analysis is to understand the nature of the risks, strengthen crisis preparedness, reduce vulnerabilities and increase the ability to manage crises. Analysing the effects of a biohazard event involves multiple scientific disciplines. A comprehensive analysis of biohazards therefore requires a systems approach to cover all complexities. From the analysis, we can conclude that the preparedness and ability to manage events are strengthened by bolstered farm biosecurity, increased monitoring and laboratory capacity, improved inter-agency communication and resource allocation. With climate change, Covid-19 and the war in Ukraine, the supply chains are challenged, and we foresee increasing food prices associated with social tensions. Our food supply chain becomes more fragile with more unknowns, thereby increasing the needs for risk and vulnerability analyses, of which FORSA is one example.

pathogenic bacteria were not enriched in damaged pods. Two herbivory-induced flavonoids, viz, 7,4'-dihydroxyflavone and coumestrol, were found to exert insecticidal activity. Our study clarified that BHBT symptoms are directly caused by the thrip. Economic thresholds for pest management need to be reconsidered and eco-economic thresholds should be proposed as thrip herbivory did not degrade cowpea nutrition.

SE25 005

STATUS/SHIFTING OF POD BORER, *HELICOVERPA ARMIGERA* POPULATION TREND IN MAJOR LEGUMES AND OTHER CROPS IN INDIA

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Pod borer, *Helicoverpa armigera* (Lepidoptera: Noctuidae) the single largest yield shrinking factor in food legumes – causes an estimated loss of US\$ 317 million in pigeon pea and \$328 million in chickpeas. In general, the estimates of yield losses vary from 50 to 100% in the tropics and 5–10% in the temperate regions. In present studies, monitoring of pod borer population using pheromones traps data of AICRP's for both crops results revealed that activity of male moths trapping was monitored for eight consecutive (2014 to 2021) years by installing 2-3 traps /acre of both crops field at different hot spot locations of India. The pooled peak population of male moths of *H. armigera* trap catches of different locations were found to be highest during the year 2015-16 (1051.16 trap catches), followed by three fourth population decrease trend was noticed during 2016-17 (306.65 trap catches) and lowest adults trap catches were noticed during 2021-22 (219.68 trap catches) in chickpea fields across India (Fig 1). A similar diminishing trend of *H. armigera* trap catches was also found in pigeonpea crop for 2014(318 trap catches) to 2021-22 (57.65 trap catches) across the different locations of India (Fig 2). In our studies also observed that, the pod borer trend gradually increased in pearl millet crop in some locations of India (Fig 3) and similarly this trend also may happen on other alternate host crops. Keeping in view present studies have been carried out to see how the population trend of pod borer declined across different hot spot locations for both crops from 2014-2021 period and these studies further help on future predication model also how the trend of this pest also effect on other hosts of horticultural and other crops.

SE25 004

BEAN BLOSSOM THRIP *MEGALUROTHRIPS USITATUS* DIRECTLY CAUSES THE BLACK-HEADS & -TAIL SYMPTOMS OF COWPEA ALONG WITH PRODUCING INSECT-RESISTANCE FLAVONOIDS

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1. Zhejiang University, Hangzhou, China, 2. Hainan Research Institute of Zhejiang University, Sanya, China

The bean blossom thrip, *Megalurothrips usitatus* (Insecta: Thysanoptera: Thripidae) damages the flowers and pods of the cowpea, causing "black-heads and black-tails" (BHBT) symptoms and negatively affecting its economic value. The mechanism by which BHBT symptoms develop was still unknown. Our results showed that the microstructure of the pod epidermis was altered and the content of the plant's resistance-related compounds increased after a thrip infestation. The contents of protein and free amino acids did not change significantly, suggesting that the nutritional value was not altered. Pathogens were found not to be involved in the formation of BHBT symptoms, as fungi and

Result & Discussion:

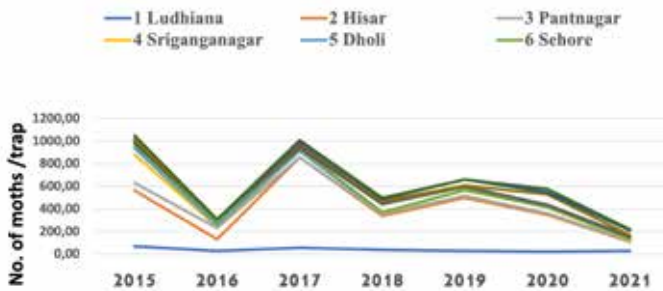


Fig 1: Status of Pod borer, H.armigera population trend different locations in chickpea crop in India

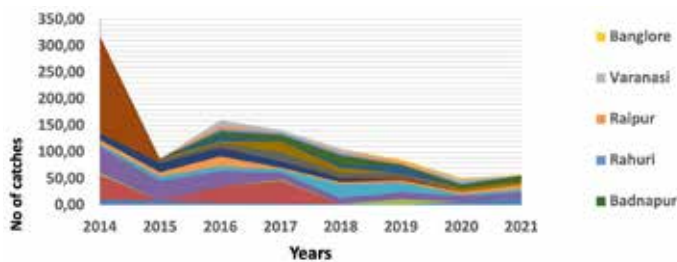


Fig 2: Status of Pod borer, H.armigera population trend different locations in Pigeonpea crop in India

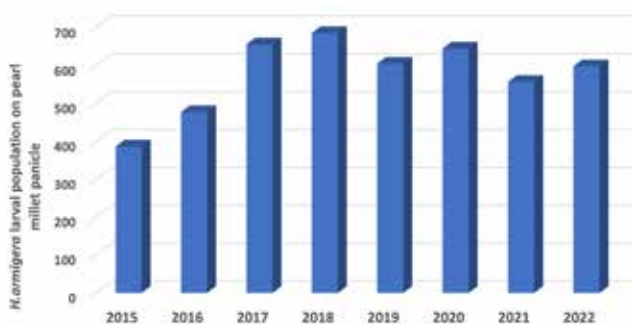


Fig 3: Field surveillance of pod borer population across seasons on pearl millet @ICRISAT, Patancheru, India

Conclusion and Way Forward Research for pod borer:

- We should try to determine not only if, when and how much migration occurs, but why it occurs
- There is a need to study migration patterns and local fluctuations in *Helicoverpa* abundance as several crops serve as collateral and alternate hosts for these pests.
- Understand host plant—insect—environment interactions in different regions to be able to develop appropriate strategies for integrated management of these pests
- There is also a need to understand the key biotic/abiotic mortality factors at different locations in different crops, at different life stages and the mortality regulated by the host plant
- Need for information on co-evolutionary trends, sustainability of the ecosystem and homeostasis in *Heliothis/Helicoverpa* populations so as to be able to develop integrated biotic stress management as a means of minimizing the extent of losses due to these pests.



CONFERENCE I

18.00-20.30 Concurrent Session **26**

Environmental Fate, Ecotoxicology, Risk Assessment, and Remediation of Pesticide Residues

Chair: Zisis Vryzas (School of Agriculture, Aristotle University of Thessaloniki, Greece)

SE26 C01

NOVEL APPROACHES IN REMEDIATION OF PESTICIDE RESIDUES BY BIOLOGICAL SYSTEMS

Paraskevas Parlakidis¹, Georgios D. Gikas², George Adamidis¹, and Zisis Vryzas³

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Pesticides can enter natural waters through agricultural wastewater from washing or loading of spraying equipment, leaks occurring due to improper maintenance of empty containers (point source pollution), and diffusion through run-off, spraying drift, leaching, and subsurface drainage. Fungicides include a broad range of compounds with different modes of action belonging to various chemical classes. Acute and/or chronic toxicity of organic fungicides to aquatic or terrestrial organisms have been reported in many studies. The extensive use of chemical pesticides in agriculture also raises concerns for public health. To reduce, eliminate, isolate or stabilize a pesticide, remediation technologies use physical, chemical, or biological processes. The selection of appropriate technologies depends on several factors, such as site characteristics and contamination (punctual or diffuse), concentration and type of pesticides to be removed, and the end use of the contaminated media. Degradation or decomposition of pesticide residues by plants is called phytoremediation. This mechanism is highly associated with the breakdown of residues either in the plant body or in the rhizosphere. It is noteworthy that phytoremediation is also an innovative, ecologically beneficial, and cost-effective technique for the degradation of pesticide residues. A phyto-based remediation technique is Constructed Wetlands (CWs). CWs comprise an environmentally friendly, low cost, and efficient fungicide remediation technique. Pesticide removal within CWs is dependent on plant uptake and metabolism, absorption in porous media and soil, hydrolysis, photodegradation, and biodegradation. Factors related to the efficacy of CWs on the removal of pesticides, such as the type of CW, plant species, and the physicochemical parameters of fungicides, are also discussed. Results from case studies will be presented. Furthermore, prospects and challenges for future research are suggested under the prism of reducing the risk related to pesticides and enhancing CW performance.

SE26 C02

GREEN APPROACHES FOR PESTICIDES CONTROL IN AGROINDUSTRIAL EFFLUENTS

Carmen Sans, Núria López-Vinent, Alberto Cruz-Alcalde, Pere Llopart, Júlia Nieto-Sandoval, Pilar Marco, Bernardí Bayarri

Chemical Engineering Section, Departament of Chemical Engineering and Analytical Chemistry, University of Barcelona, Spain

Agroindustrial facilities play a significant role in introducing pesticides and other micropollutants into aquatic environments. Consequently, adopting effective treatment methods to remove these micropollutants from wastewater effluents is crucial for environmental protection. Processes like adsorption and Advanced Oxidation Processes (AOPs) have demonstrated their efficacy in eliminating a wide range of micropollutants. However, it's important to recognize that many of these technologies come with high energy demands and associated costs for implementation and maintenance. On the other hand, Nature-based Solutions (NbS), such as constructed wetlands (CW), offer an eco-friendly and cost-effective approach to wastewater treatment. CWs have the potential to eliminate micropollutants while maintaining simplicity in operation and maintenance. However, relying solely on NbS for effluent treatment may not meet the minimum requirements for water reuse, especially regarding the removal of micropollutants or bacterial inactivation. In this context, seeking greener approaches to ensure safe, high-quality water is essential for both the environment and human health. One promising strategy involves hybrid CW-AOP systems. By combining CWs with AOPs, we can reduce land footprint, decrease energy and maintenance costs, and enhance water reuse through improved bacterial inactivation and micropollutant removal. Strategies like solar photo-Fenton using organic fertilizers as iron source, when integrated with CWs, provide a practical and adaptable solution that meets regulatory limits, tailored to agriculture destination use. Such an approach excels in cost-effectiveness, environmental sustainability, and overall efficiency. Furthermore, the exploration of effective adsorbents represents a novel strategy for waste and water valorisation. Lignocellulose wastes, rich in functional groups capable of binding to water-based pollutants, offer a valuable resource for synthesizing bio-adsorbents. These bioadsorbents include products obtained under oxygen-free conditions (biochars) and in the presence of subcritical water (hydrochars). To enhance their efficacy in removing micropollutants from complex aqueous matrices, biochar and hydrochar can benefit from tailored engineering modifications to their physicochemical properties. In addition, biochar and hydrochar hold promise as supportive, adsorptive, and catalytic materials in Advanced Oxidation Processes (AOPs). However, achieving their suitability as support



materials requires specific conditioning treatments. Acid/base treatments, chemical oxidation, surfactant incorporation, and mineral coating and impregnation are essential steps. These treatments boost up H_2O_2 , O_3 , peroxymonosulphate and persulphate activation to enhance the generation of reactive oxygen species (ROs), including $\bullet OH$, $SO_4^{\bullet -}$, HO_2 , and $O_2^{\bullet -}$, which play a crucial role in targeted pollutant degradation. The Advanced Oxidation Process Engineering group at the University of Barcelona has recently focused on developing and optimizing these environmentally friendly approaches. Their applications extend broadly to water treatment and water reuse.

SE26 C03**STRATEGIES TO REDUCE FUNGICIDE RESIDUES AND MYCOTOXINS IN WINTER CEREALS**

Palladino C.¹, Pérez C.², Pereyra S.³, Pareja L.⁴

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Fungal diseases pose a dual threat to agriculture by compromising crop yields and affecting grain quality, resulting in significant economic losses and jeopardizing global food security. The primary method to address these issues involves widespread fungicide application, leading to the emergence of resistance in fungal populations. Stringent regulations aiming consumer's health, further complicate the situation, requiring monitoring and control measures in agriculture. Uruguay faces challenges due to limited data on fungicide and mycotoxin levels in winter cereal grains, hindering informed decision-making, compromising food safety assessments, and potentially impeding compliance with international agricultural standards. This work aimed to explore effective disease control strategies that reduce dependency on chemical interventions while ensuring robust disease management and upholding stringent food safety standards. Since 2015/16 growing season, comprehensive field experiments have been conducted on both wheat and barley. The trials included the evaluation of different fungicide strategies where mixtures of fungicides applied at different times and with different application technologies in cultivars susceptible or moderately susceptible to the disease under study were evaluated. Disease incidence, severity, control efficiency, and grain yield for crucial diseases: Ramularia leaf spot (barley), Stripe rust (wheat), and Fusarium Head Blight (FHB) in both cereals were assessed. Moreover fungicide residues and micotoxinas were determined. Management strategies were identified with the lowest number of fungicidal and efficient applications in the control of major diseases in winter cereals. In addition, these strategies comply with international standards of maximum residue limits for fungicides and mycotoxin content in grains. These findings present a contribution to the management of fungal diseases in Uruguay. This information is crucial for implementing good agricultural practices within these specific disease systems.

SE26 C04**TRACKING HERBICIDE FATE AND BEHAVIOR - A CRUCIAL PUZZLE PIECE FOR OPTIMAL HORTICULTURE PRODUCTION IN SANDY SOILS**

Ramdas Kanissery

University of Florida, Department of Horticultural Sciences, Southwest Florida Research & Education Center

Herbicides, used for weed control in crop production, can enter the soil through direct spray, non-target movement, or release from dead vegetation. The retention and distribution of herbicides in the soil are influenced by soil characteristics, herbicide properties, and environmental conditions. Undesirable movement due to soil and weather conditions can lead to rapid leaching of herbicides, resulting in inadequate weed suppression or unintended crop effects. Progress in tracking herbicide fate, utilizing technologies such as rapid analytical assays and modeling, has enhanced our understanding of herbicide behavior in cropping systems. Despite the widespread use of herbicides in citrus orchards in Florida, there is limited information on how they behave and their environmental impact in the sandy soils where citrus is primarily grown. Through multi-year studies, we assessed the fate and behavior of popular herbicides (glyphosate, diuron, and indaziflam) in Florida's citrus production soils. Glyphosate had a mean half-life of 26 days, diuron had a mean half-life of 34 days, and indaziflam had a half-life of 85 days in tested sandy soils. Observations from this study suggested that herbicides dissipate relatively faster from Florida's sandy soils compared to reported soil half-life values from around the globe. However, the fate parameters depend significantly on slight nuances in the soil's physicochemical properties, even within sandy soils. This study, conducted in southwest Florida citrus production, offers insights into the unique behavior of herbicides in Florida's sandy soils and how this information can be practically applied for tree-safe weed control in horticultural production systems.

SE26 C05**CASE OF STUDY: FUNGICIDE RESIDUES IN BARLEY GRAINS AND THEIR RELATIONSHIP WITH PHYSICO-CHEMICAL PROPERTIES AND WEATHER CONDITIONS**

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Fungal disease control strategies may result in pesticide residues in grain, even when applied according to good agricultural practices. This study aimed to evaluate the residues of fungicide mixtures used to control Ramularia leaf spot on barley grains and their relationship with physicochemical properties and weather conditions. Four different alternative fungicide mixtures: fluxapyroxad + pyraclostrobin + epoxiconazole, pyraclostrobin + epoxiconazole + chlorothalonil, prothioconazole + trifloxystrobin, and isopyrazam + azoxystrobin, applied at



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GS33+GS47, were evaluated in five field trials in different environments. A triple application was also included. Barley grains were harvested from all trials and fungicide residues were determined. Fungicide residues concentrations were compared with maximum residue levels (MRLs) set by international organizations. The frequency of detection of fungicides in grains was different in each environment studied. There was a negative relationship between accumulated rainfall and rainy days in each environment and the percentage of residues detected. All fungicides with a log octanol-water partition coefficient > 2 were detected in grains, except chlorothalonil, which was not detected in this study. Chlorothalonil is a contact fungicide that may remain on the plant surface and thus undergo several biochemical degradation processes. Prothiconazole, which is not persistent in soil and is the most soluble of the fungicides evaluated, is the other active ingredient that was not detected in this study. On the other hand, fungicides with higher dissipation rate lifetime (RL50) were detected more frequently in grains than those with lower RL50. The fungicide doses and application timing used in this study resulted in residue concentrations below MRLs. Nevertheless, and given the complexity of the interactions between the presence of fungicide residues in grains, environmental conditions, and the physicochemical properties of the active ingredients, it highlights the importance of local studies to establish pre-harvest intervals at national level.



BANQUET

08.30-11.00 Concurrent Session 27A

Biological Control of Insect Crop Pests

Chair: Perdikis Dionisis (Agricultural University of Athens, Greece)

Michele Ricupero (Agriculture, Food and Environmental Science, University of Catania, Italy)

SE27 C01

BIOLOGICAL CONTROL OF CROP INSECTS IN THE MEDITERRANEAN AREA: APPLICATIONS AND PERSPECTIVES.

Luciana Tavella¹

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In the Mediterranean area, climatic and environmental conditions support a rich insect biodiversity, which provides ecosystem services, including pest biological control. Several natural enemies have been found in activity against crop pests, as a result of reduced pesticide pressure and/or accidental introduction of invasive exotic pests. The examples are numerous, starting from generalist predators and parasitoids to some more specialized parasitoids that have created successful new associations. To begin with the generalist predators, emblematic was the discovery in the mid-1980s of predatory bugs belonging to Miridae Dicyphini (e.g., *Macrolophus pygmaeus*, *Nesidiocoris tenuis*) and Anthocoridae (e.g., *Orius laevigatus*) that, with the larger-scale adoption of integrated pest management, have colonised protected crops, proving capable of preying on a variety of pests, such that they are successfully released for pest control in vegetable and ornamental crops. Recently, generalist parasitoids, such as the wasps *Anastatus bifasciatus* and *Trichopria drosophilae*, have also been shown to adapt to newly introduced pests, i.e., *Halyomorpha halys* and *Drosophila suzukii*, respectively, and therefore are now commercially available to implement augmentative biological control. Also noteworthy are the new associations created between indigenous parasitoids, particularly belonging to Eulophidae, and exotic pests, starting with *Diglyphus isaea*, which proved to be an efficient parasitoid of the exotic *Liriomyza* spp., up to *Necremnus tuta*, an indigenous species described only following the introduction of the exotic *Tuta absoluta*, the primary hosts of which are still poorly known. These biological control agents are a valuable resource for pest control in the Mediterranean area as well as in many areas worldwide. Therefore, knowledge of their biology and behaviour will enable their increasing conservation and exploitation to achieve effective and sustainable control of crop pests

SE27 C02

ENHANCING BIOLOGICAL CONTROL OF INVASIVE STINK BUG, HALYOMORPHA HALYS BY TWO EXOTIC EGG PARASITIDS

A. Pozzebon¹

1. University Of Padua – DAFNAE, Italy

The invasive alien species *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae) is a primary pest of fruit crops in the Northeast of Italy. Adventive populations of Asian egg parasitoids, *Trissolcus mitsukurii* Ashmead and *Trissolcus japonicus* Ashmead (Hymenoptera: Scelionidae), have been observed in this area since 2016. Moreover, a biological control program based on the large-scale release of *T. japonicus* was implemented starting in 2020. Investigations were performed to understand the main factors influencing the biological control of this invasive pest in fruit orchards. The parasitism impact was positively influenced by the presence of semi-natural habitats constituted by large patches of unmanaged vegetation, but a positive effect of these ecological structures was also observed on the *H. halys* population. Habitat management based on ground cover management can also enhance the activity of egg parasitoids, but attention should be placed on the selection of companion plants to avoid problems related to the unwanted increase of stink bugs. The implications of these factors on IPM in fruit crops are discussed.

SE27 C03

INSECTICIDE EXPOSURE AFFECTS FORAGING BEHAVIOUR OF THE EGG PARASITOID TRISSOLCUS JAPONICUS

Rondoni G.^{1,2}, Chierici E.¹, Brodeur J.², Conti E.¹

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While foraging in agricultural habitats, parasitoids exploit cues related to the host species to efficiently locate the suitable stage for oviposition. The innate response of the parasitoid towards host-associated cues can change during the foraging activity, for example, in response to repeated encounters with the host and successful oviposition events, eventually resulting in improved reproductive success. At the same time, in their search for hosts parasitoids may come into contact with insecticides which, if not lethal, can alter host location behaviour, including locomotor activity, arrestment response, and learning ability. The egg parasitoid *Trissolcus japonicus* (Hymenoptera: Scelionidae) is a classical biological control agent of the invasive brown marmorated stink bug, *Halyomorpha halys* (Hemiptera: Pentatomidae), in Italy. The parasitoid has also the capacity to develop in some native stink



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bug species co-occurring with *H. halys*, such as the predator *Arma custos*. We hypothesised that a low concentration (causing 20% parasitoid mortality) of a commonly used neonicotinoid (acetamiprid) would alter the behaviour and learning capacity of *T. japonicus* to exploit the chemical traces left by *H. halys* or *A. custos* reproductive females. Residence time, that is, the time spent by the parasitoid inside an arena contaminated with stink bug traces, was recorded. *Trissolcus japonicus* females responded positively to traces left by both stink bug species. Following oviposition experience and encounter with host traces (associative learning), *T. japonicus* displayed reduced foraging time. Parasitoids previously treated with neonicotinoid showed impaired foraging behaviour (increased residence time in the arena and altered arrest response). Our results suggest that exposure to low concentrations of a neonicotinoid commonly used against *H. halys* may affect parasitoid behaviour, with possible reproductive consequences.

SE27 C04

LEARNING IN PARASITOID WASPS: WHAT IMPLICATIONS FOR BIOLOGICAL CONTROL?

Gioulia Giunti

Learning is defined as the acquisition and retention of neuronal representations of new information and is ubiquitous among insects. Various species rely extensively on learning to guide essential life activities including feeding, predator avoidance, aggression, social interactions, and sexual behavior. Among these, hymenopteran parasitoids are key agents to control economically important pests in agriculture, forestry, and natural ecosystems. Associative learning can occur in parasitic wasps both at the immature stages, as well as at the adult age, usually to adapt to complex environment. Parasitoid wasps rely on a variety of stimuli including visual, olfactory, tactile, and vibrational cues as they forage for hosts, food and mates. In this context, learning can play a key role optimizing the reproductive performances and fitness of female parasitoids by improving their foraging decisions, although sometimes it can prove detrimental outcomes. Associative learning in male parasitoids is much more neglected, although their learning ability can improve searching behavior and mating performances when competing for mates. Associative learning of immature, early adult and adult parasitoids can involve different kind of stimuli, including long- and short-range chemical cues (i.e., plant, host, food and exogenous cues) and visual cues (i.e., colors and shapes) and different rewards. Visual cues can be valuable at both short and long distances when associated with food or oviposition opportunities, while chemicals are key factors during host-searching. Learning ability in parasitoids may vary according to their host specialization, physiological and genetic features. Parasitoid learning could be exploited to improve the success of biological control programs, by selecting and priming candidate parasitoids for biocontrol of a particular pest during mass-rearing. Pre-release training may improve the responses of both male and female parasitoids. However, the potential impact of associative learning on the establishment and efficacy of parasitoids post-release should be deeper investigated.

SE27 C05

PARASITOIDS OF MAJOR PESTS IN FIELD CONDITIONS: DO WE REALLY KNOW WHAT THEY FEED ON?

Ainara Peñalver-Cruz¹, Martin Luquet², Louise Lérault³, Pablo Urbaneja-Bernat⁴, Dolors Bosch¹, Bruno Jaloux³, Judit Arnó⁴

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Natural enemies controlling insect pests such as parasitoid wasps are sensitive to the availability of resources in the environment due to their short lifespan and low energy and storage capacity. Agroecosystems are often described as highly structured environments with abundant hosts within the plots, while limiting complementary resources (e.g., nectar, pollen, honeydew, plant guttation) and additional resources (e.g., alternative insect hosts) are often restricted to uncultivated areas around the crops. The nectar provision hypothesis states that the availability of nectar-producing plants in agroecosystems benefit parasitoid populations, improving their efficiency in controlling pest populations. Therefore, as part of the agroecological transition, numerous actions aimed at increasing floral diversity of cultivated plots and their surroundings, are currently recommended to provide resources for natural enemies and to promote biological regulations. However, the actual exploitation of these resources by natural enemies has rarely been evaluated and quantified, leading to rather random results. For this reason, our research presents an innovative approach that infer the recent feeding patterns of parasitoids in field conditions based on their whole-body carbohydrate profile. This involves training a classification algorithm using laboratory-reared parasitoids exposed to a controlled diet and subsequently compared to field collected parasitoid carbohydrate profiles. The latter, classifies the individuals collected from the field into the food classes originally defined in the laboratory. As a case study, the feeding patterns of *Aphidius ervi*, a generalist parasitoid of aphids in cereals and legumes, and *Aphelinus mali*, a specialist parasitoid of the woolly apple aphid (*Eriosoma lanigerum*) are illustrated and discussed.



SE27 C06

BIOLOGICAL CONTROL AND BIOTECHNOLOGICAL APPROACHES FOR CONTROLLING INSECT PESTS: A FOCUS ON OLIVE TREE PROTECTION

Paula Baptista, José Alberto Pereira

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In recent years, significant attention has been directed towards reducing chemical insect pest control, driven by the acknowledged detrimental impact of insecticides on both the environment and human health. In this regard, biotechnology has been playing a significant role in revolutionizing the development of safer and more effective pest management strategies. Indeed, biotechnology has been contributing to the development of biopesticides, pheromones and repellents, but it also contributes to the development of different strategies that allow the reduction of pest population, for example, via the manipulation of symbiotic relationships between insects and microorganisms or using RNA interference (RNAi). The effectiveness of these approaches can be maximized when combined with other control measures within the Integrated Pest Management (IPM) framework, such as the use of natural enemies or implementing cultural practices. This presentation will cover the approaches currently used to manage olive tree pests, from the conventional biological control methods to the most recent ones developed via biotechnological tools (metagenomics, metabolomics and transcriptomic). These approaches have been mostly developed for controlling two key olive pests, the olive fly (*Bactrocera oleae*) and olive moth (*Prays oleae*), and more recently for the management of the main European vector of *Xylella fastidiosa*. Finally, a holistic approach that synergistically combines biocontrol methods with cultural strategies, will be proposed aiming to maximize olive pest control efficacy while minimizing adverse effects on the environment, human health, and non-target organisms.

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SE27 C07

PREDATION OF MACROLOPHUS PYGMAEUS AND NESIDIOCORIS TENUIS UNDER DIFFERENT TUTA ABSOLUTA EGG DENSITIES AND DISTRIBUTION PATTERNS IN DISHES AND PLANTS

Perdikis Dionisis

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The aim of this work was to study the predation rates of *Macrolophus pygmaeus* (Mp) and *Nesidiocoris tenuis* (Nt) in dishes and tomato plants when eggs of *Tuta absoluta* offered at different densities and distribution patterns, including egg laying patterns used by *T. absoluta* on a leaflet or plant. *Tuta absoluta* eggs were positioned randomly on a leaflet

in a dish or following four distribution patterns. In another experiment, 4 or 8 eggs in total distributed following different patterns among 4 leaflets. In larger scale, prey consumption was searched in a cage with 4 tomato plants, in one of which 30 eggs were placed or 15 eggs were placed in each of two plants. The egg consumption was very high (i.e. 93 eggs out of 130) on a single leaflet in a dish. However, when prey allocated in more leaves, it highly reduced (i.e. 3.1 eggs out of 8). The distribution pattern affected the searching ability, with Mp showing a faster prey consumption. Generally, both predators consumed more eggs when prey was distributed in more than one tomato leaflet. They consumed significantly less when the eggs placed in one out of four plants than when in the cage there was only the plant with eggs. The prey sharing in two out of four plants caused a reduction in comparison to placing the eggs in one out of four plants. The prey consumption of *M. pygmaeus* and *N. tenuis* highly depends on their prey distribution and the scale (leaflet vs plant) and its complexity and to understand the factors affecting their efficacy studies under conditions much more field-related are necessary.

SE27 C08

HOW CAN WE EXPLOIT PLANT ESSENTIAL OILS AND NATURAL ENEMIES FOR GREENHOUSE PEST CONTROL?

Michele Ricupero

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Plant essential oils (EOs) have the potential to be key components in the sustainable control of agricultural pests as their use can help reduce the negative environmental impact of synthetic insecticides. However, the non-target effects of EOs on plants and entomophagous insects used in protected horticultural crops is limited. The target toxicity of different nanoformulated EOs belonging to Mediterranean aromatic plants was assessed in the laboratory against greenhouse key pests with different feeding strategies (i.e., a chewer, *Tuta absoluta*, and two sap suckers, *Aphis gossypii* and *Phenacoccus solenopsis*). The non-target toxicity of EOs were thus evaluated on solanaceous plants and several biological control agents (i.e. *Nesidiocoris tenuis*, *Aphidius colemani* and *Cryptolaemus montrouzieri*). Finally, we assessed the potential activation of plant defenses mediated by EOs and the cascading effects on the related food networks. Most of the assayed EOs caused significant mortality on insect pests and the estimated lethal concentrations varied significantly. Although most EOs had negligible toxicity on tomato and sweet pepper plants, they showed a changing selectivity towards the tested natural enemies. Foliar applications of garlic and peppermint EOs activated defense signaling pathways and triggered different behavioral responses on pests and natural enemies in olfactory trials. These results add new insights to the current understanding of EOs for crop protection strategies in the Integrated Pest Management framework of modern cropping systems.



SKALKOTAS

08.30-11.00 Concurrent Session **28**

Sustainable Mycotoxin Management in a Climate Change Future

Chair: Dimitris Tsitsigiannis (Agricultural University of Athens, Greece)

Paola Battilani (Università Cattolica del Sacro Cuore, Department of Sustainable Crop Production, Italy)

SE28 C01

PREDICTIVE MODELING FOR A SUSTAINABLE MYCOTOXIN MANAGEMENT IN A CHANGING CLIMATE

Marco Camardo Leggieri, Paola Battilani and Dimitris Tsitsigiannis

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The profound impact of climate change (CC) on agricultural systems, food security, and safety is widely acknowledged. The increase in temperature and CO₂ levels, coupled with the uncertainties associated with extreme weather events such as heatwaves, droughts, and heavy precipitation, significantly influences the prevalence of fungi and the occurrence of mycotoxins. Mild temperatures and rainfall favour the genus *Fusarium*, while in warm and dry periods, *Aspergillus* dominates. This behaviour is noticed in different years, but also within the same growing season, making fungal co-occurrence highly probable and mycotoxin prediction very difficult. Recent outbreaks of aflatoxin and deoxynivalenol contamination in European maize, respectively produced by *A. flavus* and *F. graminearum*, underscore the urgency of addressing mycotoxin management in the context of CC. The variability of mycotoxin-producing fungi across diverse geographic regions and crop cycles emphasizes the necessity of meteo-based mechanistic model to predict risk areas for mycotoxin co-occurrence to assist stakeholders in crop chain management, so as the need for adaptive strategies. Research endeavours are focusing on the intricate relationships between co-occurring fungi, providing insights for refining predictive models. Recently, the impact of cropping systems on mycotoxin contamination was analysed using machine learning techniques and combined with modelling; a significant improvement in prediction accuracy was obtained. While certain pathosystems have received a thorough examination, the methodologies developed hold promise for broader application across different crops and fungi. These progresses offer a pathway for global mycotoxin management strategies that are both effective and sustainable. In conclusion, it is necessary to advocate solutions for adaptive mycotoxin management practices in response to CC, with a focus on using the potential of predictive modelling. By leveraging advancements in modelling techniques, sustainable solutions can be supported to mitigate the impacts of mycotoxin contamination on crop production and global food safety.

SE28 C02

PUBLIC PRIVATE COLLABORATIVE SOLUTIONS FOR SCALING AFLATOXIN BIOCONTROL TO ADDRESS CLIMATE - AGGRAVATED AFLATOXIN CHALLENGES

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Aflatoxins are potent mycotoxins produced by *Aspergillus* section *Flavi* fungi that negatively affect health, trade, and income outcomes. Unfortunately, contamination events are increasing because of climate change. Collaborative efforts between public and private sector institutions are critical for scaling up aflatoxin control solutions. Convergence of technological, institutional, and policy actions to prevent contamination before, during, and after harvest is needed to avoid every possible deficiency across the value chain that can lead to contamination. Failing to address any of the multiple critical, fragile factors leads to aflatoxin contamination sometimes at exceedingly high levels. As for any aflatoxin control solution, scaling of biocontrol products containing atoxigenic *A. flavus* strains is challenging. Cooperation among smallholder farmers, governmental entities, research institutions, industry stakeholders, and development partners, can drive the widespread adoption of integrated management strategies centered on biocontrol. Further, inclusive approaches that account for the diverse needs, motivations and capacities of affected stakeholders are needed. The presentation will address the progress, ongoing challenges, and opportunities associated with public-private collaborations in mitigating the impact of aflatoxin contamination on food safety, which is being further complicated by the effects of climate change.

SE28 C03

POPULATION GENOMICS UNVEILS THE EVOLUTION OF ADAPTATION IN FUSARIUM HEAD BLIGHT PATHOGENS IN RESPONSE TO AGRICULTURAL PRACTICES AND HUMAN MIGRATION

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Here, we explored the population of *Fusarium asiaticum*, consisting of 245 isolates, which is the main agent of yearly epidemics of head blight in southern China, and examined the impact of human migration and agricultural practices on pathogen adaptation. Firstly, we identified the evolution trajectories over the last 10,000 years of three distinct populations that could be correlated with historically documented changes in agricultural practices due to human



migration caused by warfare, referred to as the Southern Expeditions during the Jin Dynasty. Secondly, it reveals a southward decline in 3ADON-producing isolates, along with genetic changes and ascospore dispersal patterns, demonstrating that wind-mediated long-distance dispersal (~300 km) of *Fusarium*. This is the first direct demonstration that hemibiotrophic fungi can migrate over such long distances. Finally, our insights into the population dynamics, evolutionary history, and adaptation of *Fusarium* head blight pathogens were corroborated by a genome-wide study on a global panel, including strains originating from Japan, South America and the USA.

SE28 001

CONVENTIONAL VS. ORGANIC FARMING SYSTEM: BLACK ASPERGILLI POPULATION STRUCTURE, MYCOTOXIGENIC CAPACITY AND MYCOTOXIN CONTAMINATION ASSESSMENT IN GREEK WINES

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Aspergillus bunch rot, caused by several mycotoxigenic species of Black *Aspergilli* section *Nigri* is one of the most severe pre- as well as post-harvest diseases of grapevines, while contaminated grape products and derivatives with *Aspergillus* mycotoxins may have an important impact on consumers health. During a 2-year survey, the impact of conventional and organic farming systems on Black *Aspergilli* population structure in Greek vineyards was thoroughly investigated. In total, 300 isolates of *Aspergillus* spp. were selected and identified by amplicon sequencing. Four different *Aspergillus* section *Nigri* species (*A. tubingensis*, *A. uvarum*, *A. carbonarius* and *A. niger*) were identified as the casual agents of the disease in the sampled vineyards. *A. uvarum* and *A. tubingensis* were identified as the dominant species, while higher frequencies of *A. tubingensis* and *A. uvarum* were found in the organic and conventional vineyards, respectively. In vitro production of ochratoxin A and fumonisin B₂, B₃ and B₄ was evaluated in two selective media. The analysis revealed a low frequency of mycotoxigenic isolates, mainly originated from conventional vineyards. Additionally, *A. carbonarius* was identified as the main OTA producer, whereas *A. niger* was the leading producer of FB₂, FB₄, and FB₆. In total, 74 organically and conventionally produced wines were analyzed using a new Quadrupole Time-of-Flight Mass Spectrometry (QTOF MS-MS) analytical method developed to detect and quantify various mycotoxins (OTA, FB₁, FB₂, FB₃, AOH, AME, and CIT) using a modified QuEChERS extraction protocol. Interestingly, a low frequency of mycotoxin-contaminated wines was detected. However, fumonisins were identified as the main mycotoxins in Greek wines compared to OTA which was found in only one sample. This is the first study deciphering the impact of conventional and organic farming systems on *Aspergillus* section *Nigri* species in Greek vineyards, suggesting that cropping system may affect the species composition within the vineyard.

SE28 002

APPLICATION OF MUSHROOM' METABOLITES AGAINST MYCOTOXIN PRODUCTION BY FUSARIUM SPP

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Fusarium diseases cause head blight in wheat or ear rot in maize leading to significant yield losses worldwide, even causing their contamination with mycotoxins, e.g., fumonisins and trichothecenes, which have a notable impact on human and animal health. In view of promoting the use of products capable of preserving the efficacy of function whilst reducing toxicity, we here report the use of natural products that can inhibit the biosynthesis of mycotoxins. Here, we propose the use of cultural filtrate (CF), derived by the culture of asexual mycelia of *Trametes versicolor* known for its antimicrobial and antioxidant properties, and one its bioactive component, named Tramesan, an exo-polysaccharide known for its capacity to activate the plant defenses and control the aflatoxin production in *Aspergillus flavus*. We tested the activity of both CF and Tramesan on growth and mycotoxin biosynthesis of *F. verticillioides*, *F. graminearum*, and *F. langsethiae* toxin-producing strains. *In vitro* test was chosen to mimic the cereal infection caused by *Fusarium* spp. infection. We evaluated the mycelium development and the mycotoxin production by mass spectrometry analysis. CF and Tramesan have a weak inhibition on the growth of *Fusarium* spp., but they can strongly inhibit the synthesis of diverse *Fusarium*-toxins. Further studies will focus on the mode of action of CF and Tramesan on mycotoxin biosynthesis. The discovery and the application of new fungal metabolites to counteract the mycotoxin production represent a sustainable method to prevent mycotoxin accumulation in cereals.



MC2

08.30-11.00 Concurrent Session **29A**

Improving Resistance - Key to Meet Future Challenges - Plant Breeding

Chair: Frank Ordon (Julius Kühn Institute, Germany)

SE29 C01

GENETIC IMPROVEMENT OF THE RESISTANCE OF CEREAL CULTIVARS IN RELATION TO THE POPULATION DYNAMICS OF THE FUNGAL PATHOGENS

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The aim of any breeding activity is to achieve genetic improvement in important traits, of which disease resistances are among the most important. Breeding progress depends on the genetic variation available, the heritability of the trait and the size of the fraction selected. Resistance to powdery mildew, leaf rust, scald and dwarf leaf rust generally had a high genetic gain in a long-term study, while it is lower for yellow rust and net blotch, and none at all for septoria leaf blotch (Laidig et al. 2021) and Fusarium head blight (Miedaner et al. 2023). The durability of disease resistance is mainly determined by the mode of inheritance (monogenic vs. quantitative) and the population dynamics of the fungal pathogens. In general, monogenic resistances are less durable than quantitative resistances, which depend on many genes with small effects. Fusarium head blight resistance, although difficult to achieve, is one of the most durable resistances available. In contrast, leaf rust resistance, has the highest ageing effects, i.e. resistance is significantly reduced over the lifetime of a variety due to adaptation of the highly dynamic leaf rust populations by frequent sexual recombination. As a result, there are many leaf-rust races available, almost every isolate analysed has a different pathotype. The same is true for stem rust in wheat and rye, which frequently recombines on barberry. In contrast, yellow rust has no known sexual recombination in Europe and only a few races are dominant. However, rapid race change has dramatically reduced the resistance of some varieties ("breakdown"), while others have remained stable. Quantitative resistance is one of the most durable, along with older concepts such as varietal mixtures or the cultivation of segregating varieties. In the future, genome editing could provide whole series of new resistance alleles that could potentially outpace the high reproduction rate of pathogen populations for the first time.

effectiveness and durability. Towards this goal, we assessed a panel of exotic wheats containing nineteen previously genetically defined STB resistance genes or their combinations for resistance to contemporary UK isolates of *Z. tritici* at the seedlings stage. The study revealed that STB resistance genes derived from synthetic hexaploid wheat (SHW) lines provide a broad disease resistance spectrum, and that by stacking 3-4 of these it should be possible achieve resistance to all tested isolates. Introgression of these resistance genes into UK elite wheats is under way, and we have evidence that some of these genes individually could provide acceptable levels of STB resistance in the field. Motivated by this, we are now investigating >60 new primary SHWs developed at NIAB from crosses utilising several different durum wheat and goat grass accessions in the search for novel broad spectrum STB resistance genes. These lines have up so far been assessed for STB resistance under field conditions during one season, in a region with high disease pressure, and a subset of lines have also been screened using a growth room-based bioassay against recent UK field isolates of *Z. tritici*. Astonishingly, in most cases only trace percentages of fungal pycnidia were identified. Further analysis of NAM and CSSL populations derived from these lines will provide valuable insights into the genetics underpinning STB resistance and contribute to the breeding of resistant wheat varieties.

SE29 C03

IDENTIFICATION OF NEW SOURCES OF RESISTANCE IN ASPARAGUS (*ASPARAGUS OFFICINALIS*) AND PEA (*PISUM SATIVUM*) AND WHAT IS IMPORTANT FOR SUCCESSFUL RESISTANCE BREEDING

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¹Julius Kuehn Institute (JKI), Federal Research Centre for Cultivated Plants, Institute for Breeding Research on Horticultural Crops, Erwin-Baur-Str. 27, D-06484 Quedlinburg, Germany

Asparagus virus 1 (AV1) is the economically most important virus attacking garden asparagus (*A. officinalis* L.) worldwide and is widespread in all growing regions. During a complex introgression breeding program the virus resistance gene AV-1^{pro} was transferred from the wild species *Asparagus prostratus* Dumort. to *A. officinalis*. At the diploid level (2n=2x=20), a monogenic dominant inheritance mode was confirmed and the AV-1^{pro} locus was mapped on chromosome 2. The SNP with the highest LOD score was converted into a KASP marker to support marker-assisted selection. Downy mildew (*Peronospora viciae* f. sp. *psii*) causes severe yield and quality losses in pea (*Pisum sativum* L.). The segregating F2-Population 'Topas'x'Gen.27' shows a 3:1 ratio expected of a single dominant resistance gene. The resistance gene *Rpv* was mapped to chromosome 2 of the pea genetic map, with a distance of 4.4 cM to the nearest proximal flanking marker

SE29 C02

DISSECTING WHEAT-SEPTORIA INTERACTIONS

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Septoria tritici blotch (STB) is one of the most damaging diseases of wheat caused by the fungus *Zymoseptoria tritici*. High levels of genetic diversity within the pathogen population due to a dominant sexual cycle attributes to the observed loss of effectiveness of fungicides as well as disease resistance genes deployed in breeding programmes. Therefore, it is imperative to identify and characterise new disease resistance sources and deploy them rationally, maximising their



AD147. Based on the experience in resistance breeding and the high importance of Fusarium-induced root rot in the two crops, a collection and characterization of *Fusarium* spp. isolates in the important growing areas was started to determine the isolates on species level and specify their pathogenicity and virulence. Here, clear differences in the virulence of individual isolates have already been observed in both asparagus and pea. These show us how important it is to know the current pathogen status in the growing regions in order to identify effective resistances in the genetic resources and transfer them into prebreeding lines. After the successful mapping and characterization of a resistance, advanced bioinformatics analysis tools should quickly shorten the list of candidate genes. Based on these the transfer into prebreeding material can surely be facilitated in the future using genetic engineering methods, but the need for high quality phenotypic analysis and screening methodologies in the field remains a priority.

SE29 C04

PHOMOPSIS SEED DECAY OF SOYBEAN: PATHOGEN CHARACTERIZATION, GERMPLASM EVALUATION, AND DEVELOPMENT OF IMPROVED SOYBEAN LINES WITH RESISTANCE

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Soybean is one of the most economically important crops grown globally. It is widely used for human and animal nutrition, as well as for biodiesel production, with global demand for soybean increasing. However, soybean yield can be significantly reduced by many diseases. Of the over 200 reported soybean pathogens, about 35 cause major economic impact. A seed-borne pathogen, *Diaporthe longicolla* (syn. *Phomopsis longicolla*) is the major organism that causes Phomopsis seed decay (PSD) and reduces seed yield and quality worldwide. Aggressiveness of 48 isolates of *D. longicolla* was evaluated and the most aggressive isolates were identified. To facilitate investigation of the genomic basis of pathogenicity and to understand the mechanism of disease development, the genome of an isolate from Mississippi, USA was sequenced and analyzed. Analysis of the 1,221 putative genes that encode carbohydrate-activated enzymes (CAZs) indicated that 715 genes belong to three classes of CAZy that have a direct role in degrading plant cell walls. The mitochondrial genomes of eight isolates from different states in the U.S. were also analyzed. To identify new sources of resistance to PSD, over 400 soybean germplasm accessions, originating from 28 countries, were field screened. Identified new sources of PSD-resistance, such as PI 417050, PI 424324B, PI 587982A, and PI 603756, have been used to develop improved soybean breeding lines or cultivars with resistance to PSD. Germplasm lines DS31-243 (PI 700941), derived from PI 587982A, and DS49-142, derived from PI 603756, were publicly released by the United States Department of Agriculture, Agricultural Research Service (USDA, ARS) in 2022 and 2023, respectively. This research will continue to lead to future releases of improved germplasm lines and cultivars with PSD resistance and high seed quality. It will

also aid in disease management and be a benefit to soybean producers and the industry at large.d

SE29 C05

STATUS, DIFFICULTIES AND PROSPECTS FOR IMPROVING RESISTANCE IN APPLE FROM A GERMAN PERSPECTIVE

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Eating fresh fruit is the basis for a healthy diet. Around 68.8 kg of fruit are consumed on average in Germany each year. Apples alone account for 22.4 kg. Around half of these apples (~1 million tons) are produced in Germany. To meet consumer quality demands, up to 20 fungicide treatments are used to control fungal diseases. Growing resistant varieties could help to reduce the use of fungicides, but so far, there are only a few resistant varieties with good fruit quality available. Breeding resistant varieties is a time-consuming process for which suitable genetic resources are often lacking. For this reason, great efforts have been made in recent years in Germany and other European countries to collect fruit genetic resources, to optimize their conservation and to jointly organize their evaluation and use in pre-breeding. Decentralized networks such as the German Fruit Genbank have been established, in which various partners contribute to the conservation of traditional apple varieties with their collections. The varieties available in these collections were tested for trueness-to-type and genotyped with genome-wide SNP marker arrays to enable GWAS studies. Bi-parental populations with selected donors were established, which were used for mapping genes and QTL associated with resistance to apple scab, fire blight and *Diplocarpon coronariae*. Different collecting expeditions to apple centers of origin (e.g. Caucasus) were organized to systematically expand the gene pool for breeding. As a result, new resistances for fungal and bacterial diseases could be identified and genetically mapped. In individual cases, it was even possible to isolate and functionally characterize the underlying genes. This work has considerably expanded the possibilities in resistance breeding, including marker-assisted selection. The presentation will give an overview of the work of the last two decades and show, using selected examples, the problems and difficulties associated with resistance breeding in apple.



SE29 C06

NEW DISEASE-RESISTANT GRAPEVINE VARIETIES FOR A MORE SUSTAINABLE VITICULTURE

Oliver Trapp¹ and Reinhard Töpfer¹

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Viticulture and the production of wine are some of the oldest agricultural traditions with the domestication of grapevines taking place around 11.000 years ago and the oldest evidence of wine making tracing back to at least 6.000 BC. Thus, viticulture is closely intertwined with the history of humankind and is today regarded as cultural heritage in many wine growing countries. Wine regions attract tourists and wine enthusiasts all over the world, therefore marking an important economic branch even beyond the sales of wines. However, viticulture is in an age of change, as today's winegrowers face many challenges threatening the growth of grapevines. On the one hand, the changing climate has a direct effect on the vines and the increased climatic variability in the growing season makes vineyard management very challenging and difficult. Growers might face a lack of precipitation and heat waves in one year and extremely wet and humid conditions the next year. The conditions are also getting more favorable for pests and pathogens. Above all, the fungal pathogens causing downy mildew and powdery mildew have occurred in higher intensity in recent years and make intense applications of fungicides necessary in order to protect the vines and grapes. Additionally, climate change might lead to newly emerging pathogens in regions in which they were absent before. On the other hand, there is the strong demand for more sustainable production by politics and the public. The EU, for example, set the ambitious goal to reduce the amount of pesticides applied in agriculture by 50% until 2030 in its Farm-To-Fork-Strategy. As viticulture applies around two thirds of all the fungicides used in agriculture in Europe, this goal is particularly important and impactful for winegrowers. In addition, as the amount of available plant protection products is decreasing and the abundance of pathogens is increasing, winegrowers are under high pressure. One of the possible solutions for pesticide reduction is the change of grapevine cultivars from traditional susceptible cultivar to new disease-resistant cultivars. After more than a century of grapevine breeding in Europe, breeders have developed a wealth of new disease-resistant cultivars for the winegrowers to choose from. These cultivars allow the reduction of 50-80 % of the applied fungicides. The recent advances in grapevine genetics have furthermore increased the efficiency of pre-breeding and breeding and today there are several systems for the successful early selection of the best genotypes in order to develop new and improved disease-resistant cultivars. However, as consumers tend to have a very conservative approach to the wines they are buying and rather buy wines of traditional cultivars they know, viticulture has not adopted these new disease-resistant cultivars widely, yet. However, climate change and the call for sustainability will make a change in cultivars unavoidable. Winegrowers are best advised to be open to growing new disease-resistant cultivars as the future of their businesses are at risk. The stakes are actually even higher, as not only individual businesses are at risk, but rather viticulture and the whole cultural traditions and landscapes in the winegrowing regions are threatened.



MC3

08.30-11.00 Concurrent Session 30

Microbial Interactions in Ecosystems: Negative or Positive Consequences on Plant Health

Chair: Rey Patrice (Institute of Analytical Sciences and Physico-Chemistry for Environment and Materials (CNRS-UPPA) / University of Pau, France)

Stéphane Compant (AIT Austrian Institute of Technology, Austria)

SE30 C01

BACTERIA-FUNGI INTERACTIONS AND IMPACTS ON PLANT GROWTH AND HEALTH

Adrian Wallner¹, Satish K. Verma², Tomas Kiss³, Alejandro Del Barrio Duque⁴, Livio Antonielli¹, Angela Sessitisch¹, Stéphane Compant¹

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Microbiome analyses showed that fungi and bacteria cohabit in the plant environment. Separately they have been intensively studied for their impact on plant growth and health. Except some cases, little attention has however been paid to the intricacy of symbiotic fungi-bacteria associations. Using different models, we described some interactions between fungi and bacteria, including a mycorrhiza-like fungus, a saprophyte and a phytopathogenic fungus. The mycorrhiza-like fungus *Serendipita indica* holds capabilities to enhance plant growth and to confer resistance to different stresses, but it is also known to host a symbiotic bacterium, *Rhizobium radiobacter*. However, its association with other bacteria and the potential positive effects on the fungus had not been explored. We analyzed how co-inoculations of bacteria and *S. indica* influence plant growth and protection against fungal pathogens. Bacteria and the fungus seem to cooperate in the process of root colonization and some bacteria enter inside the fungus. Another recent discovery was made on an ascomycetous saprophyte. We determined that specific fungal species host bacteria through isolation, metabarcoding, curing approaches and advanced microscopy techniques. Interestingly, all fungal isolates from a same species contain several similar bacterial taxa but differences are found between fungal species. Similar approaches were applied for a basidiomycetous wood pathogen, using a large set of fungal isolates from diverse worldwide origins. The existence of a core endohyphal bacteriome was detected with functions that remain to be explored. Besides highlighting the importance of multipartite microbial interactions, we discuss implications of our results for the development/application of microbial consortium-based bioprotectants and biostimulants as well as for the understanding of fungal pathogen-bacteria associations.

SE30 C02

BACTERIA ASSOCIATED WITH WOOD TISSUES IN GRAPEVINES : FUNCTIONAL DIVERSITY AND SYNERGY WITH FOMITIPORIA MEDITERRANEA TO DEGRADE WOOD COMPONENTS

Rana Haidar¹, Stéphane Compant², Livio Antonielli², Antoine Loquet³, Amira Yacoub¹, Eleonore Attard¹, Remy Guyoneaud¹, Patrice Rey¹

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Wood degradation is a complex process during which plant polymers (i.e. cellulose, hemicellulose, and lignin) are used by microbial communities colonizing wood. In the case of grapevine trunk diseases (GTDs), which cause a heavy economic losses worldwide, fungi have been described as the main cause of GTDs, but bacteria diversity is very high in the grapevine wood. Bacteria and fungi that co-occur in the wood have various relationships that shape the bacterial and fungal population structures, which in turn impact grapevine health. Screening of bacterial strains, isolated from grapevine, against fungal pathogens involved in GTDs (e.g. *Neofusicoccum parvum*, *Phaeoemoniella chlamydospora* and *Fomitiporia mediterranea*), showed different types of bacterial-fungal interactions. While some strains inhibited the growth of these fungal pathogens, synergistic relationships were observed with other bacterial strains and *N. parvum* or *F. mediterranea*. By using microcosms, three bacterial strains belonging to *Paenibacillus* genus were selected for their abilities to degrade wood components, and to promote *F. mediterranea*-wood degradation compared to that caused by the fungus alone. The whole-genomes of the three bacterial strains were sequenced because of their interest in wood degradation. Genome analysis of these strains revealed numerous genes predicted to be involved in cellulose, hemicellulose, and lignin degradation, as well as other genes related to fungal interactions and endophytism inside the plant. After wood samples inoculations with *F. mediterranea* alone, the bacteria alone, and *F. mediterranea* with each strain of *Paenibacillus* spp., analysis of the wood-degradations that occurred in the cellulose, hemicelluloses, and lignin, were investigated by Solid-State Nuclear Magnetic Resonance method. To understand this specific bacterial-*F. mediterranea* interaction, the volatile and diffusible bacterial metabolites were identified by using gas chromatography/

mass spectroscopy and Liquid chromatography-mass spectrometry. This type of bacteria-fungus interaction could explain, at least in part, wood degradation in GTD-affected grapevines.

SE30 C03

ANTAGONISTIC MODES OF ACTION OF GRAPEVINE BACTERIA TO CONTROL THE FUNGAL PATHOGEN FOMITIPORIA MEDITERRANEA

Quiza Mesguida^{1,2}, Rana Haidar, Amira Yacoub¹, Renaud Travadon¹, Assia Dreux-Zigha², Jean-Yves Berthon², Remy Guyoneaud¹, Thierry Pigot¹, Mickael Le Behec¹, Maxence Terrasse¹, Simon Godin¹, Ange Angaïts¹, Ryszard Lobinski¹, Eleonore Attard, Patrice Rey¹

1. E2S UPPA, CNRS, IPREM, Université de Pau et des Pays de l'Adour, Pau, France, 2. GreenCell : Biopôle Clermont-Limagne, Saint Beauzire, France

Esca is a major concern for viticulture in many regions of the world, decreasing vineyard longevity and affecting wine quality, thereby causing important economic losses throughout the viticulture sector. Because no effective treatments are available since the ban of sodium arsenate in the early 2000s, research on alternative methods such as biocontrol, using microorganisms, has become essential. Our aim is to select biocontrol bacteria inhibiting *Fomitiporia mediterranea*, one of the key pathogenic fungi of Esca and the target of sodium arsenate. We studied the antagonistic activities of 59 bacterial strains originally isolated from the wood and sap of grapevines, first *in vitro*, then on microcosms consisting of grapevine sawdust from different cultivars. These studies allowed the selection of three strains that inhibited strongly the growth of *F. mediterranea*. These bacterial strains were identified as *Paenibacillus polymyxa*, *Pseudomonas lactis* and *Pseudomonas paracarnis*. Thereafter, we conducted a metabolomics analysis of the bacteria / *F. mediterranea* interactions to identify the bacterial active volatiles (by SPME GC-MS) and the extracellular metabolites (by LC-MS/MS) with potential antifungal activities. In addition, comparative transcriptomics analyses were used to determine the effects of sodium arsenate and of the selected bacteria during their interaction with *F. mediterranea*. Finally, we are evaluating the biocontrol potential of these bacteria in vineyard trials by injecting them into the trunks of vines that have previously expressed Esca foliar symptoms. The initial screening of bacteria *in vitro* and on microcosms allowed the selection of the most antagonistic strains. Their modes of action were deciphered through transcriptomics and metabolomics studies. Their efficacy in the field is being evaluated *in planta* under vineyard conditions.

SE30 001

CULTURAL MANAGEMENT OF ARMILLARIA ROOT ROT IN PEACH ORCHARDS

Schnabel G¹, Gasic K¹, Koc B¹, Scroggs C¹, Singh S¹

1. Clemson University, Clemson, United States

Armillaria root rot (ARR) caused by *Armillaria* and *Desarmillaria* spp. is the number one cause of premature peach tree decline in commercial orchards. The fungus produces infection centers, leaving behind dead and dying trees in its path. The roots remaining in soil serve as inoculum source for the next planting and trees as young as 2 years will again succumb to the disease in infection centers from the previous planting. Management options are limited. Chemical control in the form of preplant fumigation may reduce the number of newly infected trees but only by a small margin. Biological control has been tried but without success. The most popular rootstocks are susceptible to the disease and rootstocks with resistance have been difficult to produce in large quantities. In this talk we will explore advantages and disadvantages of above-ground root collar excavation to manage ARR. Trees are planted shallow on berms that are at least 36 cm high and 48 cm wide. After two years of establishment, the root collar is excavated with hoes, shovels, and air spade, to prevent the fungus from entering the lower trunk of the tree. This planting tactic has increased the productive life of a peach orchard on replant sites by about 2 years. Efforts are underway to facilitate the labor-intensive excavation process. Part of this effort was accomplished by developing a modified weed badger to flatten the soil between trees in a row thereby increasing access to the root collar around the tree. Future work of this project is geared toward automizing the root collar excavation process.

SE30 002

IMPACT OF CROPPING PRACTICES ON THE INCIDENCE AND SEVERITY OF POTATO COMMON SCAB

Peters R¹, Goyer C², Phillips J³, Nyiraneza J¹, Jiang Y¹, Gregory D¹, Crane B¹, MacDonald K¹, MacPhail A¹

1. Agriculture and Agri-Food Canada, Charlottetown, Canada, 2. Agriculture and Agri-Food Canada, Fredericton, Canada, 3. East Prince Agri-Environment Association Inc., Kinkora, Canada

Common scab of potato is caused by pathogenic *Streptomyces* species including the widely-distributed *Streptomyces scabiei*. This chronic disease for potato growers in Canada reduces tuber quality and has limited options for control. Over several years, growers in central Prince Edward Island (PEI), Canada participated in studies where various cropping practices were evaluated. Growers split fields to evaluate and compare novel cropping practices with more traditional practices. Cropping practices evaluated included tillage regimen (conventional vs. reduced), water application (rain-fed vs. supplemental irrigation), and prior crop. In all cases, soil samples were taken at tuber initiation and again at harvest. The harvested tubers were graded for yield and quality and assessed visually for the severity and incidence of common scab. Crops including buckwheat and mustard, grown in the year before potatoes, significantly reduced the incidence and severity of common scab in the subsequent potato crop. Residues of

grasses, including ryegrass and sorghum-sudan grass grown in the year before potatoes seemed to exacerbate the disease. In general, increasing soil pH led to increased common scab, but this also depended upon other prior crop attributes. In terms of water management, of 13 fields assessed over four years, some form of supplemental irrigation reduced the incidence and severity of common scab in eight fields (62%), while results in the remaining fields were variable. In terms of tillage regimen, of 10 fields assessed over three years, tillage did not affect the incidence and severity of common scab in six fields (60%), while in three fields (30%), common scab increased under the residue tillage regimen. Our work would indicate that maintaining adequate soil moisture, preventing increases in soil pH and utilizing crops with pest-suppressive properties, can contribute to common scab management while maintaining soil organic matter and microbial diversity recognized as markers of soil health.

SE30 003

REST MATERIAL FROM THE POTATO INDUSTRY: INACTIVATION OF THE RESTING SPORES OF *SYNCHYTRIUM ENDOBIOTICUM*, CAUSAL AGENT OF POTATO WART

Van Leeuwen G¹

1. NIVIP, team Mycology, NPPO-the Netherlands, Wageningen, Netherlands

Waste material resulting from the processing industry of starch potatoes poses a serious risk to the environment, in particular when re-used on agricultural fields. Plant pathogens present in the waste can re-infest soils; one of the most persistent ones is the fungal pathogen *Synchytrium endobioticum*, which causes potato wart disease. This fungus produces resting spores which can survive in soil for more than 20 years, even without a host. In a practical application on the site of a starch factory in the Netherlands, a steam machine was installed treating heaps of composted plant material. In this process, composted potato stems/leaves/roots were steam-heated at 105-115 °C for 3 to 4 minutes, after which the material was put in well-isolated containers. Temperature in these containers was checked during 48 h after deposition, and was still 80 °C after 48 h. Based on the conditions applied and realized in practice, laboratory experiments were done treating resting spores of *S. endobioticum*, diluted in sand, at temperatures of 95 and 100 °C for 1.5 and 3 minutes followed by a period of incubation at 80 °C for 0, 24 and 48 h respectively. The survival of spores was subsequently tested in a bioassay and by a RNA-PCR method. In the bioassay the positive control reacted optimally, on average seven eye pieces out of ten showed wart formation. On the contrary, in the treatments no wart formation was observed in the combination of 95/100 °C followed by a 48 h incubation period at 80 °C. We conclude that steaming of waste material from potato industry has promising deleterious effects on the persistent resting spores of *S. endobioticum*.

SE30 004

CYLINDROCARPON-LIKE FUNGI CAUSING GINSENG ROOT DISEASES IN NORTHEAST CHINA

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Northeast China is well known for cultivating ginseng (*Panax ginseng* and *P. quinquefolius*). Ginseng roots are threatened by red-skin root disease, rusty root rot and disappearing root rot. These diseases occur in all ages of ginseng and more severely in later growing years. Although the diseases are economically important, little is known about population structure and the genetic diversity of the causal agents. To conduct this study, we collected diseased ginseng roots from 12 regions representing the main ginseng-growing areas in Northeast China. Hundreds of isolates were obtained. The pathogenic complex are really complicated and partly overlapped. Among the fungal pathogens, *Cylindrocarpon*-like fungi (CLF) which are predominant populations. A total of 169 isolates with CLF anamorph were identified in two *Dactylonectria* species and six *Ilyonectria* species using morphological and molecular methods. Cross pathogenicity tests showed that all species were pathogenic to both *P. ginseng* and *P. quinquefolius*, and most of them had slightly stronger aggressiveness in *P. ginseng*. The analysis of partial sequences of the Histone H3 gene generated a high level of genetic diversity and geographic differentiation. A total of 132 variable sites were detected in 169 sequences, which formed 20 haplotypes with a haplotype diversity of 0.824. Genetic differentiation was positively correlated with geographic distance. The geographic populations in the range of Changbai Mountain were distinctly discriminated from those in other non-Changbai Mountain populations. No significant genetic differentiation was found between ginseng hosts. *Cylindrocarpon*-like fungi causing ginseng root diseases are geographically correlated in the genetic distance in Northeast China and should be managed correspondingly.

SE30 005

THE IMPACT OF GLUFOSINATE AMMONIUM AND GLYPHOSATE APPLICATION TO THE SOIL FUNGAL DIVERSITY AND POPULATION DYNAMICS

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1. Malaysian Palm Oil Board, Bandar Baru Bangi, Kajang, Malaysia

The soil microbiome is a dynamic and intricate environment that plays a crucial role in maintaining plant health and ecosystems. Alterations in the composition of the soil microbiota can have both beneficial and harmful impacts on plants, either directly or indirectly. The application of agricultural chemicals, such as herbicides, for weed management has the potential to impact soil microorganism populations. We have conducted a pilot study to examine the effects of glyphosate and glufosinate ammonium applications on the fungal microbiomes present in the soil of a mature oil palm plantation. Glyphosate isopropyl-ammonium (41% w/v) and glufosinate-ammonium (13.5% w/v) were administered

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at a rate of 3.0 L/ha and 3.3 L/ha using a knapsack sprayer with a spray volume of 450 L/ha. Soil samples were obtained at three sites within each plot at day 0 (before) and 1, 7, 28, 56, and 84 days after application (daa). The soil fungal population and diversity were assessed using amplicon sequencing data of the rDNA internal transcribed spacer (ITS). Analyses of OTUs revealed a variation in the richness (number of species) and diversity of fungal species within the treated plots before and after spraying. Nevertheless, the changes are not statistically significant, except at 84 daa. The trends were similar for the control plot; hence, it is improbable to attribute the changes to the use of both herbicides. The abundance of some species was shown to have significant changes (decrease or increase). However, additional research is required to determine whether these changes are a result of herbicide usage or other factors such as climate conditions (such as rainfall and temperature). The findings in this study show that there is no strong evidence that the application of both glyphosate and glufosinate ammonium at the recommended rate has a significant impact on overall soil fungal diversity and population.

different isolates has been verified through Koch's postulates fulfilment, further work is needed to conclusively elucidate the role of *P. vexans* and other oomycetes (mainly *Phytophthora spp.*) in the wider context of plant-microbiome-environment interactions.



Figure 1.

SE30 006

STATE OF THE ART ON THE KIWIFRUIT VINE DECLINE SYNDROME IN ITALY

Li Destri Nicosia M¹, Guaschino M², Mosca S¹, Malacrin A¹, Li Destri Nicosia M¹, Garelo M², Ermacora P³, Spadaro D², Martini M³

1. Mediterranean University of Reggio Calabria, Reggio Calabria, Italy, 2. University of Turin, Turin, Italy, 3. University of Udine, Udine, Italy

Kiwifruit Vine Decline Syndrome (KVDS) is dramatically impacting kiwifruit industry in Italy. KVDS causes kiwi vine root rots with the consequent collapse of plants within a few months after the appearance of the first symptoms (Figure 1). Once symptoms appear on the canopy, the root system is already compromised (Figure 2). After over 10 years from the first reports, the mechanisms behind KVDS are not yet fully understood, although its biotic origin has been demonstrated and the climate change is likely to play an important role in promoting the syndrome. Several putative pathogens including Fungi and Bacteria have been associated to KVDS, but only Oomycete species have been constantly detected on symptomatic plants in different geographic areas. Here we summarize results of investigations conducted in northwest, northeast and southern Italy to characterize the microorganisms associated to KVDS by analysing roots, rhizosphere and soils using conventional isolations, baiting and metagenomic analyses. Taken together, our results support the role of oomycetes, belonging to different genera, as causal agents of KVDS. In northwest and southern Italy *Phytophthora vexans* is the most frequently detected species and it is significantly more abundant in both soil, rhizosphere, and root samples collected from symptomatic plants, although, genetically different populations seem to be associated to different geographic areas. Several other oomycete species have also been detected including *Phytophthora litorale*, *P. chlamydospora*, *Phytophthora* spp., *Globisporangium* spp., and *Pythium* spp.. In particular, a *Phytophthora* taxon closely related to *P. sojae/asiatica* (clade 7) seems to play a relevant role in northeast Italy. Even if pathogenicity of



Figure 2.



BANQUET

11.30-14.00 Concurrent Session **27B**

Biological Control of Insect Crop Pests

Chair: Perdikis Dionisis (Agricultural University of Athens, Greece)

Michele Ricupero (Agriculture, Food and Environmental Science, University of Catania, Italy)

SE27 002

TAXONOMIC STUDY OF APHIDS (APHIDIDEA: HOMOPTERA) FROM PUNJAB, PAKISTAN

Tayyib M¹, Zain-ul-Abdin Z¹, Dr. Waseem Akram D¹

¹. University Of Agriculture, Faisalabad, Pakistan, Faisalabad, Pakistan

Small winged or wingless insects belonging to the family Aphididae are known as Aphids. Aphids are polyphagous in nature and feed on a large number of host plants like field crops, fruit trees, vegetables, ornamentals, and weeds. A pair of tubes are present on the upper sides of the 5th abdominal tergum. These tubes are called cornicles or honey tubes. A total of 98 species were collected from Punjab, Pakistan. Major species include *Rhopalosiphum padi* (L.), *Rhopalosiphum rufiabdominalis* (Sasaki) *Schizaphis graminum* (R.) *Sitobion avenae* (F.) *Aphis gossypii*, *Lipaphis erysimi* (Kalt) *Brevicoryne brassicae* (L.) *Myzus persicae* (Sulzer) *Eriosoma lanigerum* (Hausmann) *Sitobion avenae* *Diuraphis noxia* (M.) *Rhopalosiphum padi* *Aphis pomi* *Metopolophium dirhodum* (Walker) *Sipha flava* were recorded from the nymph cases infesting different plants. These specimens were infesting *Triticum* sp., *Jasminum sambac*, *Murraya exotica*, *Gossypium* sp., *Rosa hybrid*, *Saccharum officinarum*, *Sapindus saponaria*, and *Syzygium cumini*. *Rosa* L. (Rosales: Rosaceae) *Malus* spp. Specimens were collected from Faisalabad, Multan, Rahim Yar Khan, Sargodha, Lahore, Rawalpindi, Mianwali, Layyah, and Dera Ghazi Khan respectively. These species differ in antennae, color, size, a little horns projecting from the rear end of the abdomen. Shannon-Weiner Index value for richness is maximum 1.72 at Faisalabad followed by Sargodha, Rahim Yar Khan, Rawalpindi, Multan, Mianwali, Lahore, Dera Ghazi Khan, and Layyah respectively. A key for the aphid species attacking different host plants in Punjab is also prepared.

SE27 003

INTERNATIONAL PLANT PROTECTION CONGRESS: HEALTHY PLANT SUPPORT HUMAN WELFARE

Egbe John E¹

¹. College Of Education Oju.p.m.b 2035, Otukpo, Oju Lga Benue State Nigeria, Oju Lga, Nigeria

From time immemorial, invasive insects pose significant threats to productivity in agriculture, ecosystem balance and human health in general. The use of chemicals pesticides which is the traditional pest control method is usually accompanied with environmental and health challenges. For this reason there is every reason to explore other alternatives and sustainable approaches to manage these pests thereby impacting fewer hazards to our health and environment. A promising strategy to explore is the use of Biological Control which uses the natural enemies of these pests to regulate pest population. This abstract gives the principles and applications

of biological control in curbing and managing invasive insects. This abstract discusses the importance of identifying and understanding the ecology and biology of the target pest and the potential natural enemies. Biological control can be effectively achieved by selecting appropriate biological agent that can effectively suppress pest population while minimizing non-target impacts. The abstract highlights various biological control agents including wasps and flies which lay their eggs inside or on the target pest which leads to the death of the pest. Some pest predators such as ladybug and lacewings consume insect pests directly there by helping to reduce and manage the population of invasive pest. Pathogens such as fungi and bacteria infect pests with toxins which leads to the death of the pests. This abstract discusses the importance of combining both biological control and Integrated Pest Management (IPM) approaches such as cultural practices and monitoring. Finally, the abstract emphasizes the need for ongoing research and collaboration to ensure applicability of biological control methods. This can be achieved by advancement in, ecological modeling, biotechnology and interdisciplinary collaboration. In conclusion, the use of biological control in management and control of invasive pests promotes ecosystem health and resilience.

SE27 C09

ESTIMATING PREDATION RATES FOR ARTHROPOD PREDATORS USING MOLECULAR GUT CONTENT DATA

David A. Andow and Débora Pires Paula

Applied Ecology, Ncsu

Several methods have been published to estimate per capita predation rates from molecular gut content analysis relying on intuitive understanding of predation, but none have been formally derived. We provide a theoretical framework for estimating predation rates to identify an accurate method and lay bare its assumptions. *Per capita* predation can be estimated from the average prey quantity in the predator, the decay rate of the prey in the predator and a conversion constant to convert measured prey quantity into prey numbers or biomass. We used previously published data on aphid consumption by a ladybird beetle in a controlled laboratory feeding trial and found that the estimated predation rate (3.29 ± 0.27 aphids/h) was not significantly different from the actual predation rate, 3.11 aphids/h and significantly more accurate than previously published methods. We used our method to estimate the relative per capita predation rate for a single prey species consumed by one predator species, which eliminated the need to estimate the decay rate of the prey in the predator and the conversion constant. Ten independent estimates from a controlled laboratory feeding trial were statistically similar, indicating that the method was accurate. We then estimated the relative per capita predation rate from

field samples by the ant *Pheidole flavens* on another ant *Pheidole tristis* and by the ladybeetle *Hippodamia convergens* on the aphid *Lipaphis pseudobrassicae*. We found that higher *P. flavens* activity-density was associated with lower relative per capita predation rates. We found no significant variation among farms in relative per capita predation by *H. convergens* suggested that the farms were biological replicates. Using relative per capita predation rates can provide a rapid way to assess how a predator-prey interaction changes over space and time and may help identify factors that limit or enhance biological control of pests.

SE27 C10

ASSESSING THE BENEFITS AND RISKS OF NON-INDIGENOUS BIOLOGICAL CONTROL AGENTS FOR THE EPPO REGION

Rob Tanner, Nico Horn

European and Mediterranean Plant Protection Organization (EPPO), 21 Bd Richard Lenoir, Paris, 75011 France

The European and Mediterranean Plant Protection Organization (EPPO) is an international organization responsible for cooperation and harmonization in plant protection within the European and Mediterranean region. Under the International Plant Protection Convention, EPPO is the Regional Plant Protection Organization for the Euro-Mediterranean region. To harmonise approaches for the introduction of biological control agents (BCAs) for research and releases, the Joint EPPO/IOBC Panel on biological control agents (BCAs) has developed Standards PM 6/1 'First import of exotic biological control agents for research under contained conditions' and PM 6/2 'Import and release of non-indigenous biological control agents'. These Standards provide guidance on the dossiers for import of BCAs, and on information which should be included in these dossiers. They also provide guidance to relevant national authorities on the supervision of BCA releases and on general safeguard measures to be applied during import and releases. To help take decisions on the import and releases of BCAs, the Panel has developed the Standard PM 6/4 'Decision-support scheme for import and release of biological control agents of plant pests'. It provides guidance on a risk/benefit assessment of BCA releases. This is based on the comparison of potential negative non-target effects with potential positive environmental effects. The EPPO Standard PM 6/3 'Biological control agents safely used in the EPPO region' provides a list of BCAs which are already present in the EPPO region and have been safely used without negative non-target effects. EPPO recommends its countries to use a simplified procedure of decision taking for these BCAs.

SE27 004

SPREAD OF AN INVASIVE APHID SPECIES ON APRICOTS IN EUROPE AND ITS CONTROL

Spitaler U¹, Oberhofer S¹, Facchini T¹

¹ Laimburg Research Centre, Terlan, Italy

An invasive aphid species appeared on apricots (*Prunus armeniaca* L.) in Italy's Emilia-Romagna region in 2016, followed by Hungary in 2020, Serbia in 2021, and Germany in 2022. Panini et al. (2017) identified this species as *Myzus umecola* (Matsumura, 1917) (Hemiptera: Aphididae). First reports for European countries and preliminary studies on the control of this new pest in apricot cultivation used COI sequences from Japan for the identification of this species. In the presented study, the phylogeny by COI and 12S sequences, the overwintering in Europe, egg-laying, and its life cycle were studied. The egg-laying period and the different life stages were observed in the field in Italy. Laboratory bioassays using treated leaf disks of apricot were conducted to test the efficacy of biological and conventional insecticides according to the maximum field dose. Additionally, dose-response curves were established for selected insecticides, and the effects on colony development were evaluated. Furthermore, a field trial was conducted to investigate the effectiveness of commonly used insecticides in apricot cultivation. Samples collected in different countries provided the first insight into the phylogeny of this aphid. In South Tyrol, Italy, it was observed that less severely infested leaves showed deformations, which were visible until the leaves fell in autumn. In the case of heavy infestation, the shoots showed a decreased growth and dieback of the tips was also observed. At very high infestation densities, the aphids also directly fed on fresh shoots, particularly causing severe damage to young trees. The life cycle characterized this species as holocyclic. Regarding organic insecticides, pyrethrins showed a significant effect on adults and nymphs, while azadirachtin was less effective. Based on the results, it can be concluded that successful control of this aphid is possible with synthetic chemical insecticides, while insecticides allowed for organic production were less effective in achieving the same level of control.



**SE27 005****BIODEGRADABLE BIOPOLYMERIC CONTAINING NANO-CLAY FOR ENCAPSULATION OF ENTOMOPATHOGENIC FUNGI VIA DRYING PROCESS****Albuquerque P¹, Machado J¹, Almeida J², Bortoletto-Santos R³, Barud H¹**

1. LATEFBIO - Laboratório de Tecnologia Farmacêutica e Controle Biológico, Araraquara, Brazil, 2Universidade de Ribeirão Preto (UNAERP), Ribeirão Preto, Brazil, 3Instituto Biológico de Campinas, Campinas, Brazil

The use of bioinsecticides has become increasingly popular in recent years, especially those based on entomopathogenic fungi. The growing market demand has led to investigations to determine their potential application in protecting different species of arthropods. To enhance the viability and adaptability of these bioinsecticides against climatic factors such as UV light, oxygen, heat, and humidity, the drying process has shown to be a promising technology, in which pathogenic fungi are microencapsulated. The aim of this study is the encapsulation of entomopathogenic fungi (*Beauveria bassiana* and *Metarhizium anisopliae*) using different formulations with biopolymeric bases and nano-clay. Different mass concentrations were mixed in water under agitation until complete dissolution, followed by the drying process, with the intention of obtaining encapsulated formulations. The developed formulations containing the fungi were evaluated using techniques such as Fourier Transform Infrared Spectroscopy (FTIR), X-ray Diffraction (XRD), and Thermal Analysis (Thermogravimetric and Differential Scanning Calorimetry). The results showed that the combination of the biopolymeric matrix and nano-clay ensures the protection and viability of the microorganisms. Moreover, Scanning Electron Microscopy (SEM) showed the formation of microspheres with similar size distribution and average diameters of 6.8 µm. This innovative system suggests that biodegradable biopolymers containing nano-clay represent an effective and promising approach to improving the viability of microorganisms and increasing agricultural productivity. Additionally, this study contributes to the development of effective and ecological bioinsecticides, offering an innovative and sustainable solution to the challenges faced in modern agriculture.



SKALKOTAS

11.30-14.00 Concurrent Session **31**

Tropical Pest Management - Challenges in the Post-Covid World

Chair: Trevor Jackson (AgResearch, Lincoln Research Centre, New Zealand)

Sulav Paudel (AgResearch, Lincoln Research Centre, New Zealand)

Souleymane Nacro (INERA, France)

SE31 C01

TROPICAL PEST MANAGEMENT – CHALLENGES IN THE POST-COVID WORLD

Sulav Paudel and Trevor Jackson

AgResearch, Lincoln Research Centre, New Zealand

In tropical regions, implementing effective insect pest management programs encounters escalating difficulties due to a surge in invasive species, the impacts of climate change, intensified problems from secondary pests and challenges in managing resistance against existing pesticides. Increased global transport of people and goods provides avenues for invasive pests, such as the coconut rhinoceros beetle, to travel across the tropical regions. Climate change is expanding the favourable area for tropical pests and diseases which are moving into formerly temperate regions. In addition, tropical climates exacerbate the challenges posed by secondary pests, like the spiralling whitefly in India, which thrives in warm and humid conditions. Furthermore, continual use of insecticides is leading to pesticide resistance, allowing pests like the diamondback moth to escape control. Combination of these issues in the tropics leads to massive crop losses, the failure of production systems and loss of food security in the affected countries and underscores the urgency of developing sustainable pest management practices appropriate for tropical ecosystems. Moreover, the socio-economic dynamics of small-holder and fragmented farming systems in many tropical regions compound these challenges. Agriculture serves as the backbone of the economy in many tropical countries and the livelihoods of millions of small-scale farmers are at stake due to the challenge of pest infestations. Despite the critical role these farmers play in global food security, there is a significant disparity between their numbers and the limited resources allocated to extension and research programs. To overcome these problems there is a need for a holistic approach that integrates research, collaboration, and technological innovation supported by international financing. Technological advances can support this approach. Mobile phone technology and artificial intelligence can facilitate the rapid dissemination of pest management information to farmers, empowering them to make informed decisions in real-time before damage occurs. By fostering partnerships between researchers, policymakers, and local communities, context-specific solutions can be developed that effectively mitigate the impact of insect pests on tropical agriculture.

SE31 C02

RETHINKING IPM - CHALLENGES AND OPPORTUNITIES FOR IMPLEMENTATION AND SCALING IN AFRICA

Thomas Dubois¹

¹. International Centre of Insect Physiology and Ecology (icipe), Nairobi, Kenya

In sub-Saharan Africa, a 'perfect storm' is brewing, which has fundamental consequences for sustainable food production. Africa's population is the fastest growing in the world, creating a large contingent of youth, coupled with a fast urbanization rate. Yields of the major crops are not increasing in Africa compared to other regions in the world, suggesting that increased production is the result of increased land conversion to agriculture. Third, Africa is disproportionately faced with the burden of climate change. To facilitate a sustainable agricultural transformation, IPM should be a key principle of sustainable intensification, but needs to be adapted to the intrinsicities of smallholder farmer systems. This presentation will focus on the building blocks of bio-based IPM interventions targeting smallholder farmers on the continent - conservation, augmentative and classical biological control, highlighting opportunities and challenges for their implementation and scaling.

SE31 C03

CHALLENGES IN MANAGING SPORADIC SUBTROPICAL PESTS – A CASE OF DESERT LOCUSTS IN EASTERN AFRICA

Alexandre V. Latchininsky

Food and Agriculture Organization, Rome, Italy

The recent desert locust *Schistocerca gregaria* (Forskål) upsurge of 2020-2021 in the Near East, the Greater Horn of Africa and Southwest Asia posed an unprecedented risk to livelihoods and food security in some of the most food-insecure countries in the world. This was exacerbated by the impacts of the COVID-19 pandemic and the global response to it. In May 2021, 36.6 million people in locust-affected countries faced crisis-level food insecurity (IPC 3+). Despite the timely and accurate predictions of locust spatio-temporal spread made by the Food and Agriculture Organization of the United Nations (FAO) Desert Locust Information Service (DLIS), the response to the dramatic increase of the locust populations lagged. One of the reasons is that for many countries in the Greater Horn of Africa, desert locust emergencies are insufficiently frequent to justify maintaining permanent desert locust units. Consequently, the technical capacity of national staff in the survey and control of desert locusts, as well as regional institutional capacity, were inadequate to cope with the fast-evolving locust menace. As a result, massive swarms



of desert locusts flew across Ethiopia and invaded Kenya. The crisis was considered the worst in 70 years. Needless to say, Kenya was totally unprepared. FAO and its partners mobilized more than USD 243 million from January 2020. The response included three key pillars: i) curbing the spread of desert locusts through control and surveillance operations; ii) safeguarding livelihoods and promoting recovery through livelihood protection and farmer re-engagement packages; and iii) coordination and preparedness of the rapid surge support. Overall, FAO ensured coordination of a complex and multi-actor response building and maintaining good partnerships, including foundations and private companies, despite the uniquely challenging external context. Several innovations were deployed in the 2020–2021 response, across areas including apps, models and control techniques. Somalia stood out as a country where less hazardous locust control products, such as biopesticides and Insect Growth Regulators, have been successfully applied to a vast area of over 200,000 ha. The main lessons learned for the future are to ensure that early warning translates into early action and to continue survey operations during remission.

SE31 C04

FARMER-ORIENTED AND SCIENCE-DRIVEN PLANT HEALTH MANAGEMENT FOR WEST AFRICA

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In Africa, farmers' decision to spray their crops is most often either calendar-based or triggered by visual damage symptoms. Although IPM was introduced about sixty years ago, smallholder farmers continue to use hazardous pesticides as their first control option. We are therefore proposing a new paradigm for farmer-oriented and science-driven plant health management, anchored on three pillars: (1) real-time farmer access to decision-making, (2) pest management options relying on nature-based approaches, (3) the integration of genomics, biopesticides and habitat management practices. The development of the Farmer Interface Application (FIA) illustrates progress in educating and empowering farmers to take informed decisions. FIA assists farmers in recognizing early stages of pests in the field, guided by GPS coordinates, and determine an intervention threshold. Animation videos, voice recognition and commands guide low-literacy users through the different functions, addressing the needs of all genders, ages and social groups. For many years, the pod borer *Maruca vitrata* has been tacitly categorized as an 'indigenous' pest in Africa. However, recent population genetic studies, supported by biodiversity assessment of its natural enemies, have revealed a likely Asian origin. Based on these findings, three hymenopteran parasitoids were introduced from Asia to West Africa for confined biocontrol

assessment against the pod borer. Two of them, *Liragathis javana* and *Phanerotoma syleptae*, were experimentally released in Benin, Burkina Faso and Nigeria. Encouraged by the establishment and preliminary impact data (of up to 86% reduction of pod borer populations at pilot release sites), releases are continuing in Ghana and Nigeria. As an example of integrative solutions, women-led cooperatives in Niger have initiated the commercial production of a ready-to-use biopesticide made of locally available neem seed powder, which is packed into a small tissue bag, soaked in water overnight, with the resulting solution sprayed the next day on a range of crops.

SE31 C05

IMPROVING BIODIVERSITY FOR RESILIENT RICE-BASED ECOSYSTEMS: SUSTAINABLE STRATEGIES FOR PLANT HEALTH AND CLIMATE ADAPTATION IN CAMBODIA

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Rice is a key staple crop in Cambodia, supporting the livelihoods of most farmers. However, the country faces increased vulnerability to climate change, exacerbating risks such as the spread of insect pests. Innovative solutions are needed with biodiversity management in rice-based ecosystems showing promise for increasing resilience, but widespread adoption by farmers remains an obstacle. This study aims to evaluate the effectiveness of biodiversity management in rice-based ecosystems through a farmer participatory field study in Prey Veng province. Initial consultations identified mung bean and watermelon as preferred crops for integration into rice-based ecosystems. The field study, conducted during the 2022 wet season, with rice as the primary crop and mung bean grown on the bunds, followed by watermelon during the fallow period. The study compared four treatments: 1) rice grown without pesticides and with no bund crop in the wet season and no crop in the dry season, 2) rice grown without pesticides, but with mung bean as a bund crop in the wet season and watermelon in the dry season, 3) rice grown with pesticides but without a bund crop in both seasons, and 4) rice grown with pesticides but without a bund crop in the wet season and watermelon in the dry season. Parameters such as insect pest damage, arthropod diversity and abundance, agronomic metrics and cost-benefit analysis were evaluated. Preliminary analysis suggests lower thrips damage in pesticide-free treatments compared to those with pesticides, with consistent grain yield across treatments. Treatment and crop stage influenced the relative abundance of arthropods while diversification of rice-based systems with mung bean



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and watermelon showing higher benefit-cost ratio than other treatments. Further analysis of the first- and second-year data is underway and promises to provide insights into the effectiveness of biodiversity management strategies in enhancing resilience of rice-based ecosystems.

SE31 C06

CHALLENGES AND OPPORTUNITIES IN IMPLEMENTING VEGETABLE IPM IN AFRICA

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The vegetable production area has increased by over 2 million hectares in the last decade (2013-2022) in sub-Saharan Africa (SSA). Currently, Africa holds about 18% of the vegetable production area of the world. However, productivity remains significantly lower in Africa (8.62 t/ha) compared to the Asian (22.23 t/ha) and world (20.13 t/ha) average. Pests and diseases continue to be major production constraints, leading to low productivity of vegetable crops in SSA. Hence, vegetable producers predominantly rely on the use of hazardous chemical pesticides, which are perceived to be insurance to protect their crops from significant yield losses. However, adverse impacts of the over- and/or misuse of chemical pesticides on human as well as environmental health and raising consumer awareness of food safety have created the demand for alternatives such as integrated pest management (IPM) packages. In recent decades, significant investments have been made in researching and developing IPM packages, primarily focusing on resistant cultivars, pheromones, biological control, and bio-pesticides. These IPM innovations are gradually being adopted, especially for high-value horticultural crops, including vegetables across Africa, at varying levels. However, several barriers to IPM adoption in SSA remain to be addressed. Specifically, barriers related to the institutional environment and policy frameworks require immediate attention for the large-scale promotion and adoption of IPM in Africa. Hence, the major challenges and potential opportunities in implementing vegetable IPM in Africa will be discussed in detail based on the R&D experiences of the World Vegetable Center (WorldVeg) in SSA.

SE31 C07

NAVIGATING CHALLENGES IN BIOPESTICIDE UTILIZATION FOR TROPICAL PEST MANAGEMENT: LESSONS FROM THE AMERICAS AND BEYOND

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Biopesticides are widely used in the tropical Americas and this presentation focuses on successes and how biopesticides can be used to meet the challenges ahead for tropical pest management. The tropics are a region of high

biodiversity where there are complex food webs and pest species are accompanied by many natural enemies. Invading insects often arrive in a new environment without their natural enemies which opens possibilities for introduction of natural enemies (classical biological control) with predators, parasitoids and diseases. Insect pathogenic microbes are widely used as biopesticides and often produced in small scale biofactories. Microbial variation for virulence and environmental persistence provides the opportunity to match microbial strains to pests within the local conditions and, if necessary, reselect strains to best respond to climate change altering pest complexes and local conditions. Biopesticides can also offer an alternative control to conventional pesticides and delay development of resistance to chemical pesticides. Examples will be given from countries in the American tropics. Drawing from these case studies and others, we will examine the challenges inherent in biopesticide utilization, including product quality, formulation stability, efficacy under variable climatic conditions, regulatory hurdles, and farmer adoption. Overall, we aim to deepen understanding of the use of biopesticides in the tropics and identify factors leading to successful use and uptake in tropical pest management. The need for tailored approaches, collaborative research, and knowledge exchange is emphasized to maximize the potential of biopesticides in addressing pest challenges in tropical agriculture.

SE31 001

INTEGRATING LATE BLIGHT MANAGEMENT STRATEGIES IN POTATO PRODUCTION FOR ENHANCED EFFICACY AND SUSTAINABILITY

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Potato late blight, caused by *Phytophthora infestans*, remains a significant global threat, requiring precise interventions. This study explores integrated management strategies, including targeted fungicide application and disease forecasting using decision support systems (DSS). SimCast, a DSS combining weather-based forecasting and algorithms, and a handheld decision support tool (HH-DST) factoring in environmental conditions, management practices, cultivar resistance, and customized farmer education, were used. Alternative options like combination of phosphonates and silicon amendments, and resistant cultivars complemented these strategies. Field trials were conducted on-station and on-farm in Kenya to assess the late blight efficacy of HH-DST, comparing it against calendar and local spray regimes on potato, with additional on-station trials in Rwanda, Peru, and Ecuador. SimCast was compared with other DSS customized for late blight forecast. The efficacy of phosphonate, either applied alone or in combination with silicon amendments, was tested against mancozeb in Kenya. Disease severity,



number of sprays, yield, environmental impact, and cost-benefit were assessed. Across test countries, HH-DST reduced fungicide sprays from 6 to 11 per season, proving more effective on susceptible cultivars and yielding similar results to weekly sprays, but with reduced costs (23-57%), higher net returns (USD 76-102 per hectare higher), and increased environmental benefits. SimCast improved spray effectiveness, particularly for moderately to highly resistant cultivars. Phosphonate, combined with silicon, enhanced disease suppression comparable to conventionally used Mancozeb, with phosphonate targeting the pathogen and silicon alleviating heat stress, thus enhancing crop resilience against late blight. The integration of decision support systems, disease-tolerant cultivars, and phosphonate-silicon amendments improved late blight control on potatoes while providing educational resources to farmers. These strategies underscore the potential of innovation and crop genetics to enhance productivity, reduce environmental impact, and strengthen resilience in potato blight management. **Keywords:** Late Blight; Phosphonate; Silicon; Decision Support Systems; Potato

SE31 002

POPULATION DYNAMICS OF FALL ARMYWORM (LEPIDOPTERA: NOCTUIDAE) ON MAIZE FIELDS IN UGANDA

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Spodoptera frugiperda (Lepidoptera: Noctuidae) commonly known as fall armyworm was first detected in Uganda in 2016 and has spread to all the maize-growing districts. Different methods have been deployed to control this pest. However, there is a limited understanding of the role of the environment and farmers' practices in the abundance and damage of *S. frugiperda* in Uganda. This study, therefore, assessed the abundance of *S. frugiperda* and leaf damage levels in 69 farmers' fields of Kileleshwa, Kiryandongo, and Nakaseke districts using a longitudinal survey. We also explored the association between agronomic practices, crop phenology, and weather parameters on *S. frugiperda* damage and abundance in the smallholder farmers' maize fields. Leaf damage and incidence of damaged plants differed significantly between locations and seasons. Leaf damage and abundance of larvae varied significantly in the different growth stages. Conservation tillage, reduced weeding frequency and increased rainfall were associated with reduced *S. frugiperda* damage. No significant relationship was observed between pesticide or cropping systems with *S. frugiperda* leaf damage. However, the influence of fertilizer use on leaf damage was contradicting in

different seasons and locations. Timely and vigilant scouting, proper timing of control measures, minimum tillage practices, and crop diversity should be included in an IPM strategy for *S. frugiperda*.

SE31 003

EFFECTIVENESS OF VARIOUS SYSTEMIC AND BIOLOGICAL FUNGICIDES AGAINST COFFEE LEAF RUST IN HAWAII

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Coffee is a tropical crop with worldwide importance. In USA, Hawaii and Puerto Rico are the only commercial coffee producing state and territory. In Hawaii, coffee is #2 agricultural commodity only below seed corn. Coffee leaf rust (*Hemileia vastatrix*, CLR) is considered one of the world's most devastating pathogens of coffee and is found in nearly all coffee growing regions. Hawaii was one of the last locations to grow coffee without the disease until CLR was confirmed on Maui in October 2020. Within a year, it spread to all the islands. CLR damage reduces the tree's photosynthetic capabilities and causes premature defoliation of infected leaves. In turn, future growth and yield can be lost, and in severe cases, CLR can cause tree death. In terms of chemical fungicides, currently Hawaii's coffee growers can only use contact fungicides and one translaminar fungicide to battle CLR. Considering the urgent research need, this project aimed to identify effective systemic and biological fungicides against CLR in Hawaii. This is a multiple-year project in which we tested 7 systemic fungicides in 2021, 8 fungicides (6 systemic and 2 biological) in 2022, and 5 fungicides (3 systemic and 2 biological) in 2023 in replicated field plots (Randomized Complete Block Design) at two major coffee farms in Kona, Hawaii. CLR disease incidence (% infected leaves) and infection severity (0-5 scale) were measured at least 5 months post initial treatment in each year. Our results showed that ipyrfluxam and azoxystrobin were both very effective against CLR. Although biological fungicides tested in this research showed some efficacy, they were not as effective as ipyrfluxam and azoxystrobin. This research provided critical foundation for CLR management in Hawaii and beyond.

SE31 004

THE POPULATION BIOLOGY, PREDATION AND COMPETITIVENESS OF TWO PREDATORY MIRIDS IN RICE ECOSYSTEMS WERE ASYMMETRICALLY EFFECTED BY ELEVATED TEMPERATURE

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As temperature is an important abiotic factor in agriculture, affecting individual organisms and the entire agri-ecosystem, and natural enemies are generally weak thermal tolerant, the tangible global warming may negatively affect insect pest biological. The two mirids *Cyrtorhinus lividipennis* and *Tytthus chinensis* (Insecta: Hemiptera: Miridae) are important natural predators of planthoppers and leafhoppers in Asian tropical



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and subtropical rice. However, the effects of thermal stress on these predators remain poorly understood. The thermal tolerance, fitness, predation abilities, and transcriptomic response of *T. chinensis* and *C. lividipennis* at elevated temperatures were investigated in laboratory. *T. chinensis* was more heat tolerant than both its prey *Nilaparvata lugens* and its competitor *C. lividipennis*. *T. chinensis* not only exhibited better development, survival, reproduction, and predation capacities compared with *C. lividipennis*, but also indicated higher competitiveness when the two mirids co-persisted under high-temperature conditions. To understand the underlying mechanisms, their transcriptomes at different temperatures were sequenced. Heat shock protein (HSP) genes were identified and analyzed due to their high co-regulation during heat treatment. Quantitative polymerase chain reaction results showed that *T. chinensis* induces HSPs expression quickly and strongly over a wider range of temperature in response to heat stress compared with *C. lividipennis*. Taken together, the potential of *T. chinensis* as a biological control agent in future global warming conditions and provided insight into the thermal adaption of mirid species could be highlighted.



SKALKOTAS

11.30-14.00 Concurrent Session **29B**

Improving Resistance - Key to Meet Future Challenges - Plant Breeding

Chair: Frank Ordon (*Julius Kühn Institute, Germany*)

SE29 C07

IMPROVING RESISTANCE LEVEL TO IMPORTANT PHYTO-PATHOGENIC FUNGI IN WINTER WHEAT - WHAT WE CAN LEARN FOR THE FUTURE FROM 50 YEARS OF BREEDING HISTORY?

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Wheat is one of the world's three most important cereals and is also the most important crop for Europe, where wheat yields are among the highest worldwide. Plant breeding has led to a tremendous increase in yields over the past decades and, hence, has made considerable contributions to meet the growing demand. However, phytopathogenic fungi remain major constraints of wheat production in Europe with substantial impact on grain yield. Genetic improvements through plant breeding in order to embed heritable host resistance represents the most environmentally friendly and for growers the most cost-effective way of crop protection. However, given the adaptability of fungal pathogens and the intraspecific emergence of new virulent strains widely used resistances can often become ineffective and require relentless efforts of resistance breeding (Zetzsche et al. 2019, 2020). To assess breeding progress over the past five decades, the resistance level of 191 wheat cultivars were assessed in detail in field trials for three consecutive years (2014-2015, 2015-2016, and 2016-2017). Moreover, the same genotypes were tested in isolate-specific infection assays in the greenhouse. The results show a very clear empirical evidence that not only grain yield but also the resistance level against all four diseases have been substantially increased over the past five decades in face of strong artificial infection pressure provoked by artificial inoculation. Modern wheat varieties are also significantly more resistant against 11 of 13 different isolates of stripe rust, leaf rust and *Fusarium* than older varieties at the seedlings stage. The widespread assumption in public that breeding has tended to neglect disease resistance and that wheat varieties have been adapted only to the intensive use of crop protection cannot be substantiated with the data collected. On the contrary, in the scenario of a limited availability of fungicides the resistance status of the cultivars gains on importance in an event of strong disease pressure. In order to continue the genetic gain in resistances into the future conditions it will be necessary to exploit resistances genetic resources to be introgressed and pyramided in elite lines together with already introduced long-term durable quantitative resistances. For this purpose, we regularly assess the resistance status of large diversity panels against leaf and stripe rust isolates in order to identify potential resistance donors. By bringing together high-throughput phenotyping technologies,

the availability of full genome sequence information and the use of the latest statistical methods, there is reason to assume that the resistance level of the varieties can be further increased.

SE29 001

CONSEQUENCES OF MANAGEMENT PRACTICES ON DISTRIBUTION OF RUSSIAN WHEAT APHID, DIURAPHIS NOXIA, IN WHEAT PRODUCTION AREAS OF SOUTH AFRICA

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Russian wheat aphid (RWA) is an international pest on wheat in most countries with large scale wheat cultivation. Considerable efforts have been made to manage RWA globally and the two management options used are chemical control and breeding for deployment of resistant wheat. With widespread and continuous use of the same active ingredients of insecticides, susceptibility of RWA to these insecticides may decrease. With resistance breeding RWA populations can overcome the resistance in the wheat cultivars, resulting in new biotypes virulent to resistant wheat cultivars. For continued success of these management practices, it is necessary to monitor how the distribution of RWA populations are affected. RWA was surveyed annually, from 2010 to 2023, during the wheat growing season in South Africa in all main wheat production areas. RWA clone colonies were established from field collected samples. The biotype of each RWA clone was determined by screening its feeding damage on 11 wheat resistant sources containing resistance genes *Dn1* to *Dn9* and *Dnx* and *Dny*. Insecticide resistance of RWA clone populations were determined by screening against commonly used active ingredients, using IRAC susceptibility test (Method No:001; version 3; June 2009). In South Africa RWA showed considerable biotypic adaptations to resistant wheat cultivars, resulting in five RWA biotypes occurring in wheat production areas where RWA resistant wheat was deployed. There was widespread decrease in susceptibility of RWA populations to commonly used active ingredients in areas where chemical control was used as management practice. Given the invasive ability evolutionary adaptability, virulence, and fecundity of RWA, it remains a threat to global wheat production. RWA remain present in all major wheat production areas of South Africa. It is important that future management practices focus on sustainability instead of indiscriminate use of insecticides to control RWA.

SE29 002

VIRULENCE DIVERSITY OF PUCCINIA TRITICINA IN SOUTH AFRICA AND THE RESPONSE OF WHEAT CULTIVARS AND BREEDING LINES TO NEW RACES

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Leaf rust, caused by *Puccinia tritricina* (Pt), is an important disease of wheat in South Africa (SA). Genetic resistance provides effective control of leaf rust. However, new Pt races with wider virulence frequently emerge. Monitoring the Pt population is essential for early detection of virulence changes and the identification of effective resistance sources. To determine the virulence diversity of Pt detected in SA from 2017 to 2022 and evaluate commercial cultivars and breeding lines for their response to new races. Samples of wheat leaves infected with Pt were collected annually from commercial fields and rust trap nurseries across the major production regions of SA. The virulence variability of Pt isolates was determined using standard differentials. Seedlings of recommended cultivars and breeding lines were evaluated for their response to existing and new races. Assessments were performed in the greenhouse. Eleven Pt races were identified from 609 isolates, of which four, MFPSK, MCPSK, MFPSJ, and CNPSK represent first reports. Races MFPSJ, CFPSJ, MCDSK, CBPSG, and CDPSK were most found with average frequencies ranging from 13% to 27%. Seedling infection types of 109 wheat entries against the new and existing races revealed CFPSK as the most virulent (79 entries susceptible) followed by CFPSJ and MFPSJ (73), CDPSK (66), CNPSK (64), MFPSK (60), CCPSJ (51), MCDSK (47), MCPSK (46), CBPSG (32), and SDDNP (5). The results showed increased susceptibility of wheat cultivars SST O166 to MFPSJ and MFPSK, Tredou to MCPSK, MFPSJ and MFPSK and SST O117 to CNPSK, indicating a change in the response of cultivars to the new races. Effective resistance genes which can be incorporated into new cultivars in SA were identified. Four new Pt races were detected in this study with increased virulence to recommended wheat cultivars indicating their potential to cause rust outbreaks and significant yield losses.

have been modified and developed. These include a seedling test in which several cultivars and wild species were tested for resistance to *F. oxysporum* using two single-spore lines of different virulence. Generally, the tested cultivars showed high levels of infection. The wild relative *A. aethiopicus* was free of symptoms such as root browning or growth depression. A greenhouse test was developed to analyse resistance behaviour at later plant stages, to determine whether resistance in the seedling is maintained in the adult plant. Following the resistance tests, *A. aethiopicus* was tested for its defence response to *F. oxysporum* infection, in comparison with *A. densiflorus* and *A. officinalis* cv. Thielim. *A. densiflorus* and *A. aethiopicus* showed a hypersensitive response as a defence response, whereas *A. officinalis* cv. Thielim show no storage of oxygen compounds (Fig. 1). *A. aethiopicus* was identified as a potential new source of resistance to *F. oxysporum*.

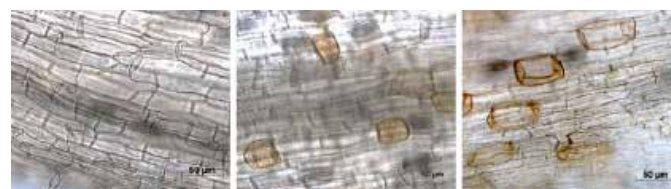


Fig. 1 DAB stain of hydrogen peroxide on *F. oxysporum* infected roots. Arrows in (b,c) mark enhanced hydrogen peroxide concentration stained by 3,3 Diaminobenzidine (a) *A. officinalis* cv. Thielim 12 hpi; (b) *A. densiflorus* 12 hpi; (c) *A. aethiopicus* 12 hpi.

SE29 004

COMPARATIVE TRANSCRIPTOME ANALYSIS OF SYNCHYTRIUM ENDOBIOTICUM, PATHOTYPE 18(T1), TO UNDERSTAND PATHOGENICITY BEHAVIOR ON RESISTANT, TOLERANT AND SUSCEPTIBLE POTATO VARIETIES

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Potato wart, caused by the soil-borne obligate fungus *Synchytrium endobioticum* (SE), is the most devastating disease of potatoes (*Solanum tuberosum* L.) worldwide. Infected tubers develop tumor-like growths on emerging sprouts, causing significant yield losses. Because of its longevity in soil (> 40 years) and lack of chemical control, SE has a quarantine status worldwide. The only effective strategy to prevent its spread is phytosanitary measures and resistant varieties. Nevertheless, SE can overcome host resistance by developing pathotypes. Today, more than 40 pathotypes exist, with pathotype 18(T1) being the only pathotype present in Greece. In this study, SE (pathotype 18(T1)) and meristematic tuber tissues of three potato varieties with varying levels of phenotypic resistance were analysed using transcriptomics. While detection of SE GAPDH transcripts at similar levels in all varieties demonstrated the pathogen's presence, 5080 and 104 transcripts were identified in the sensitive (S) and the tolerant (T) variety, respectively, and only 7 transcripts with low counts in the resistant (R) variety. Among the 47 SE transcripts with more than 1,000 counts detected in S variety, were HSP90 and HSP70 genes, components of the HSP chaperone machinery. HSP90 and HSP70 expression was highly decreased in T variety, while no expression was observed in R variety, indicating a noteworthy

SE29 003

STUDIES ON THE IDENTIFICATION OF RESISTANCE TO FUSARIUM OXYSPORUM IN DIFFERENT GENETIC BACKGROUNDS OF ASPARAGUS OFFICINALIS AND ITS DEFENSE RESPONDSS

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Fusarium oxysporum is a serious disease that causes root and crown rot in asparagus (*A. officinalis*). Large areas of asparagus production in Europe are infected, resulting in yield and quality losses at harvest. *F. oxysporum* cannot be effectively controlled by plant protection methods. Breeding resistant cultivars is therefore an important strategy to control *F. oxysporum* in asparagus. The only resistance described so far was detected in *Asparagus densiflorus*, a wild relative of garden asparagus. Until now this resistance could not yet been transferred to cultivars, so that an important aim of this work is to identify other potential sources of resistance. For this purpose, several test methods



role of the chaperone in *SE* infection. Although no significant differences were observed in *S* variety following *SE* inoculation, the HSP chaperone was significantly upregulated in *R* and *T* varieties, mainly by *sHSPs* and *HSP70* transcripts. Functional analysis of the upregulated genes in both *R* and *T* varieties revealed considerable enrichment in hydrolase and peptidase activities that are key factors in plant defense. Our results suggest that there is dynamic competition in the exploitation of the stress HSP chaperone machinery as the pathogen needs it for attack and the host for defense.

SE29 005

MANAGING OIL PALM BASAL STEM ROT DISEASE THROUGH SELECTION OF OIL PALM RESISTANT MATERIALS AS PART OF INTEGRATED GANODERMA MANAGEMENT

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Ganoderma boninense, identified as a major threat to the palm oil industry in Southeast Asia. The effect of *G. boninense* infection on productivity decline in oil palm have been considerable concerns whereby RM 1.5 billion losses due to the disease was reported. This pathogenic fungus spread through three main ways which are inoculum left by host plants, inoculum from infected trees spreading by mycelial root contact and airborne basidiospores. To date, the only practical and sustainable approach to controlling *G. boninense* is by breeding disease resistant palm lines as one of the approaches in Integrated Ganoderma Management programme. However, the success of this strategy depends on the variability of *G. boninense* isolates as the pathogen is ideally suited to cope with this selection pressure through outcrossing and prolific spore production to adapt for aggressiveness traits. This study aims to identify potential resistant and susceptible palms based on the world largest collection of oil palm germplasms, clones and commercial progenies in the world against the moderate and most aggressive strains of *G. boninense*. Based on ten months assessments of the percentage of seedling mortality due to *G. boninense* disease infection, the results show that progeny TUP 1319 was the most partial resistance at 16% mortality, followed by PUP 6189 (17%), KTG 582 (23%), PRC 1743 (23%) and PUP 5610 (25%), while the most susceptible was TUP 1508 at 88%. Therefore, this study identified potential partial oil palm progeny that could be used to minimize the impact of *G. boninense* infection.

fruit and how strawberry responds to *B. cinerea* during fruit development and ripening. The RNAseq analysis of flowers, immature fruit, and mature fruit after flower inoculation with *B. cinerea*, revealed that the majority of the defense-related genes were upregulated in flowers and mature fruit but downregulated in immature fruit. However, the preformed defense mechanisms decreased drastically from immature to mature fruit (without *B. cinerea* inoculation). Based on the transcriptome analysis we could confirm that the strong induced defense response emerging in mature fruit was not sufficient to compensate for the reduction of the preformed defense mechanisms. When focusing specifically on volatile compounds (VOCs), including alcohols, aldehydes, terpenes, we observed a strong induction of several of these VOCs upon infection but also here the induction did not compensate for the strong decrease in VOCs during ripening. We further confirmed that several of these VOCs have *in vitro* and *in vivo* antimicrobial activity against *B. cinerea*. Finally, we show that the volatilome profile can be used as a potential alternative to detect *B. cinerea* in strawberry fruit during quiescent infection prior to symptom development.

SE29 006

THE STRAWBERRY - BOTRYTIS CINEREA INTERACTION: FROM FLOWER TO FRUIT

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Botrytis cinerea is a fungal pathogen that causes grey mold in many crop species, leading to significant economic and production losses. In strawberry plants, the fungus usually infects the flower and remains quiescent in the immature fruit without causing any symptoms, while the infection progresses to symptomatic disease in mature fruit. Using transcriptome and volatilome profiling, we aimed at understanding why the fungus remains quiescent in immature



MC3

11.30-14.00 Concurrent Session 32

Eco-Epidemiological and Pathobiome Perspectives on Diseases Caused by Mycotoxigenic Fungi

Chair: **Antonio Moretti** (Institute of Sciences of Food Production of Research National Council, CNR-ISPA, Italy)
Slavica Stojkov (Maize Research Institute of Belgrade, Serbia)

SE32 C01

PATHOGENICITY, MYCOTOXIN PROFILE AND GENETICS OF PHYTOPATHOLOGICAL AND TOXICOLOGICAL ALTERNARIA SPECIES, A "FARM TO FORK" GLOBAL CONCERN

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Mycotoxigenic *Alternaria* species, causal agents of important crop diseases, represent an increasing problem in the food supply chains for their phytopathological activity and toxicological risk. Recently, phylogenetic and genomic analyses led to a deep revision of *Alternaria* taxonomy, grouping all morpho-species in 26 taxonomic Sections. *Alternaria* species can produce more than 70 metabolites, including the harmful mycotoxins Alternariol, monomethylether Alternariol, tenuazonic acid, altenuen. In 2011, EFSA published the first scientific opinion on the toxicological risk due to the presence of *Alternaria* toxins in feed and food, suggesting to pose attention to *Alternaria* contamination in food commodities. In this scenario, our activities focused on the study of phytopathogenic *Alternaria* occurring on a broad range of agri-food crops in different geographical areas. By using a poliphasic approach, based on morphological traits, gene sequencing and mycotoxin profile, about 1000 selected strains from wheat, tomato, potato, date palm, medicinal plants collected in different regions of Mediterranean basin, Iran, and Argentina were studied. Phylogenetic analyses revealed that the most frequent species occurring on cereal belonged to *Alternaria* and *Infectariae* Sections while majority of the strains isolated from the other crops belonged only to *Alternaria* Section. Very few strains belonged to *Ulocladiodes*, *Chalastospora*, *Pseudoalternaria*, *Embellisia* Sections. More than 90% of *Alternaria* Section strains were able to produce multiple mycotoxins, at a very variable amount, while all strains belonging to other Sections were not producers. The in-depth investigations carried out allowed us to demonstrate that geographical origin and host plants did not influence *Alternaria* species distribution and genetic diversity, and that *Infectariae* Section species are pathogenic only on wheat. Finally, the great genetic plasticity of this genus, that allows it to grow under a wide range of environmental conditions, represents a further factor of pathological and toxicological risk at worldwide level, also in the incoming climate change scenario.

SE32 C02

CHARACTERIZATION OF MYCOTOXIGENIC *FUSARIUM FUJIKUROI* SPECIES COMPLEX PATHOGENS ON SMALL GRAIN IN SERBIA

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The *Fusarium fujikuroi* species complex includes cosmopolitan and toxigenic species that causes Fusarium Head Blight of small grains, an economically significant disease. In Serbia, the species mostly found associated with head blight of wheat are *F. graminearum*, *F. culmorum*, and *F. avenaceum*. Species of the section Liseola such as *F. verticillioides*, and *F. proliferatum* were sporadically isolated before 2007. However, due to climate changes, a more intense occurrence of these species on small grain has been observed in the last decade. Moreover, the first report on the presence of fumonisin on wheat was recorded in 2009 in Serbia, which indicated the growing importance of this complex and the need to pay special attention to it. The aim of this study was to identify and characterize the *Fusarium fujikuroi* species complex based on morphological, pathogenic and molecular characteristics, as well as to examine the virulence of isolates of these species and their ability to synthesis fumonisins. Samples of wheat, durum wheat, barley and triticale were collected from 55 locations in Serbia. Molecular identification of isolates was performed with specific primers (VER1/VER2, PRO1/PRO2, SUB1/SUB2, FV-F2/FV-FR, FS-F1/FS-R and FS-F2/FS-R), after which the selected isolates were characterized on the basis of three gene loci (ITS, TEF 1- α and RPB2). By sequencing three gene loci, sequences were obtained on the basis of which phylogenetic trees were reconstructed for each region. The virulence of the isolates was tested on spikes of durum wheat, wheat, barley and triticale. The concentrations of synthesized fumonisins were tested using the ELISA and HPLC method. Based on the conducted molecular analyses, three species (*F. verticillioides*, *F. proliferatum* and *F. subglutinans*) belonging to *F. fujikuroi* complex were identified on small grains. A significant difference in morphological characteristics, virulence as well as toxigenicity was observed between these species.

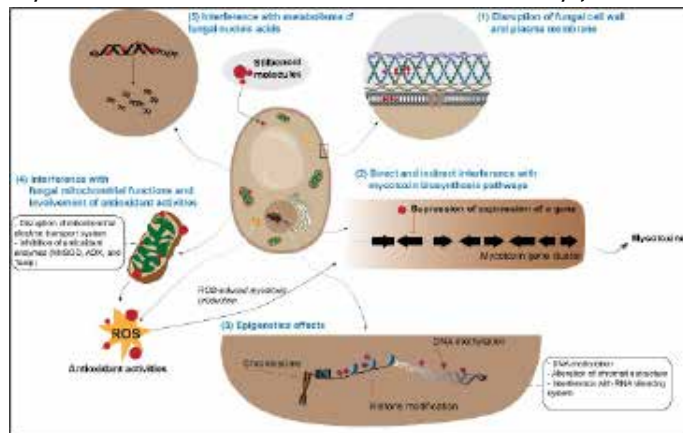
SE32 C03

UNCOVERING THE ANTIFUNGAL AND ANTIMYCOTOXIN MECHANISMS OF STILBENOIDS AGAINST *FUSARIUM GRAMINEARUM* VIA COMPREHENSIVE OMICS INTEGRATION

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An increase in contamination of agricultural commodities with mycotoxins forceful by climate change becomes an intractable problem worldwide, alarming intercontinental food security. On the other hand, to lessen the use of agrochemicals, seeking environmentally-safe antimycotoxin agents is urgent action. This current study aims at elucidating the antifungal and antimycotoxin modes of action of main active stilbenoid molecules derived from vine by-product extracts via treatments with *Fusarium graminearum* fungus - the primary causal agent of Fusarium head blight of wheat and the main producer of type B trichothecene (TCTB) mycotoxin. A monomer resveratrol (RES, 35 μ M) and a tetramer vitisin B (VIT B, 8 μ M) were the two designated candidates due to their predominance in the extracts. An array of the in vitro fungicidal bioactivities of the molecules, consisting of anti-spore germination, and inhibition of biomass production and TCTB yield, were proved in which the fungus was significantly susceptible to VIT B rather than RES. To deeper understand their modes of action underlying the antifungal and antimycotoxin activities, multi-omics approaches including mRNA-seq-based transcriptomics and LC-MS/MS-based nontargeted metabolomics were performed. The mRNA-seq data illustrated a significantly fungal global transcriptomic regulation induced by VIT B. Approximately 7,000 genes were differentially expressed upon exposure to VIT B compared to the untreated control at both 3 and 5 days post inoculation (dpi), while a very slight fungal transcriptomic regulation was observed for RES. In conjunction with metabolomics data, our study provided comprehensive insights into the modes of action of the active molecule VIT B. These findings hold promise for developing novel biofungicides exploiting viticulture wastes in order to contribute to lowering the contamination of cereals with mycotoxins and add value to the undervalued vine by-products.



SE32 001

PISTACHIO FRUIT CONTAMINATION WITH AFLATOXINS IN CALIFORNIA: SENSITIVITY ANALYSIS AND VERIFICATION
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Aflatoxins, carcinogenic mycotoxins primarily produced by soilborne fungi *Aspergillus flavus* and *A. parasiticus*, are strictly regulated in food and feed due to their health risks to humans and animals. These fungi proliferate on plant debris, release airborne conidia and contaminate crops like corn, peanuts, or pistachios. In pistachio orchards, prematurely shell opened fruits (called Early Split, ES) make 1-7% of nuts, depending on agronomical factors like irrigation, cultivar, rootstock, or pests. ES exposes seeds to *Aspergillus* contamination. In California, navel orange worm moth damage correlates with aflatoxin contamination in pistachios. Although biocontrol strategies using non-toxigenic *A. flavus* strains like AF36 have gained importance, their effectiveness is highly variable. We aimed to develop a sensitivity analysis addressing key aflatoxin contamination risk factors in pistachios by considering data from commercial orchard under different scenarios. We parameterized a mechanistic model using bibliographic data, easily implemented in Excel and R. Model verification was conducted using library samples from certified laboratory analyses of commercial orchards. Field data reveals that *A. flavus* causes more batch contamination but with lower aflatoxin concentrations (<20 ppm), while *A. parasiticus* leads to higher concentrations, albeit less frequently. Simulation results show that using the UCB-1 rootstock ensures minimal aflatoxin contamination. A two-week harvest delay escalates samples exceeding US and EU limits by 12% and 40%, respectively. The effect on the reduction of aflatoxin contamination when AF36 biocontrol agent is not applied was evaluated concerning applications that increase its proportion in the field by 20%, 40%, and 60%. In the future, the conclusions obtained with this model, raising several scenarios, can be integrated into a decision support system to design management strategies ensuring the safety of pistachios produced in California.

SE32 002

EXPLORING BIOCONTROL POTENTIAL OF GREEK NON-AFLATOXIGENIC ASPERGILLUS FLAVUS ISOLATES IN AFLATOXINS MITIGATION: INSIGHTS INTO POPULATION DYNAMICS

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Eco-friendly strategies for an effective aflatoxin mitigation seem to be a necessity to ensure food and feed safety



Concurrent Sessions

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worldwide. The most effective and common approach for pre-harvest aflatoxin management is the use of *Aspergillus flavus* non-aflatoxigenic strains. Several conducted experiments showcased aflatoxin inhibition in rates above 90%, by selected Greek endemic non-aflatoxigenic *A. flavus* strains based on *in vitro*, *in situ* and in field experiments in maize kernels and pistachios. Additionally, the biocontrol potential of 14 selected most promising non-aflatoxigenic strains was further confirmed, during a two-year field experiment, in maize, presenting significant reductions in aflatoxin contamination in rates above 94%, as compared to the control. Notably, aflatoxin levels in the treated plants were found below the quantification method's Limit of Detection (LOD). Adaptability to the local environment was subsequently investigated, by subjecting selected isolates in Vegetative Compatibility Analysis (VCA). Results demonstrated that the pistachio Greek non-aflatoxigenic *A. flavus* isolates ranked in 56 distinct VC groups, while the maize isolates in 23 VCGs, proving their widespread distribution in Greek territory. Moreover, a plausible overlapping of maize and pistachio VCGs was examined to identify population patterns between the two plant hosts. A study on Cluster Amplification Patterns (CAP) among the 14 selected non-aflatoxigenic isolates was carried out, towards the exploration of deleted genes among the aflatoxin, cyclopiazonic acid and sugar clusters. Results revealed a great genetic diversity within non-aflatoxigenic strains, varying from single to multiple gene deletions in the clusters of interest. The findings of this study contribute to the establishment of biological control systems, aiming to the prevention of food crises and the reassurance of safe-food trade in Greece and other Mediterranean regions.



BANQUET

18.00-20.30 Concurrent Session 33

Unveiling Nature's Arsenal: The Role of Volatile Compounds in Plant Protection

Chair: Nutan Kaushik (Food and Agriculture Foundation, Amity University, India)

Vicente González (4Centro de Investigación y Tecnología Agroalimentaria, Spain)

Naceur Djebali (Centre de Biotechnologie, Technopole Borj Cédria, Tunisia)

SE33 C01

VOLATILE COMPOUNDS IN POST-HARVEST PRESERVATION

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Maria Fe Andrés³, Asha Kumari¹, Yingmian L Antuhu²,
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Postharvest losses of fruits and vegetables, caused by senescence, microbial infections, and pathogen attacks, significantly impact their quality and quantity, leading to substantial economic losses. Current methods to control pathogen infestation and extend shelf life include physical techniques (refrigeration, low-pressure storage, and UV treatments) and chemical treatments. However, physical methods can cause chilling injury, and chemical methods harm the environment, human health, and lead to pesticide-resistant pathogens. There is an urgent need for sustainable methods to reduce postharvest losses. Volatile compounds (VOCs) from natural sources, such as plants and microbes, offer an environmentally friendly approach to disease control during pre- and post-harvest stages. These VOCs are antimicrobial, less harmful to humans, and can replace chemical fungicides. VOCs produced by endophytes like *Bacillus subtilis*, *Bacillus siamensis*, and *Bacillus megaterium* show antagonistic activity and promote plant growth. They reduce infections caused by plant pathogens such as rotting on tomato fruits, and improve tomato seedling growth parameters. The compounds in VOCs from these *Bacillus* strains include 2-Heptanone, Pyrazine, 2,5-dimethyl-, Naphthalene, and Benzenamine, N-ethyl-. Benzenamine, N-ethyl- has the highest antifungal activity against *R. solani*, with an IC₅₀ of about 0.09 mL L⁻¹ headspace. This demonstrates the effectiveness of *Bacillus* VOCs in reducing. Similarly, VOCs from *Zingiber officinale* (ginger) and *Piper nigrum* (black pepper) essential oils inhibit the growth of *Botrytis cinerea* and *Alternaria alternata* by 74.0-85.0%, maintaining fruit quality by reducing weight loss and preserving total soluble solids, firmness, pH, and acidity. In summary, volatile compounds from essential oils, plant extracts, and *Bacillus* strains provide an effective, natural method for enhancing the postharvest quality and shelf life of fruits and vegetables by mitigating fungal diseases.

SE33 C02

HARNESSING ENDOPHYTIC BACTERIA AS A SOURCE OF VOLATILE ANTIFUNGAL COMPOUNDS FOR POSTHARVEST DISEASE CONTROL

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Botrytis cinerea is one of the most important postharvest fungal pathogens causing significant losses in fresh fruits, vegetables and ornamentals. Synthetic fungicides are primarily used to control postharvest decay loss, but due to their hazardous use the recent trend is shifting toward safer and more eco-friendly alternatives. The use of antagonistic microorganisms is becoming popular throughout the world. This study investigated the antifungal, the plant growth promoting activities and the identity of volatile organic compounds (VOCs) produced by tomato-derived endophytic bacteria strains. The capacity of selected strains to prevent postharvest *B. cinerea* infection on tomato fruit through VOCs and soluble compounds was also studied. A collection of 50 bacterial strains was established from different organs of tomato plants sampled from six localities in Cape Bon region (Tunisia). Despite the small geographical scale, complexity and abundance of endophytic communities varied greatly according to the site of origin. Healthy tomato plants harbor diverse endophytic bacteria of *Bacillus* and *Enterobacter* genera colonizing mainly leaves with a significant enrichment with *Bacillus* strains. The *in vitro* dual culture assays showed that 36 % of the endophytic bacterial strains produce antifungal VOCs against *B. cinerea*. To our knowledge, this is the first report of VOCs antifungal activity produced by *B. nakamurai*, *B. pseudomycoides*, *B. proteolyticus*, *B. thuringiensis*, *E. asburiae* and *E. cloacae* against *B. cinerea*. About 14 % of bacterial strains produce VOCs with *in vitro* specific promoting effects on tomato seedling length or biomass production. The five selected antagonistic endophytic bacterial strains produced a core set of seven VOCs along with different strain-specific and known antifungal VOCs such as 3-Methylbutan-1-ol, sulfur-containing compounds, 2-Heptanone and Dodecanal. Tomato fruit bio-protection assay showed that the *Enterobacter* strain TR1 produces the most protective VOCs against *B. cinerea* infection with 3-Methylbutan-1-ol as a major volatile compound which totally suppressed *B. cinerea* growth and infection on tomato fruit at 0.442 mL L⁻¹ headspace, whereas the *Bacillus* strains showed better protection against fungal infection when applied as vegetative cells on tomato fruit. These results support the use of the selected strains as potential biocontrol agents to reduce postharvest decay of *B. cinerea*, as well as 3-Methylbutan-1-ol as promising antifungal volatile to apply during postharvest commercialization of tomato fruit.



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SE33 C03

ROLE OF PUSH-PUSH COMPANION PLANT VOLATILES IN THE MANAGEMENT OF THE INVASIVE FALL ARMWORM (*SPODOPTERA FRUGIPERDA*) PEST

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Fall armyworm, *Spodoptera frugiperda*, is a serious invasive pest in Africa but "Push-Pull" companion cropping can substantially reduce infestation. Our study elucidates the underpinning chemical ecology mechanisms. Headspace volatiles were collected from companion plants (*Desmodium intortum*, *Desmodium uncinatum* and *Brachiaria mulato II*) and used in bioassays and electrophysiological recordings with *S. frugiperda* and parasitoid wasps. Insect populations, plant damage and herbivore parasitism were assessed under field conditions. Coupled GC-electroantennogram (GC-EAG) recordings showed robust responses to certain aromatic and terpenoid volatile compounds. In wind tunnel bioassays, maize volatiles mixed with *Desmodium* volatiles were less attractive to moths than maize alone. In oviposition bioassays, *S. frugiperda* laid fewer eggs on maize when *Desmodium* volatiles were present. In an olfactometer bioassay, parasitoid wasps were attracted to the scent of both *Desmodium* spp. (intercrop) and the *Brachiaria* border crop. Our data provide evidence of the mechanisms underpinning the reduced *S. frugiperda* infestation in the Push-Pull companion cropping system i.e. volatiles from companion crops repel *S. frugiperda* while attracting its parasitoid natural enemies. These findings explain why Push-Pull field plots had fewer *S. frugiperda* larvae and lower crop damage than monocropped maize.

formulation, in parallel with different formulations (alginate encapsulation and application on cellulose microfiber dishcloth) of the above mentioned 4 essential oils for the same doses. Activity assessed on cellulose microfiber dishcloths remained the highest after lowering doses to 100µg/insect. Also, another follow-up experiment was designed to better assess repellent activity. Dose was doubled in such oils where the knock-down was the lightest and halved in those where the knock-down was stronger. Nearly every oil saw its repellent activity enhanced following this strategy, thus dose-response experiments were undergone in those where the knock-down had been the greater. Again, the same 4 essential oils returned the best bioactivity and were diluted to 1/10 of the original dose. The same alternative formulations as for the toxicity assays were tested for such 4 essential oils, with remarkable differences in the bioactivity presented.

Essential oils	Essential oils
Artemisia absinthium	Rosmarinus officinalis
Dittrichia graveolens	Salvia lavandulifolia x officinalis
Lavandin grosso	Salvia officinalis
Lavandin super	Satureja montana
Lavandula luisieri	Tanacetum vulgare
Mentha rotundifolia	Thymus vulgaris
Oreganum virens	Thymus zygis

Table 1.- Plant species from which the essential oils tested were obtained.

Acknowledgements: The realization of this work was possible thanks to the project: Nuevas estrategias de control del escarabajo de la trufa en Teruel. ColeopTE: I+D+i CIBR - FITE 2021

SE33 C04

POTENTIAL OF SELECTED ESSENTIAL OILS FOR THE CONTROL OF THE EUROPEAN TRUFFLE BEETLE (*LEIODES CINNAMOMEUS*)

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The bioactivity of 14 essential oils extracted from semi-industrial pilot plant (table 1), were tested *in vitro* against one of the most important emergent pests of truffle plantations nowadays, the so-called European Black Truffle beetle *Leiodes cinnamomeus* in terms of repellent activity and contact toxicity. First, a preliminary screening was conducted using filter paper as carrier in both assays, in order to select the most promising plant species. For this purpose, 0.5% (v/v) and 400µg/insect were the initial doses. From this screening, *Mentha rotundifolia*, *Satureja montana*, *Tanacetum vulgare* and *Origanum virens* were the most effective in contact toxicity (100%); regarding repellent activity, this effect was observed, with a certain knock-down effect in most of the species chosen due to neurotoxicity of essential oils. In the light of these results, a follow-up toxicity dose-response experiment was undergone for filter paper

SE33 C05

METABOLOMIC AND AGRONOMIC CLUSTERING OF BIOACTIVE ESSENTIAL OILS FROM CULTIVATED SPANISH AROMATIC PLANTS

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Essential oils (EOs) are volatile natural components extracted from aromatic plants with many biological activities including these related to plant protection against pests and diseases and are being considered as an alternative to synthetic pesticides. In this work, aromatic plants from 19 species belonging to different genera have been adapted to cultivation in preliminary field trials located in Aragón, Spain. These fields, with 50 plants of each species, were supplemented with drip irrigation from June to August (4 L h⁻¹, 5-6 h week⁻¹) for 3 years. Plants were manually harvested at 75% of blooming to evaluate yearly biomass production to be distilled in a steam distillation pilot plant, and the essential oil yield (EOY) (%)



calculated. The EOs distilled between 2017 and 2019 were analysed by GC-MS and tested against a stored product pest model (*Tribolium confusum*) in repellency, direct contact (with filter paper or crystallized cellulose as carriers) or contactless toxicity (fumigation). Thirteen of the EOs showed at least one type of bioactivity against the model insect. Some species share similar compounds that could explain their bioactivity, despite agronomic data showed significant differences. Considering chemical composition, bioactivity and agronomic performance, the EOs were grouped using Rstudio as clustering and dendrogram plotting tool, resulting in five groups. Groups 3 (thymol, carvacrol and p-cymene) and 4 (piperitenone and piperitenone oxide) hold the strongest bioactivity (lowest EC₅₀ values for repellency and toxicity, respectively). Therefore, this composition-bioactivity based grouping method holds potential to predict additional bioactivities of essential oils based on their chemical compositions, rather than phylogenetic relationships. Additionally, the agronomic data gathered for each species allows for the election of the best performing species for cultivation.

SE33 C06

COMPARISON OF EXTRACTION METHODS FOR THE DETERMINATION OF ESSENTIAL OIL CONTENT, COMPOSITION AND ANTIFUNGAL ACTIVITY OF DIFFERENT PLANT SPECIES

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Plant essential oils (EOs) are gaining interest as biopesticides for crop protection. The antifungal effect of some EOs is one of their most important crop protection properties. In addition to these direct effects on the pathogen, it has been reported that EOs can induce plant defenses (priming effects) resulting in better protection against the pathogen. Considering the growing importance of EOS as active ingredients, the domestication and cultivation of Aromatic and Medicinal Plants (AMPs) to produce chemically stable EOs contributes to species conservation, provides sustainability of the production and lower variations in active ingredients. Otherwise, the essential oil content and consequently their bioactivity can vary with development stage and by extraction methods. The aim of the study was to compare a laboratory distillation in a Clevenger apparatus and steam distillation in a pilot plant to extract EOs from plant species belonging to the genera *Artemisia*, *Dittrichia*, *Geranium*, *Lavandula*, *Mentha*, *Origanum*, *Rosmarinus*, *Salvia*, *Satureja*, *Santolina*, *Tanacetum* and *Thymus* experimentally cultivated. The EOs were analyzed by GC-MS and tested in vitro against phytopathogenic fungi (spores of *Alternaria alternata*, *Botrytis cinerea*, *Fusarium oxysporum*) to select the most active and less phytotoxic one (against *Lolium perenne*) as plant candidates. Differences in activity and composition have been found between the laboratory and pilot plant extracted oils. Therefore, the semi-industrial conditions for

the extraction of bioactive EOs must be optimized case by case to improve the EO content in bioactive compounds.

SE33 C07

VOLATILE COMPOUNDS FROM *BACILLUS SIAMENSIS* NKIT9 INHIBIT MYCELIAL GROWTH OF *RHIZOCTONIA SOLANI*

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Rhizoctonia solani, a widespread fungus, affects crops in both greenhouse and field environments, leading to plant damping-off and fruit rot, and consequently substantial yield losses. This study explores the use of bacterial volatile organic compounds (VOCs) as an alternative to chemical pesticides to control *R. solani* infections. Bioassays conducted in sealed plates demonstrated that VOCs produced by *Bacillus siamensis* strain NKIT9 significantly inhibited *R. solani* mycelial growth by over 60%. Solid-phase microextraction-Gas chromatography-mass spectrometry (SPME-GC-MS) analysis identified more than 40 volatile compounds, including 2-Undecanone, 6-methyl-2-heptanone, Heptadecane, and β -fenchol, which are known for their antifungal properties. Among these, 2-Undecanone exhibited the highest antifungal activity against *R. solani*.



SKALKOTAS

18.00-20.30 Concurrent Session **34**

Advances in Biosecurity Measures for Emerging and Invasive Pests

Chair: Rajan Sharma (ICRISAT, India)

Lava Kumar (IITA, Ibadan, Nigeria)

SE34 C01

BIOSECURITY FOR FOOD SECURITY: SAFEGUARDING CROP GENETIC RESOURCES FROM INVASIVE BIOLOGICAL THREATS

Rajan Sharma¹

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The availability of diverse germplasm is essential for the success of any crop improvement program. The germplasm conserved in the genebanks and improved breeding lines available with the breeding programs are vital and irreplaceable heritage resources, which must be shared with the global community. The sharing of germplasm will help to realize the dream of food security in most of the developing countries. Studies on the origin and use of food crops indicated that about 69% of national food supplies are derived from crops of foreign origin. This emphasizes the importance of the intra- and intercontinental exchange of seed for use in crop improvement and grains for consumption. However, the inherent ability of seeds to serve as carriers of pests and pathogens increases the risk of transboundary spread of pathogens along with the seed movement which has been recognized as a high-risk factor for international germplasm exchange. Though quarantine and phytosanitary procedures have been established to facilitate safe exchange of germplasm, most of the developing nations are generally not able to maintain adequate levels of biosecurity capacity and capability to safeguard their plant resources. Improving pest diagnostics protocols, pest surveillance, treatment protocols, capacity building and establishing network of diagnostic laboratories will help safeguard plant resources from exotic/ alien invasive pests. The plant quarantine lab at ICRISAT performs seed health testing according to the standard ISTA protocols and facilitates export and import of germplasm of ICRISAT mandate crops across the globe. As of 2023, ICRISAT through National Bureau of Plant Genetics Resources, India has facilitated the export of 1,349,395 seed samples to 174 countries and import of 202486 seed samples of its mandate crops from 96 countries. This has been achieved by strictly adhering to the phytosanitary standards and protocols with no report of introduction of any exotic pests, pathogens, or weeds.

sub-Saharan Africa (SSA). BBTv has caused serious damage in 17 countries and is continuing to spread. Surveillance for BBTv is challenging due to the small and fragmented production of bananas in heterogeneous fields across large areas, making conventional surveys difficult, time-consuming, and expensive. To help solve this problem, we developed a Banana Mapping Tool that uses remote sensing, machine learning (ML), and artificial intelligence (AI) to support BBTv surveillance. Utilizing synthetic aperture radar (SAR) data from Copernicus Sentinel-1 satellite and AI algorithms like Random Forest (RF) and Convolutional Neural Networks (CNN), we mapped banana fields in Nigeria, Benin, Togo, and Ghana. The model development and training involved about three million data points extracted using the training polygons of known classes amounting to 3,256,321 pixels, with 66% being for training and 33% for testing. Model training involved UAV imagery and ground truthing using GoPro action camera photos, with seven classes considered: banana, built-up areas, croplands, shrubland, forest cover, water body, and herbaceous wetland. RF and CNN models achieved 86 and 87% accuracy in identifying banana fields, respectively, demonstrating the potential for risk modeling and surveillance support. The SAR VH and VV dual polarization band data were obtained over the study area for two months (August and September) in 2017, 2020, and 2023, and it was used to create precise banana footprints by extracting banana-predicted pixels. Utilizing SAR data, our models generated hectareage estimations exceeding previous crop datasets like SPAM and CropGRID, validated against known banana plantations. The banana crop mapping method established in this study offers a reliable approach for remotely sensed banana mapping. It contributes particularly to BBTv surveillance, aiding informed biosecurity decision-making to protect bananas from invasive diseases.

SE34 C03

ROLE OF RESEARCH LABORATORIES ACCREDITED WITH ISO/IEC 17025:2017 TO FOSTER BIOSECURITY AND BIOSAFETY: A CASE STUDY IN NORTHERN ITALY

Mezzalana Monica¹

1. DISAFA, University Of Turin, Italy, 1. AGROINNOVA, University of Turin, Italy

In the frame of the regulation EU 2002/89/CE, a National Plant Protection Organization (NPPO) can accredit university and research diagnostic laboratories to carry out plant testing with official recognition. The role of these reference laboratories is primary to provide updated scientific competence in the development of diagnostic tools and in the study of aetiology and epidemiology of plant pathogens to support NPPOs surveillance and monitoring activities. The laboratory of the Interdepartmental Centre for Innovation in the Agri-environmental sector AGROINNOVA, University of

SE34 C02

REMOTE SENSING AND AI TOOLS FOR ENHANCING BIOSECURITY MEASURES: THE CASE OF BANANA MAPPING AND BBTv SURVEILLANCE IN SUB-SAHARAN AFRICA

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1 International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria

The Banana Bunchy Top Virus (BBTV) is an introduced virus and has emerged as a major threat to banana production in



Turin, Italy, accredited with ISO/IEC 17025:2017, carries out standardized diagnostic activities on horticulture, cereal, fruits, and ornamental crops in the private sector and in the frame of national and international research projects. In the area of seedborne diseases, the laboratory developed molecular diagnostic tools, such as qPCR and LAMP, for the detection and quantification of *Fusarium fujikuroi* in rice seed. Lately an exhaustive study was carried out on the aetiology of maize root and stalk rot caused by several species of *Fusarium* spp. reporting for the first time in Italy the presence of three species of *Fusarium* belonging to three different species complexes causing the disease in seed multiplication plots of important seed companies. The results of seed testing activities carried out on maize by the laboratory has also helped to monitor the quality of the seed commercialized in Italy and monitor the presence of new or emerging pathogens such as *Trichoderma afroharzianum*, causing a severe cob rot of maize. The data provided supported seed companies in the certification process and the NPPOs to gain knowledge on the phytosanitary situation of important economic crops produced on the territory. A successful strategy to prevent the outbreak of dangerous plant diseases is the continuous surveillance that requires resources, efficient communication, effective national and international networks between the academy and the NPPOs.

SE34 C04

THE INTERNATIONAL POTATO CENTER (CIP) - GERmplasm HEALTH UNIT (GHU) SAFEGUARDING GENETIC RESOURCES FROM EMERGING PESTS FOR THE FUTURE

Giovanna Müller¹, Jan Kreuze

1. International Potato Center, La Molina, Lima, Peru

The Consultative Group on Agricultural Research (CGIAR) consists of fifteen research centers, eleven of which have Genebanks that distribute worldwide materials from their very diverse germplasm collections with the aim of preserving their biodiversity but also to promoting its use through research projects and breeding purposes; to accomplish this critical objectives these Genebanks must comply with national and international regulations to guarantee the phytosanitary quality of the material and ensure their safety trans-national movement. To comply with these phytosanitary regulations, the CGIAR proposed the establishment of the Germplasm Health Units (GHUs) in the early 1990s, with the goal of performing the phytosanitary testing and sanitation on germplasm collections held in custody by the CG Genebanks. The CIP - Germplasm Health Unit was one of, if not the first, GHUs established within the CGIAR Centers and has been instrumental in the successful distribution of CIP's genebank collection as well as breeding material which has contributed to efforts to increase clonal crop productivity and biodiversity, for instance, through repatriation of clean materials. The services provided by CIP-GHU are constantly being improved, as is the main principle acquired after 12 years of ISO/IEC 17025 accreditation and virus testing procedures have evolved over time, starting from evaluating symptoms directly or after transmission to biological indicator plants, evolving to electron microscopy, ELISA, PCR, nucleic acid hybridization and more recently high throughput sequencing (HTS) approaches. We will discuss the progression of ongoing efforts of CIP-GHU to protect genetic resources from emerging pests in the future.

SE34 C05

MANAGEMENT OF EMERGING AND INVASIVE PESTS OF FOOD LEGUMES AND CEREALS GERmplasm IN CWANA REGION

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1. International Center for Agricultural Research in the Dry Areas (ICARDA), Terbol Station, Zahle, Lebanon, 2. ICARDA, Rabat, Morocco

The global economic cost of invasive pests has been estimated at \$1.29 trillion over the past 50 years. Numerous reports of the invasion of transboundary pests into new areas have indicated that these pests are driven by agricultural intensification, international trade and climate change with adverse ecological, environmental and/or economic impacts. The Germplasm Health Units (GHUs) network that represents 11 CGIAR centers is a significant step to prevent the spread of invasive pests through global germplasm distribution activities. ICARDA's GHU conducts regular monitoring, clearance and documentation of germplasm movement for food legumes and cereals in collaboration with the National Plant Protection Organizations (NPPOs) in Lebanon and Morocco, where ICARDA has platforms for crop breeding, germplasm multiplication, evaluation and genetic resources, by ensuring compliance with the International Plant Protection Convention (IPPC). It uses comprehensive methods that focus on preventing the entry, establishment and spread of invasive and emerging pests. In this context, ICARDA's GHU applies international phytosanitary measures as part of inclusive and multidisciplinary procedures to eradicate and manage invasive and emerging pests to save germplasm. Testing of all exchanged germplasm is an essential task by investing in different modern diagnostic tools starting with a direct inspection method to DNA/RNA-based techniques. Rejected germplasm that cannot be treated or cured is terminated by autoclaving. In the last five years, ICARDA's GHU tested about 506k seed samples with a total of 3,356k diagnostic reactions and removed 1.2% of the tested samples that were found to be infected with pests to facilitate exchanging of 1,580 consignments from/to 93 countries. By that ICARDA's GHU demonstrates a proactive approach to addressing the challenges posed by invasive and emerging pests and serves as a key mechanism for ensuring the health and integrity of germplasm movement.

SE34 001

REVISIT A CASE OF FRESH TABLESTOCK INTERCEPTED IN CFIA REGULATORY ACTION

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Numerous fungi, viruses, nematodes and bacteria can infect potato plants and tubers and cause various diseases, many of which are regulated worldwide. In Canada, the CFIA is responsible for the Potato Post-Entry Quarantine (PPEQ) Program, which guarantees the supply of seed of high varietal integrity and free of potato diseases. Prior approval is required to import field-grown seed potatoes and fresh potatoes from areas other than the continental US through PPEQ program. In this presentation, we revisit a specific case in 2015 where CFIA intercepted the illegal import of fresh table stock potatoes

originating from Bangladesh in three retail locations in Toronto. The shipments were seized for quarantine disposal and samples were analyzed for the presence of soil and potato diseases for research purposes including but not limited bacterial brown Rot (*Ralstonia solanacearum* R3Bv2, and common potato viruses. Sub samples were also provided to the PPEQ program for grow-out and testing for additional common and exotic potato viruses. NGS detection followed by diagnostic confirmation using PCR, qPCR and reverse transcriptase (RT)-PCR analyses indicated the presence of potato viruses PVS, PVX, PLRV and PVY^O, PVY^{NTN} and the presence of *R. solanacearum* R3Bv2. *R. solanacearum* R3Bv2 detected using real time PCR was further confirmed using immunostrip testing and bacterial isolation as *R. solanacearum* Phylotype II and race 3 biovar 2 using multiplex PCR assays, multiple real-time PCR assays, and biotyping. The NGS results were validated and supported by bioassay, conventional and real-time RT-PCR and some other diagnostic methods (e.g. ELISA, RFLP). Other potato pests were also revealed by NGS including viruses (Tomato chlorosis virus and Pepino mosaic virus) and phytoplasmas with PCR and Sanger sequencing confirmation. NGS followed by validation provides a rapid strategy (~5 weeks) that can significantly reduce the time required for general PPEQ program (55 weeks).



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SE34 002

USING PHEROMONE AND SMART TRAPS TO CONTROL LYMANTRIA DISPAR IN EUROPEAN COUNTRIES

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The gypsy moth, *Lymantria dispar* (L.) (Lepidoptera: Erebidæ) is a polyphagous species that infects forests in Europe, Asia, Africa, and North America. It constitutes a severe environmental problem as it is a voracious eater that defoliates entire trees and causes health problems to humans and animals. The LIFE eGymer project aims to utilize non-chemical control by developing and implementing e-traps, novel trapping devices, mass larval trapping and mating disruption techniques to remotely monitor and effectively control *L. dispar* in three European countries (Slovenia, Spain, and Greece). Based on our results in 2022 and 2023, there are specific trap devices that capture more *L. dispar* adults than others, but the ranking of traps varied according to the geographical zone. For the mating disruption application, the gel was applied on time when the first captures were noted (early July). This technique works perfectly, since same results have been received at all strategic sites, where the gel was applied. Furthermore, there was a significant reduction in the trap captures, as compared with the control areas. The overall results clearly illustrate that there are good potentials for the techniques that have been evaluated here to be used in area-wide management strategies to mitigate *L. dispar* expansion.



MC2

18.00-20.30 Concurrent Session 35

Chemical Ecology and Biological Control

Chair: Antonino Cusumano (University of Palermo, Italy)

Panos Milonas (Benaki Phytopathological Institute, Greece)

SE35 C01

PLANT-PLANT COMMUNICATION VIA VOLATILES - UNEXPLORED PHENOMENON OF PLANT RESISTANCE TO PESTS

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Increasing diversity within fields through cultivars mixtures could be an effective strategy to reduce aphid pressure. Recent studies on the impact of cultivar mixtures on pest suppression demonstrated mixed findings. Some studies report a reduction in the size of the aphid population, while others show no discernible effect. Therefore, it is crucial to comprehend these inconsistent outcomes both ecologically and practically. Volatile organic compounds (VOCs) play a pivotal role as mediators in the mutualistic interactions between plants, as they can trigger defense responses in neighboring plants. In our studies, we explored the influence of cultivar mixtures on aphid suppression, delving into the potential involvement of volatile organic compounds in interactions between undamaged cultivars as an underlying mechanism contributing to observed aphid responses. Our findings present novel evidence that specific cultivar interactions via VOCs can affect the growth of neighboring plants and prime defense responses. These induced responses proved highly effective in reducing aphid feeding, inhibiting performance, settling, and thus reducing population development. Notably, these bottom-up trophic effects were only evident in selected cultivar mixtures, highlighting the genotype-dependent nature of plant-plant interactions through volatiles. The findings of our research on the volatile interactions between specific cultivars in mixtures will help to develop innovative pest management strategies and implement sustainable and environmentally friendly crop production systems.

SE35 C02

FRIENDS OR FOES? USE OF HOST AND HOST PLANT-DERIVED VOLATILES BY AN OLIVE FRUIT FLY PARASITOID

Giannoula Bogka^{1,2}, Eirini Anastasaki³, Panagiotis Milonas³, Aikaterini Psoma³, Emmanouil Kabourakis⁴, Bas Zwaan¹, Bart Pannebakker¹, Nina Fatouros²

1. Wageningen University & Research, Laboratory of Genetics, Wageningen, The Netherlands, 2. Wageningen University & Research, Biosystematics Group, Wageningen, The Netherlands, 3. Benaki Phytopathological Institute, Laboratory of Biological Control, Kifissia, Greece, 4. Hellenic Mediterranean University, Laboratory of Olive, Vine & Agroecological Production Systems, Heraklion, Greece

The olive fruit fly, *Bactrocera oleae* is the most important pest of olive (*Olea europaea*), and its larva causes considerable damage to olive fruits, resulting to serious quantitative and qualitative losses. In response to herbivore attack, plants “cry for help” by emitting volatile organic compounds which play an important role in the communication between plants, herbivorous insects, and the natural enemies of herbivores (e.g., parasitoids). Parasitoids rely on chemical signals (i.e., semiochemicals), derived from the habitat, host plant or/and host insect itself, in order to detect their host. We studied the olive, the olive fruit fly, and its larval endoparasitoid, *Psyttalia concolor*. The aim of the study was to investigate: (a) whether volatiles induced by egg deposition (oviposition-induced plant volatiles – OIPVs) or larval feeding (herbivore-induced plant volatiles – HIPVs) by *B. oleae* attract *P. concolor* wasps, (b) whether olive responds locally (local response) or/and systemically (systemic response) to feeding damage by the *B. oleae* larvae, and (c) the role of host cues to parasitoid attractiveness. We conducted two-choice bioassays to test the preference of *P. concolor* wasps to different olfactory stimuli. Moreover, we collected headspace volatiles and analysed the composition of the volatiles emitted from fruits and branches. We found that the wasps showed a strong preference to HIPVs emitted by larval-infested fruits and to cues emitted by virgin and mated *B. oleae* females. Qualitative differences in volatile profiles of egg and larval-infested fruits were found. Furthermore, volatile analysis showed that olive changes volatile emissions locally and systemically in response to larval infestation. Our results suggest that HIPVs and host cues play an important role to the parasitoid attractiveness. Chemical analysis revealed promising candidate compounds for further testing in order to improve the efficacy of *P. concolor* wasps as biological control agent of the olive fruit fly.

SE35 C03

EFFECTS OF NECTAR-INHABITING BACTERIA ON PARASITOIDS' LONGEVITY

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Bacteria from the phyla Actinomycetota, Bacillota, and Pseudomonadota have been associated with the nectar of buckwheat *Fagopyrum esculentum* Moench (Polygonaceae: Polygonales). This flowering plant has been considered important for Conservation Biological Control due to its beneficial effect on the performance of parasitoids attacking economically important insect pests. However, the impact of nectar-inhabiting bacteria from buckwheat nectar on the performance of parasitoids is not known. In this study, the effect of bacterial isolates from buckwheat nectar on the longevity of female adult parasitoids was tested. Synthetic nectar was prepared and inoculated with 14 individual bacterial isolates. The non-fermented (control) and bacterial-fermented synthetic nectars were provided *ad libitum* to the hymenopteran parasitoids *Anagyrus vladimiri* Triapitsyn sp. n (Encyrtidae), *Anastatus bifasciatus* (Geoffroy) (Eupelmidae), *Aphelinus abdominalis* Dalman (Aphelinidae), *Aphidius colemani* Viereck (Hymenoptera: Aphidiidae), *Diglyphus isaea* (Walker) (Eulophidae), *Encarsia formosa* Gahan (Aphelinidae), *Eretmocerus eremicus* Rose and Zolnerowich (Aphelinidae), *Ooencyrtus telenomicida* (Vassiliev) (Encyrtidae), *Trichogramma brassicae* Bezdenko (Trichogrammatidae), *Trichopria drosophilae* Perkins (Diapriidae), and *Trissolcus basalis* (Wollaston) (Scelionidae). For each adult female parasitoid, the total number of days alive (longevity days) was recorded for the statistical analysis. The bacterial-fermented nectars affected the longevity of the parasitoids in a positive, negative, or neutral species-specific manner compared to the control, with most positive and few negative effects recorded mainly for Bacillota isolates.

resistance has not been found in oilseed rape and *B. napus* resyntheses, but could be demonstrated for *Eruca sativa* and *Sinapis alba*. To identify potential chemical resistance markers, we compared semi-polar metabolite fraction profiles of resistant and susceptible *E. sativa* (5) and *S. alba* (15) accessions. Discriminating metabolites (derivatives of glucosinolates, amino acids and small phenolic compounds) were tested for bioactivity. b) Biotests with the Colorado potato beetle and screening wild and cultivated potato lines by liquid chromatography/tandem mass spectrometry revealed a new, bioactive metabolite from *Solanum bulbocastanum* and numerous 100 structural derivatives in different *Solanum* species, which are promising candidates for resistance breeding. Using chemical diversity in crop plants and their wild relatives can help to understand the mechanism of crop plant resistance to insect pests causing high losses in yield, quality and economy. Moreover, this diversity provides a source of bioactive plant metabolites that are promising candidates for natural plant protection and can serve for marker-assisted resistance breeding or the development of eco-friendly, biological plant protection measures.

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SE35 C05

OVIPOSITION INDUCED PLANT VOLATILES PRIME BRASSICA NAPUS DEFENCE RESPONSE

P. Otto¹, A. Kergenteuil², G. Célestin¹, M. Valantin-Morison¹, F.G Pashalidou¹

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Plants frequently use warning cues from their environment that provide information about the risk of encountering herbivores or pathogens. Plant responses to such cues often entail defense priming, which enables faster deployment of induced defenses following subsequent attack in a wide range of systems. Priming can be used within the framework of IPM towards alternative solutions to the chemical control of pests. We have recently show that the annual brassicaceous plant, *Brassica nigra*, responds to warning cues associated with oviposition-induced plant volatiles (OIPVs) emitted by undamaged neighboring plants that harbor *Pieris brassicae* eggs. Focal *Brassica nigra* plants can detect volatiles produced by egg-infested plants in their vicinity and these volatiles not only prime defenses in the focal -receiver plant, but also elicit changes in life history strategies, namely a rapid shift from growth to reproduction that enhances fitness via reproductive escape. Here we tested whether *Brassica napus*, one of the utmost importance economic crop plant, responds with priming of plant defenses and increased reproduction to oviposition induced plant volatiles (OIPVs) emitted by (1) neighboring plants of the same species. This was done by measuring *Pieris brassicae* larvae performance on OIPV-exposed *B. napus* as a proxy for defence. We found pronounced effects on herbivores performance. To further explain the observed results, we conducted OIPVs collection and identification experiments for each conspecific and heterospecific treatment group.

SE35 C04

USING CHEMICAL DIVERSITY APPROACHES TO CONTROL POLLEN BEETLE (*BRASSICOGETHES AENEUS*) AND COLORADO POTATO BEETLE (*LEPTINOTARSA DECEMLINEATA*)

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The lack of effective plant protection agents for crop cultivation due to societal restrictions and the rapid development of resistance increase the urgency to find new resistance traits and plant protection agents. Here we apply a chemical diversity approach using wild relatives of crop plants and non-targeted metabolomics to protect a) oilseed rape (*Brassica napus* L.; Brassicaceae) from the pollen beetle (*Brassicogethes aeneus*) and b) potato (*Solanum tuberosum*) from the Colorado potato beetle (*Leptinotarsa decemlineata*). a) We studied the natural variation of metabolite-mediated resistance to adult pollen beetles in brassicaceous plant species and initiated the intergeneric transfer of resistance into oilseed rape. Such



SE35 C06

WHAT MAKES LARVAL ORAL SECRETION OF THE MEDITERRANEAN CORN BORER SO ATTRACTIVE TO ITS PARASITOID?

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Biological control (BC) offers environmentally friendly solutions for protecting crops against pest insects. The performance of parasitoids, which are successfully used in BC programs worldwide, rely on their ability to effectively locate and recognize their hosts in the field. In sub-Saharan Africa, the parasitoid *Cotesia typhae* (Hymenoptera: Braconidae), is specialized on larvae of the Mediterranean corn borer *Sesamia nonagrioides* (Lepidoptera: Noctuidae) on wild Poales plants. However, as *S. nonagrioides* is an important maize pest in southern Europe, the use of *C. typhae* as a biological control agent against *S. nonagrioides* is currently under study. Here, we focus on host recognition and acceptance mechanisms underlying parasitoid adaptation to a new host population, by studying the role of α -amylase enzymes from the oral secretions of host larvae on these processes. Based on three genes encoding *S. nonagrioides* α -amylases (SnAmy1, SnAmy2 and SnAmy3), we developed a protocol to synthesize these enzymes and test its influence on the behaviour of parasitoid wasps in the lab. We measured the intensity of antennation (proxy of recognition) and attempted of ovipositor insertion (proxy of acceptance) in the presence of these α -amylases (isolated or in synergy). Each of the α -amylases tested triggered host recognition and acceptance behaviour by *C. typhae*. These results open new perspectives for using these enzymes as egg-laying stimulants in the context of *C. typhae* mass rearing for biological control of *S. nonagrioides* in France. Recognition, by the sensory system of an insect, of a protein of this size has very rarely been observed. The perception mechanisms of these α -amylases by the wasp's sensory system remain to be elucidated. Other chemical cues, as primary and secondary metabolites, on oral secretions and frass of *S. nonagrioides*, are currently being identified, and their potential implication on *C. typhae* host adaptation will be discussed.

SE35 C07

REGULATION OF SORGHUM DEFENSES TO INSECT PESTS

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Aphids are important pests of plants that cause substantial loss in plant productivity. The sugarcane aphid (SCA; *Melanaphis sacchari*) is a relatively new and devastating pest of sorghum (*Sorghum bicolor*) in the United States. Previously, we identified varied levels of resistance to SCA in founder lines of the sorghum nested association mapping (NAM) population. Compared to wild-type plants, SCA proliferation was lower on SC265 and higher on SC1345 plants. Monitoring of aphid feeding behavior using electrical penetration graph (EPG), a technique to unveil feeding process of sap-feeding insects, revealed that SC265 plants

restricted SCA feeding from the phloem sap. Our preliminary results also indicate that the constitutive and SCA feeding-induced volatiles were significantly higher in SCA-resistant SC265 plants compared to SCA-susceptible SC1345 plants. Moreover, transcriptomic profiling of sorghum plants reveals that the SCA-resistant SC265 line is better adapted to activate early defense signaling mechanisms in response to SCA feeding. Taken together, our results indicate that SCA feeding, in addition to altering direct defenses can also modulate indirect defenses in sorghum.

SE35 001

POTENTIAL OF SELECTED WILD PLANTS FOR USE IN CONSERVATION BIOLOGICAL CONTROL OF TOMATO INSECT PESTS

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The establishment of selected flower strips adjacent to or within the crops is an increasingly adopted practice to enhance natural enemies' habitats and associated ecosystem services in the agroecosystems. Here, weed vegetation surveys were conducted in two important production areas of Solanaceous crops in Greece to identify wild plant species that would be suitable to support natural enemies of tomato insect pests, with a focus on parasitoids. Initially, the parasitoid presence on potentially suitable wild plant species was recorded by suction samplings. Volatiles were collected from the plants in full blossom using dynamic headspace technique and analyzed by gas-chromatography-mass spectrometry in the laboratory. The response of the egg parasitoid *Trichogramma achaeae* (Hymenoptera: Trichogrammatidae) to odour combinations between different species of flowering plants vs air, was tested by Y-tube olfactometer. The suction samplings showed that specific wild plant species are promising in supporting the conservation of parasitoids of important tomato pests. In particular, the wild plant species *Calendula arvensis* (Vaill.) L., *Melilotus indicus* (L.) All., *Eruca vesicaria* (L.) Cav., *Reichardia picroides* (L.) Roth and *Vicia villosa* Roth, attracted parasitoid species of the families *Braconidae*, *Eulophidae*, *Pteromalidae* and *Trichogrammatidae* (only by *M. indicus*) which could contribute to the control of the Lepidoptera *Helicoverpa armigera* (Hübner) (Noctuidae) and *Tuta absoluta* (Meyrik) (Gelechiidae), and the family Scelionidae including egg parasitoids of pentatomid bugs such as *Halyomorpha halys* (Hemiptera: Pentatomidae). The olfactometer choice tests indicated a preference of *T. achaeae* to certain plant species (e.g., *C. arvensis*). These findings are of great importance in exploiting promising wild plant species when designing functional flowering plant mixtures for agroecological schemes in crop protection.

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Concurrent Sessions

Thursday, 4 July

SE35 002

ABELIA × GRANDIFLORA AS POTENTIAL FUNCTIONAL BANKER PLANT FOR CONSERVING BIOCONTROL IN RICE ECOSYSTEM

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Abelia × grandiflora (Dicotyledon: Dipsacales: Caprifoliaceae), commonly called glossy abelia, is a flowering shrub making perfect road-side hedge, addition to garden borders, beds, and screens in the suitable climates. Its long-flowering duration and faint scent volatile provide the possibility of being a functional banker plant for conserving natural enemies of crop insect pests. We carried out a series of population, behavioral and chemical ecological experiments and indicated that *Abelia × grandiflora* can increase the parasitism of rice brown planthopper eggs by *Anagrus* spp. (Insecta: Hymenoptera: Mymaridae) in open garden experiment, and increased *Anagrus*' parasitizing activities. The main volatile compounds were identified and validated as the main attractive chemicals to the parasitoid. Field experiment showed the *Abelia* could enhance main parasitoids' abundance around the monitoring sticky board. In conclusion, *Abelia × grandiflora* is one the potential functional plant for rice insect pest management through conserving natural enemies.



MC3

18.00-20.30 Concurrent Session 36

Challenges in Sustainable Plant Disease Management in a Changing Climate

Chair: Jay Ram Lamichhane (INRAE, France)

Emilia Markellou (Benaki Phytopathological Institute, Greece)

SE36 C01

INFLUENCE OF CLIMATE CHANGE ON FUSARIUM HEAD BLIGHT AND ASSOCIATED MYCOTOXINS

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Climate change, or long-term shifts in temperatures and weather patterns, can significantly influence the prevalence, incidence and severity of plant diseases. Human-induced greenhouse gas emissions have led to global warming and increased precipitation, which favor the development of certain plant diseases. Among them is Fusarium head blight (FHB), a devastating disease of small grain cereal crops, mainly wheat and barley. FHB is caused by fungal pathogens in the genera *Fusarium* and *Microdochium*, notably *F. graminearum*, *F. culmorum*, *F. avenaceum*, and *M. nivale*. In addition to grain yield and quality reduction due to aborted or Fusarium-damaged kernels (FDK), these pathogens also produce mycotoxins which are harmful to humans and animals. The major mycotoxin detected in FHB-affected grain is deoxynivalenol (DON), but others including zearalenone, HT2, T2 and NX-2 are also detected. FHB is favored by warm temperatures and abundant rainfall before and during anthesis. Simulation models by various researchers indicate that globally, the risk for outbreaks of FHB epidemics will increase due to climate change, and it will be accompanied by an increase in mycotoxin contamination of grain. Results from field experiments showed that FHB index, DON and FDK were much higher and yield was much lower in a wet compared to a dry growing season and in irrigated compared to dryland plots. Rising ocean temperatures due to global warming increase atmospheric water content due to evaporation. The risk for FHB outbreaks is increased due to the resulting intense precipitation over land coupled with warm temperatures. Efforts to curb greenhouse gas emissions and slow the pace of climate change will contribute to sustainable food production and global food security by reducing crop losses caused by plant diseases.

SE36 C02

CAN MATHEMATICAL MODELS SUPPORT GRAPEVINE DISEASE MANAGEMENT UNDER CLIMATE CHANGE?

Georgia Fedele¹, **Vittorio Rossi**¹

1. Research center on Plant Health Modelling (PHeM), Department of Sustainable Crop Production (DiProVeS), Università Cattolica del Sacro Cuore, Via Emilia Parmense 84, 29122, Piacenza, Italy

Climate change and consequent extreme events have a severe impact on viticulture, affecting grapevine growth and development, arthropod pests, and diseases, as well as grape yield and quality. Specifically, since both crops and crop pathogens are affected by weather variables, climate change is

expected to affect plant pathogen epidemiology and the host-pathogen interactions at different rates. A multi-modelling approach, which combines mathematical models for both grapevines and pathogens, would be useful for assessing the potential influence of climate change on disease development during the season by computing future projections of disease outbreaks in the grape-growing areas. Plant disease models can be classified in data or process-based models. Despite data-based models provide useful insight to explore the relationships within a system, they are limited in providing explicit information on the underlying processes and impacts with the environmental driving variables; for this reason, they are not able to predict plant-disease-environment relationships under climate change. Conversely, process-based model, which describe the process based on the underlying functional mechanisms, are useful for evaluating the biological responses as function of environmental drivers. The use of process-based model for Botrytis bunch rot and downy mildew are case-studies that show the benefits of integrating process-based models for plants and pathogens under a variable climate.

SE36 C03

MANAGING SOILBORNE DISEASE COMPLEXES OF ANNUAL FORAGE LEGUMES IN A CHANGING CLIMATE

M. J. Barbetti¹, **M. P. You**¹

1. University of Western Australia

Studies on soilborne diseases of annual forage legumes have generally involved infections by a single oomycete or fungal pathogen (e.g., individual species of *Pythium*, *Phytophthora*, *Aphanomyces*, *Rhizoctonia*, or *Fusarium*). However, the reality is that they actually exist as variable complexes of co-occurring pathogens. Such complexes not only devastate annual forage legume stands but the severe damage to root systems ensures a lack resilience to counter the increasing adverse weather conditions, including dry periods, associated with a changing climate. The unpredictable and ever-changing proportions of different pathogen species within complexes, including individually variable, multifaceted, and contrasting interactions with each other, and the effects of temperature, moisture, type of soil, nutrition and cultivar, pose further challenges to their management. Recent studies highlight how fungicides effective against single pathogens are ineffective in managing soilborne disease complexes. Host resistance against all pathogens within a complex is unavailable, and generally, individual resistances can breakdown readily. Therefore, identifying and deploying effective in-field host tolerance(s) and combining these within an integrated disease management framework to manage these soilborne complexes is required. Current studies emphasise how forage legume cultivars with in-field-tolerance are not only up to four times more productive, but that they also demonstrate enhanced



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resilience against adverse weather conditions. This approach of in-field-tolerance provides the opportunity to achieve major increases in biomass, seed production, stand persistence, and both greater, more stable and consistent forage productivity over variable seasonal conditions exacerbated by a changing climate. Such an approach would not only benefit graziers and forage legume seed producers but also provide farmers with reliable forage stands even in seasons of adverse weather conditions due to a changing climate.

SE36 C04

NEM-EMERGE: TACKLING THE EMERGENCE OF TROPICAL ROOT KNOT NEMATODES IN EUROPE

Daniel Bebber¹

1. Department of Biosciences and Global Systems Institute, University Of Exeter, Exeter, United Kingdom

Soil-borne plant-parasitic nematodes are a biosecurity risk for global food production with an estimated annual loss of €110 billion worldwide. Root-knot nematodes (RKN) and potato cyst nematodes (PCN) rank 1 and 2 in the Top 10 of high-impact plant-parasitic nematodes with RKN alone accounting for ~5% of global crop losses. RKN and PCN are A2 quarantine pests or emerging species listed on the EPPO Alert List. The two PCN species are also included in EU Commission implementing regulation 2021/2285. Recent reports document the emergence of new RKN and PCN problems in tomato and potato cropping across Europe and beyond due to two independent drivers: global warming and genetic selection. For decades, non-specific, environmentally harmful agrochemicals have been applied to manage RKN and PCN. The increasing awareness about their negative impact prompted the phasing out of most nematicides. Consequently, there is an urgent need for novel, durable control strategies that enable adequate responses by stakeholders to prevent crop losses in the EU and beyond. A new Horizon Europe project, NEM-EMERGE, will provide a spectrum of sustainable, science-based solutions for both the conventional and organic farming sector based on the principles of IPM, including (1) optimized crop rotations schemes including cover crops, (2) tailored host plant resistances, and (3) optimal use of the native antagonistic potential of soils. Moreover, monitoring and risk assessment tools will be generated to support Plant Health Authorities in decision and policy making. To ensure the adoption and implementation of NEM-EMERGE tools in the sector, a bottom-up co-creation process and multi-actor approach will be used based on stakeholder demands from both the conventional and organic sector. This makes NEM-EMERGE a key driver for the transition to sustainable farming in line with the Farm to Fork Strategy thereby contributing to the challenging targets set by the Green Deal.

SE36 001

CHALLENGES FACING U.S. COTTON PRODUCTION: A FOCUS ON PATHOGENS AND NEMATODES

Bissonnette K¹

1. Cotton Incorporated, Cary, United States

Cotton production in the U.S. spans from the eastern to the western seaboard representing multiple climates, each favoring different pathogens and nematodes. As environmental

conditions have begun to change in U.S. cotton production regions, so too have the pathogen and nematode threats. These include not only re-emerging and endemic threats, but also newly emerging and invasive threats. This talk will focus on Cotton Incorporated's priorities in pathology and nematology for the U.S. cotton production system. To prioritize threats in the U.S., the cotton producing states are divided into production regions, eastern and western, based on environmental conditions and dominant pathogen and nematode threats. Emphasis will be placed on research advances and Cotton Incorporated's engagement with researchers to address threats relevant to the global cotton community.

SE36 002

TRICHODERMA EAR ROT – EFFECT OF TEMPERATURE AND PRECIPITATION ON A NEW EMERGING DISEASE OF MAIZE IN EUROPE

Pfördt A¹, von Tiedemann A¹

1. Georg-August Universität Göttingen, Goettingen, Germany

Trichoderma afroharzianum is a ubiquitous species occurring worldwide. Due to its mycoparasitic and endophytic properties, certain *Trichoderma* isolates are used in agriculture as biological plant protection and biocontrol agents. However, in 2018, a massive occurrence of *T. afroharzianum* on maize cob was observed for the first time in Germany. Since then, *Trichoderma* ear rot has been observed at several locations in Germany, France, and Italy, especially after dry and hot vegetations periods. *Trichoderma* ear rot are shown as soft rotten infections characterized by excessive moisture and low starch content with massive production of green to gray-green conidia on the kernels and husk leaves. This results in a significant reduction in cob weight and quality with a reduced germination rate. Greenhouse and climate chamber experiments highlighted the preference of *Trichoderma* for hot temperatures and dry conditions, aligning with observations in natural field occurrences. The growth rate of pathogenic *T. afroharzianum* isolates were significantly higher compared to other *Trichoderma* species, with an optimal temperature range between 28°C and 32°C. In vivo climate chamber experiments further confirmed a strong positive correlation between disease severity and increasing temperature. Elevated temperatures, particularly during the months of June, July, and August, corresponding to the period of flowering, are potentially favor a natural infection in the field. The climatic tendencies suggests that its occurrence may become more frequent in the future. In additions, greenhouse and field experiments have shown that *Trichoderma* is capable of infecting other cereals beyond maize like wheat and barley causing discolorations and a reduction of thousand-kernel weight. This expanded host range and continues shifts in temperature and precipitation patterns underscores the potential threat posed by *Trichoderma* infections in Europe.

SE36 003

KEY INFECTION STAGES DEFENDING HEAT STRESS IN HIGH TEMPERATURE RESISTANT *BLUMERIA GRAMINIS* F. SP. *TRITICI*

Zhang M¹, Fan J¹, Liu W¹, Zhou Y¹

1. Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China

With the increase of temperature in the winter wheat-growing regions in China, the high-temperature-resistant *Blumeria graminis* f. sp. *tritici* (Bgt) isolates developed in the fields. To clarify the key infection stages and the roles of heat shock protein (HSP) genes of high-temperature-resistant Bgt isolates defending high temperature, 3 high-temperature-resistant and 3 sensitive Bgt isolates were selected from 55 isolates after determination of temperature sensitivity. And then they were used to investigate the infection stages and the expression levels of HSP genes, including *Bgthsp60*, *Bgthsp70*, *Bgthsp90*, and *Bgthsp104*, at 18°C and 25°C. The formation frequency of abnormal appressoria and inhibition rate of haustoria formation of high-temperature-resistant isolates at 25°C were lower than those of high-temperature-sensitive isolates, while major axis of microcolonies of high-temperature-resistant isolates was higher than those of high-temperature-sensitive isolates at 25°C. The results indicated that haustoria formation and hyphal expansion were the key infection stages of defense against heat stress in high-temperature-resistant isolates. Further analyses of HSP genes found the expression levels of *Bgthsp60* and *Bgthsp70c* were upregulated at 24 and 72 h post-inoculation in high-temperature-resistant isolates, while no significant difference was observed for *Bgthsp90* and *Bgthsp104* genes. Taken together, the basis of high-temperature-resistant Bgt isolates is associated with induced expression of *Bgthsp60* and *Bgthsp70c* response to heat stress in haustoria formation and hyphal expansion stages.

SE36 004

INVESTIGATING COLLETOTRICHUM SPP. FROM GREECE FOR GROWTH UNDER DIFFERENT TEMPERATURES AND THEIR VIRULENCE ON FLOWERS AND FRUITS OF OLIVE CULTIVARS

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1. Agricultural University Of Athens, Athens, Greece

Olive anthracnose has evolved to a destructive disease as a result of climate change, with most damaging symptoms being fruit rot and blossom blight. In the present study, five *Colletotrichum* spp. strains from different regions of Greece were evaluated for their ability to grow under different temperatures and to infect olive flowers and fruits of the Kalamon, Lianolia and Koroneiki variety. The three strains were isolated from Epirus (Preveza), one of them belonging to *C. godetiae* based on sequence analysis of the ITS (internal transcribed spacer) region. These strains were compared to a *C. guajavae* isolate from Messina (Peloponnese) and the *C. acutatum* reference strain (island Zakynthos). Results show that strains from Epirus were more capable to grow on lower temperature (15°C), compared to the intermediately growing *C. guajavae* isolate. On the other hand, the *C. acutatum* isolate showed slow growth at low temperature (15°C) and satisfactory

growth at high temperature (30°C). Regarding infection of flowers, the Koroneiki variety was more susceptible compared to the Kalamon variety against infection by the *C. guajavae* and *C. godetiae* isolates. Among isolates, *C. acutatum* and *C. guajavae* were more virulent infecting olive flowers already at 2 dpi (day post inoculation) compared to *C. godetiae*. In olive fruits, the *C. guajavae* and *C. acutatum* isolates were more aggressive, compared to *C. godetiae*. Both on the Kalamon and the Koroneiki variety. However, in the Lianolia variety, *C. guajavae* and *C. godetiae* were more aggressive to infect the fruit in comparison to *C. acutatum*. In conclusion, the present study gives an insight on *Colletotrichum* spp. strain adaptation to different temperatures and differential aggressiveness on fruit and flowers of three Greek olive cultivars. This knowledge combined with population dynamics and epidemiology, might contribute to develop effective disease management.

SE36 005

GENETIC STRUCTURE AND EXCHANGE OF *BLUMERIA GRAMINIS* F. SP. *TRITICI* POPULATIONS IN CHINA

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Wheat powdery mildew caused by an obligate airborne biotrophic pathogen *Blumeria graminis* f. sp. *tritici* (Bgt), is one of the most important economic diseases in wheat production in China. The genetic diversity and genetic structure of 412 Bgt isolates grouped into twelve populations which collected from 13 wheat-producing regions, covering almost all the major wheat growing areas of China were studied by microsatellite molecular markers (SSR), and the relationship between airflow transmission and possible pathogen dispersal were analyzed by the forward and backward trajectories of air mass transport. The results showed that a total of 370 multilocus genotypes were detected which revealed a high genetic diversity of Bgt populations. There existed a significant positive association between Nei's genetic distance and geographic distance, which demonstrated that the genetic variation increased along with increasing in geographic distance over the studied areas. Yunnan population were clearly separated from other populations; The populations exchanged between Shaanxi and Hubei by crossing the Qinling Mountains and possible asymmetric migration from Gansu at high elevation to Sichuan at low elevation. There are gene flow in Shandong, Hebei and Henan populations and the migration were mainly from Henan to Shandong and Hebei; Furthermore, it was inferred that the source of inoculum in Jiangsu were mainly from Anhui and probably from Shandong Province, and the pathogen of Jiangsu-Zhejiang and Beijing might have the same sources of inoculum. These results were valuable to understand the wheat powdery mildew epidemics and to develop the effective disease management strategies.



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SE36 006

IDENTIFICATION OF WHEAT STRIPE RUST TRANSPORT PATHWAYS AND SOURCE APPORTIONMENT FOR INITIAL RE-COLONIZATION ON WINTER-WHEAT IN SOUTHERN HENAN OF CHINA

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Wheat stripe (yellow) rust caused by *Puccinia striiformis* f. sp. *tritici* (*Pst*), is an important airborne disease worldwide. *Pst* inoculum strength of southern Henan in winter or early spring is vitally important for spring epidemic in most of autumn-sown wheat-growing regions of China annually. We calculated the backward trajectories within a more likely 'Window' for initial infection to occur by grasping the first time of wheat stripe rust onset in each growing season among 2011-2022 (11 growing seasons), based on the epidemiological investigation. The resulted trajectory clusters hinged on prevalent winds indicated that quantitative information of the spatial extent of possible spore-cloud movement apparently differed between receiving heights or re-colonization events. Air masses were detected mainly from northwest where covered core rust sources at all tested heights when rust onset occurred in winter, particularly 10 and 500 m. The similar results were found at 3000 m height when the disease firstly reestablished in early spring, while more air masses originated from east and southeast below 1500 m, scarcely any rust sources. Source contribution suggested that the initial *Pst* inoculum which reestablished disease in southern Henan during winter mainly originated from Gansu overwintering areas of China, whereas mainly from adjacent winter spore production regions (southern Shaanxi and northwestern Hubei) when disease onset in early spring. Slightly lower of *Pst* migration contribution was found on southern Hubei. We supposed that the 'green bridges' in southern Hubei may provide a vital link of *Pst* inoculum between Yunnan-Guizhou and southern Henan. Source-receptor relationship showed that the early onset of rust in southern Henan may cause by abundant overwintering inocula in Gansu overwintering areas where commonly regarded as key source areas and transporting on favorable high-altitude winds. Our results provide a new perspective on tracing *Pst* source and migration contribution forward southern Henan.



BANQUET

08.30-11.00 Concurrent Session **37A**

Recent Advances in Plant Virology

Chair: Xueping Zhou (*Institute Of Plant Protection, Chinese Academy Of Agricultural Sciences, China*)

SE37 C01

KNOCK-OUT OF THE VIRUS REPLICATION-RELATED GENES *UBE1B* AND *CCR4/NOT3* BY CRISPR/CAS9 CONFERS HIGH-EFFICIENCY AND BROAD-SPECTRUM RESISTANCE TO GEMINIVIRUSES

Hao Li¹, Pan Gong¹, Xiongbiao Xu¹, Fangfang Li¹, Xueping Zhou¹

1. Institute of Plant Protection, Chinese Academy of Agricultural Sciences, China

Developing and generating resistant germplasm resources is crucial to preventing and controlling devastating diseases caused by geminiviruses. We employed CRISPR/Cas9 system to confer plant resistance against geminiviruses through two strategies. One strategy is to target and edit the viral genome sequences (iterons). On the contrary, similar to the naturally resistant plant germplasm variants that might avoid the penalty of defense, the generation and application of transgene-free plants through CRISPR/Cas-mediated host susceptibility gene editing for antiviral breeding would be promising. The two novel susceptible genes *NbUbe1B* and *NbCCR4/NOT3* were first identified, and knock-out displayed high-efficiency and broad-spectrum resistance to multiple geminiviruses. Notably, deficiency of *NbUbe1B* or *NbCCR4/NOT3* did not display any observable growth-defense trade-off. Therefore, editing these recessive resistance genes would have a promising perspective in future engineering resistant plants against geminiviruses under field conditions.

SE37 C02

UNRAVELING THE PLANT MANIPULATION BY GEMINIVIRUSES

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Viruses, as obligate intracellular parasites, need to subvert the host cell in order to enable their replication and efficient spread. Due to strict restrictions in genome size, viruses commonly produce a limited number of proteins; this is the case of geminiviruses, plant viruses with circular single-stranded (ss)DNA genomes that are believed to contain only 4-8 translated open reading frames. Strikingly, despite their limited armoury, geminiviruses are able to establish an infection, overcoming plant defence, dramatically altering plant development and physiology, and ultimately causing devastating diseases to crops worldwide. How these viruses successfully invade and manipulate their plant hosts by deploying only a handful of proteins is a long-standing, fascinating question. In our group, we are interested in understanding how geminiviruses co-opt the plant cell and lead to disease, for which we use a combination of approaches, including molecular biology, cell biology, and

genetics. Our results have shed light onto the molecular mechanisms underlying the replication of viral DNA, plant anti-viral defence and geminiviral counter-defence, and symptom development, and hint at novel virulence strategies potentially employed by geminiviruses to maximize their coding capacity and their impact on the host cell. We expect that our work will contribute to a deeper understanding of the infection process, which may in turn pave the way to the design of effective and sustainable anti-viral strategies and assist breeding programs to obtain virus-resistant plants.

SE37 C03

PROTEIN, LIPID, AND MEMBRANE INTERACTIONS IN POSITIVE-STRAND RNA VIRUS GENOME REPLICATION

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Positive-strand RNA viruses reorganize host intracellular membranes to assemble their viral replication complexes (VRCs) at specific organelles. However, mechanisms by which viral replication proteins recognize specific organelle membranes, modulate host lipid metabolism to accommodate such membrane proliferation and rearrangements, and recruit host proteins to VRCs are unclear. We use an engineered brome mosaic virus (BMV)-yeast system to examine how BMV regulates phospholipid synthesis during viral replication. The aims of this project are to reveal mechanisms whereby BMV replication protein interact and recruit phospholipid synthesis enzyme to VRCs to synthesize a pool of lipids specific to VRC formation. We first used lipidomics to analyze the compositions of major phospholipids in the absence or presence of BMV replication; we next used antibodies in immunofluorescence microscopy to specifically recognize and localize phosphatidylcholine (PC) and its synthesis enzyme, Cho2p; tested BMV replication by northern blotting using viral RNA specific probes and checked its VRCs by electron microscope in the mutant lacking the CHO2 gene. A significantly increased levels of total phospholipids and particularly PC were found in cells replicating BMV compared to mock cells. The increased PC was primarily colocalized at the perinuclear ER membrane with BMV replication protein 1a, whose expression in yeast cells induced the formation of VRCs. We further showed that BMV 1a interacted with Cho2p and redistributed Cho2p from ER membranes to the perinuclear ER membrane, suggesting the synthesis of PC at VRCs. Deletion of the CHO2 gene inhibited BMV replication substantially, indicating a critical role of Cho2 and PC synthesis in BMV replication. The VRC-enriched PC was also found in hepatitis C virus and poliovirus, suggesting a conserved feature among positive-strand RNA viruses.



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Friday, 5 July

SE37 C04

OCCURRENCE, PREVENTION AND CONTROL OF PEPPER VIRUS DISEASES IN CHINA

Fei Yan, Jiejun Peng, Jian Wu, Yuwen Lu

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China is the world's largest pepper producer. From 2000 to 2021, more than 30% of the world's peppers were grown in China. In 2021, China's pepper planting area reached 12.405 million acres, accounting for approximately 36.72% of the global planting area. Viral diseases are one of the most important diseases on peppers. There are more than 20 kinds of pepper viruses that have been reported, among which PMMoV is an epidemic virus whose occurrence area has gradually increased in recent years. The virus is stable in nature and can persist and remain viable in both water and soil environment. The virus has also been employed as a valuable indicator for tracking fecal pollution in such environments. How to block virus infection and effectively prevent and control the occurrence of diseases is the key to pepper Problems that need to be solved urgently in production. Our research found that the virus-infected plant disease sap can still infect plants from the roots through the soil after dilution, which suggests a way of viral infection and may be the cause of the virus epidemic. Furthermore, our in-depth research found that PMMoV can infect and traffic within *N. benthamiana* plants in non-virion forms. On this basis, we combined a microbial combination. We use soil microorganisms to degrade soil organic matter on the one hand and cut off the source of viruses. On the other hand, we regulate soil microecology to promote plant growth and enhance plant immunity. The application has effectively reduced the incidence of PMMoV and other viruses in the field.

SE37 C05

STRENGTH IN DIVISION: THE COSTS AND BENEFITS OF A MULTIPARTITE GENOME ORGANIZATION IN PLANT VIRUSES

Mark P. Zwart¹, Marcelle L. Johnson^{1,2}, Dieke Boezen^{1,2}, Dimitris Karapliafis³, Rosalinde M. Keijzer¹, Alexey A. Grum-Grzhimaylo^{1,4}, Anne Kupczok³, J. Arjan G.M. de Visser⁵, Carolyn M. Malmstrom⁶, René A.A. van der Vlugt²

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Many viral genomes consist of multiple segments, with some viruses packaging these segments individually into virus particles. These multipartite viruses are anticipated to spread inefficiently due to the potential loss of essential genome segments during transmission. Surprisingly, they

are prevalent among plant viruses, prompting questions about associated benefits. We investigated the ecology and evolution of multipartite viruses using experimental evolution to analyze virus adaptation patterns and metagenomics to probe their occurrence in natural ecosystems. In laboratory experiments, we observed high variability in the genome formula – the relative frequency of viral genome segments – of cucumber mosaic virus (CMV). Despite this variation, the mean genome formula did vary across plant host species. Large shifts in the genome formula correlated with reduced virus accumulation and could even lead to virus extinctions during evolution experiments. Contrary to expectations, our experimental work therefore revealed additional costs associated with multipartite organization. These results reinforce the conclusion that within-host selection may drive the emergence of multipartite viruses, rather than any obvious benefits at higher levels of selection. In field studies, we analyzed the viromes of wild plants in re-wilded former agricultural land. While we detected numerous multipartite plant viruses, their prevalence patterns did not differ from viruses with other genome organizations. However, we noted systematic differences in the genome formula of two prevalent multipartite viruses across field sites, independent of host-plant species or virus phylogeny, indicating a dependency on the successional stage of communities. Unlike experiments under controlled conditions, our field observations suggest that the genome formula may indeed play a role in the adaptation of multipartite viruses in real-world systems.

SE37 C06

DSRNA-MEDIATED RESISTANCE AGAINST PLANT VIRUSES

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Plant viruses cause nearly half of the emerging plant diseases worldwide, contributing to 10-15% of global crop yield losses. At present, the management of plant viral diseases relies mainly on extensive use of chemicals targeting the vectors (i.e., insects, nematodes, fungi) transmitting these viruses. However, the application of these chemicals has negative consequences on human health and the environment. Aiming at the reduction of usage of pesticides in agriculture (main aim of the European Green Deal) the genetic resistance will have to play an important role in sustainable agriculture. The method of pathogen-derived resistance (PDR) in plants seems to be an excellent means to induce antiviral resistance taking advantage of the virus' genetic information. RNA interference (RNAi) is an endogenous, cellular, sequence-specific RNA degradation mechanism present in almost all eukaryotes, induced by double-stranded RNA (dsRNA) molecules, and has been exploited as an antiviral strategy through transgenesis. Since genetically modified crop plants are not accepted for cultivation in several countries of the world, including the European Union, the top priority is to embrace alternative non-transgenic strategies for crop protection. This demand has boosted research on exogenous application of the RNA-based biopesticides that are shown to exhibit significant protective effect against viral infections. In addition, there are notable advances in the use of carrier molecules to improve



longevity and efficiency of the dsRNA molecules applied for the induction of plant pathogen resistance. The dsRNA-mediated approach along with the gene editing and amiRNA-mediated approach are environment-friendly and efficacious antiviral strategies for crop protection to contribute to global food security, without undesirable effects on human health.

SE37 001

INVESTIGATING PLANT PERSISTENT VIRUSES IN CHILLI PEPPER

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Plant viruses have been mostly studied for their role as pathogens. However, research on plant-viral interactions in populations of wild plants has shown that some viruses have non-pathogenic effects on their hosts. Plant persistent viruses (PVs) are vertically transmitted, i.e., inherited through pollen and seed, and are not infectious. PVs cause little or no disease symptoms in their hosts, and it has been hypothesised that these RNA viruses are mutualists rather than pathogens. Building on these ideas, PV-mediated effects on host phenotypes are being investigated in pepper (*Capsicum annuum*). Using RT-PCR, genomic RNAs of *Partitiviridae* and *Endornaviridae* were detected in eight accessions of pepper. PV RNAs were observed in all tissues of PV-harboured lines. Virus-virus interactions between PVs and pathogenic viruses were investigated in pepper using tobacco mosaic virus (TMV) and cucumber mosaic virus (CMV). Preliminary experiments show that TMV and CMV RNA levels were lower in PV-harboured pepper varieties compared to PV-free varieties, possibly due to interaction between PVs and pathogenic viruses. To improve future analyses, virus-induced gene silencing using tobacco rattle virus vectors has been used successfully to generate isogenic PV-harboured and PV-free pepper plant lines. The effects of PVs on a broad range of host traits, from seed yield, resistance to aphids and aphid-vectored pathogens, to drought tolerance are being examined using these isogenic lines.

SKALKOTAS

08.30-11.00 Concurrent Session **38**

Precision Agriculture and Remote Sensing for Future Plant Disease Management

Chair: Spyros Fountas (Agricultural University of Athens, Greece)

SE38 C01

AI-DRIVEN PRESCRIPTION SPRAYING MAPS: PIONEERING HERBICIDE REDUCTION IN PRECISION AGRICULTURE

Spyros Fountas¹

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This study presents an AI-driven framework for the generation of Prescription Spraying Maps (PSMs) aimed at optimizing plant protection products usage in precision agriculture. By integrating artificial intelligence (AI) with real-time and historical agricultural data, the proposed system enables precise herbicide application, potentially reducing overall usage and environmental impact. The methodology employs artificial intelligence algorithms to analyze multi-source data, including soil conditions, meteorological data, and remote sensing imagery. These AI models predict weed proliferation zones, allowing for targeted plant protection products application (Smith et al., 2020). The approach is underpinned by the development of digital twin technology, offering a virtual representation of physical fields to simulate and optimize spraying strategies (Jones & Williams, 2019). A key component of the system is the adaptation of autonomous ground vehicles equipped with direct injection spraying technology, ensuring precise herbicide delivery (Doe et al., 2018). The integration of these technologies aims to refine herbicide application, aligning with sustainable farming practices and regulatory frameworks aimed at reducing agrochemical reliance (EU Green Deal, 2021). Preliminary results indicate a potential reduction in herbicide use by up to 30%, with corresponding decreases in environmental contamination risks. The study suggests that AI-driven PSMs could represent a scalable solution for precision agriculture, promoting efficient resource use and supporting ecological balance in farming ecosystems.

to a revolutionary era in agriculture. UAVs offer novel opportunities to improve agricultural practices by providing precision, efficiency, and safety in chemical application. This study presents a comprehensive investigation into the application of UAV spraying technology to improve spray coverage and deposition in vineyards. Field trials were conducted using a commercial hexacopter spraying UAV with water-sensitive papers serving as the primary tool for assessing coverage and deposition. Analysis of these water-sensitive papers was performed using DepositScan software, providing valuable insight into the effectiveness of different application techniques and parameters. Moreover, spraying droplet displacement, commonly referred to as spraying drift, was measured using a standardized experimental protocol, namely ISO-22866. The results of the study showed that inter-row application methods outperformed over-row applications and demonstrated superior canopy coverage. In addition, the study showed that a 2.5-meter flight altitude outperformed a 2-meter flight altitude, resulting in a better coverage rate. A flight speed of 1 m/s proved to be more efficient than 1.5 meters per second. Notably, specific treatments, such as Treatment A for over-row applications and Treatment E for inter-row applications, showed promising results due to their common operational parameters - a flight altitude of 2.5 meters and a flight speed of 1 m/s. This research underscores the transformative potential of UAV spraying in modern agriculture, demonstrating its ability to increase efficiency and reduce environmental impact, thus promoting environmentally sustainable crop protection practices. Moreover, our findings underscore the importance of optimizing the operational parameters of UAV spraying, which is also one of the most important open problems in conventional crop protection, to further advance agricultural practices.

SE38 C02

OPTIMIZING CROP PROTECTION: A COMPREHENSIVE STUDY OF SPRAYING DRONES IN VINEYARDS

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In modern agriculture, the precise and efficient application of agrochemicals is essential to ensure crop health and increase productivity while minimizing adverse environmental impacts. While traditional spraying methods have long been the cornerstone of crop protection, the introduction of unmanned aerial vehicles (UAVs), or drones, has led

SE38 C03

CAN SPRAY APPLICATION TECHNOLOGICAL IMPROVEMENTS CONTRIBUTE TO PESTICIDE USE AND RISK-RELATED REDUCTION?

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The EU Farm to Fork Strategy strives to reduce by 50% the overall use and risk of agrochemicals within 2030. Two possible ways to comply with the stringent requirement are i) to increase the spray application efficiency and ii) to reduce the amount of plant protection product (PPP) applied by adapting the spray application rate to the canopy

characteristics. In this context, any pesticide application equipment (PAE) technological improvements and the use of decision support systems (DSS), aimed to properly adjust the PAE, are key factors in reducing both pesticide use and related risk. This work presents the main results derived from the EU Life PERFECT, EU H2020 OPTIMA, and NOVIAGRI projects. A common experimental approach based on the measurements of a) canopy spray deposit, b) canopy spray coverage, c) infield ground losses and d) spray drift (both ground sediment and airborne) was used in the three projects to quantify the possible PPP use reduction as well as PPP use related risk while ensuring the biological efficacy of treatments in vineyards. Within the PERFECT project a DSS was developed and tested through field trials to demonstrate that according to the different vineyard training systems by the proper selection of spray application rate according to the canopy characteristic it was possible to reduce the PPP use between 20 and 45 %. Within the OPTIMA project an innovative smart sprayer able to automatically and continuously vary the airflow rate according to the canopy characteristics density, was developed and tested. The sprayer demonstrated a significant increment of spray efficiency by increasing the canopy deposit between 25 and 40% while concurrently reduce the spray drift up to 74% at 30 m downwind distance. Finally, within the NOVIAGRI project an innovative sprayer able to provide variable spray application rate according to the canopy characteristics was developed and tested showing a direct pesticide saving comprised between 43 and 73% and concurrently reduced the airborne spray losses up to 90%.

SE38 C04

DIGITAL TECHNOLOGIES FOR PLANT HEALTH, EARLY DETECTION, TERRITORY SURVEILLANCE AND PHYTOSANITARY MEASURES?

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The EU funded project STELLA (<https://stella-pss.eu/>) is developing a holistic digital real time pest surveillance system (STELLA PSS) to aid in the early warning and detection of quarantine and regulated plant pathogens, together with a response strategy that uses modern sensing technology and Artificial Intelligence. The system consists of 3 subsystems: 1) an early warning system using novel forecasting models and IoT sensors, 2) a pest detection system using drones, satellites, and a smartphone application, and 3) a pest response system providing data-driven recommendations for containment and counteractive measures. The system will be tested over three years on field, farm, and regional levels across 6 Use Case Pilots and will focus on 8 different quarantine and regulated non-quarantine pest (RNQP) diseases, expanding across 4 European countries with different climate and geological characteristics and New Zealand. Capacity building activities will be developed with a focus on providing training, education, and resources to farmers, agronomists, and other stakeholders involved in crop protection. These activities will aim to equip stakeholders with the skills and knowledge necessary to effectively use the

STELLA system and implement environmentally friendly crop protection alternatives. Policy recommendations generated by the project will aim to support the European Commission's goals of reducing pesticide use and managing priority plant pest outbreaks. The recommendations will be based on the data and insights gathered through the early warning, detection, and response system. A networking strategy will be developed to leverage existing knowledge and enable links with relevant organisations, networks, projects, and initiatives. Collaborating with stakeholders as well as other projects will provide opportunities to exchange ideas and learn from others' experiences.

SE38 001

SYNTHETIC DATA GENERATION FOR ENHANCED WEED AND CROP SPECIES DETECTION?

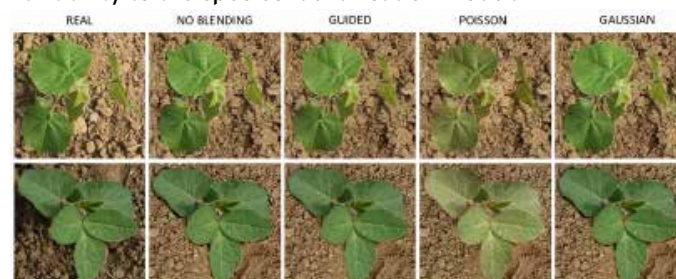
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Precision weed management has emerged as a promising solution for targeted herbicide application and thus, environmental impact reduction. While deep learning-based methods provide valuable solutions for weed detection, they often rely on large volume of annotated data, a resourceintensive and time-consuming task. We propose an approach that reduces human intervention during algorithm training by generating synthetic images from existing annotated data. Our method involves segmenting and extracting objects from real plant images, followed by pasting these objects on clean ground backgrounds with different advanced blending techniques:

- Guided image filtering: We create a 3-channel mask using guided filters to indicate transparency levels. This preserves border details and ensures smoother transitions.
- Poisson local illumination: Seamlessly integrates object and background colours.
- Gaussian blur: This technique smooths sharp transitions along plant edges.

These techniques are applied to generate synthetic images using 360 real images of 13 varied species, including crops and weeds. These synthetic images will provide valuable variability to the species identification model.



Synthetic images realism is measured by comparing image embedding representations between real and synthetic images extracted by a Dinov2 model. To validate the use of synthetic images and evaluate the performance of the different blending methods, species identification models

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are trained first only with real annotated images, and then, by combining real and synthetic images. Subsequently, the models are evaluated with 366 real annotated images from a different collection. This process is repeated across 10 different train subsets, and accuracy and F1-score metrics are reported. The inclusion of synthetic images allows more accurate identification of weed and crop species.

	REAL	REAL + NO BLENDING	REAL + GUIDED	REAL + POISSON	REAL + GAUSSIAN
Accuracy	85.25	88.8	89.78	89.26	89.62
F-1 Score	84.75	88.48	89.50	88.76	89.40

SE38 002

PRECISION WEED MANAGEMENT AND CROP COVER ANALYSIS AND WEED IDENTIFICATION IN AGRICULTURE: USING CNNs FOR ENHANCED PRACTISES

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In response to mounting global concerns over food security and stringent herbicide regulations, innovative approaches to weed management are crucial. In the context of limiting the use of chemical herbicides to protect the environment, as defined by the Green Deal, cover crops would be used between planting lines to limit weed growth. In recent years, digital cameras and Artificial Neural Networks (ANNs) have undergone rapid development, offering innovative techniques and tools for applications in agriculture and weed management. A multitude of methods have emerged, yet there remains a need for continued improvement in addressing challenges related to the speed, robustness and accuracy of algorithms and recognition systems. This research aims to investigate the effectiveness of three common Convolutional Neural Networks (CNNs), specifically VGG16, ResNet-50, and Xception, to analyse plant coverage, cover crop mix ratio and weed coverage ratio among various cover crop combinations. It is being investigated whether these models can be used to assess the effectiveness of cover crops or to calculate the spread of weeds in a field. Utilizing a 12 MP camera at a 3-meter distance from the ground (0.60 mm² / pixel), RGB images were captured for analysis. The images in this study encompassed 5 cover crop mixtures, at different plant growth stages, as follows: *Lathyrus sativus*, *Hordeum vulgare*, mixture of *Hordeum vulgare* with *Vicia sativa*, *Trifolium subterraneum* and *Pisum sativum*. Additionally, control groups were incorporated, totalling 12 crops combinations. For each CNN model and crop combination, 70% of the images were utilized for network training, 15% for validation during each training, and the remaining 15% for testing. The CNNs models achieved high accuracy in identifying the cover crops, and analysing the appropriate cover percentages, providing valuable insights for informed decision-making in agricultural management practices. Further research and implementation of this methodology hold promise for addressing challenges related to food security on a global scale.

SE38 003

AIR-SEQ: A NEW TOOL FOR DETECTING AIRBORNE PATHOGENS

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Airborne crop pathogens pose a serious threat to food security through devastating loss of yield. Early detection of pathogens can provide valuable information to allow targeted fungicide application or the use of alternative pathogen management strategies. However, existing detection regimes typically rely on analysis post-damage, may be specific to a single pathogen and can't distinguish different pathotypes. Here, we present Air-seq, a rapid sequencing-based method that is unbiased, can detect multiple pathogens simultaneously and can detect them before evidence of disease is present on plants. The Air-seq method combines collection of airborne particles (e.g. fungal spores, plant pollen, bacteria) using commercial air collectors with a bespoke DNA extraction, purification, sequencing and analysis pipeline. Air-seq has been trialled in a range of open and closed environment conditions. In a recent experiment air samples were collected in two greenhouses and one field growing strawberries. Sequencing and analysis with the Air-seq pipeline allowed comparison of airborne pathogen levels to manual disease scoring and fungicide application information. This showed that Air-seq could detect high levels of pathogens before symptoms were visible on the plants. Air-seq is also being applied to understand how the dynamic air microbiome on a cereal farm varies over the course of a 24-hour period and for pathogen monitoring in wheat fields over a whole growing season. Air-seq has demonstrated potential as a useful tool in a plant protection context. In future, it may be possible to use Air-seq to detect fungicide resistance genes in pathogens, or to promote awareness of new invasive species before environmental conditions are conducive to infection. Additionally, airborne pollen from problematic plants could be detected with Air-seq enhancing monitoring across large areas with minimal effort. These applications mark Air-seq as a valuable tool in plant protection strategies.

SE38 004

EARLY DETECTION OF VERTICILLIUM WILT OF OLIVE: ANALYZING THE INTACT LEAF SPECTRA FROM THE VISIBLE-NEAR INFRARED RANGE IN THE FIELD

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One of the key challenges facing global olive cultivation is the Verticillium wilt of olive (VWO), caused by the pathogenic fungus *Verticillium dahliae* (Vd) Kleb. Most olive cultivars are susceptible, while tolerant varieties are often unsuitable for mechanized olive hedgerow systems. Early detection approaches guarantee effective management; however, it is quite difficult to visually detect symptoms during the early stages of disease development. Our aim in this study was to develop a non-invasive method for early Vd detection using leaf spectroscopy and machine learning (ML) algorithms. Thereunto, forty-five 7-year-old 'Picual' trees were used in the trial conducted in Cordoba, Spain, in 2022, one year after drip-inoculation with the defoliating pathotype of Vd. During the trial, pre-inoculated trees were drip-irrigated for 100% crop evapotranspiration while control trees were subjected to three water regimes (rainfed, 50%, and 100% crop evapotranspiration) to contrast biotic and abiotic stress. Each treatment was surface or subsurface irrigated in equal numbers. Pre-inoculated trees were categorized based on VWO symptoms, and leaf spectral data were collected using a field spectrometer every 2-3 weeks from 5th July to 10th October. The ML algorithms included support vector machine, artificial neural network, multi-layer perceptron classifier, and gaussian process classifier. Our ML models accurately discriminated between inoculated and non-inoculated trees, achieving overall accuracy (OA) of 70-95% with a Kappa coefficient (κ) of 0.43-0.85. The ML models increased their OA by adding vegetation indices, compared to those based on exclusively spectral band (700-2100nm). Specifically, the gaussian algorithm demonstrated superior performance, achieving an OA of 79% ($\kappa=0.57$) for reflectance spectra and 95% ($\kappa=0.85$) for vegetation indices. This novel method allows the early detection of VWO in asymptomatic cases, facilitating control measures even under water stress conditions and providing an efficient tool for early disease detection, avoiding the processing of olive tissue samples.

SE38 005

TESTING INNOVATIVE SPRAYING SYSTEMS FOR MITIGATING THE IMPACTS OF LOBESIA BOTRANA (DENIS & SCHIFF.)

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The effective control of the grape berry moth *Lobesia botrana* (Denis & Schiffermueller) (Lepidoptera: Tortricidae), requires multiple treatments commonly applied using traditional air sprayers. The issue of insecticide drift, combined with the application rate not being tailored to the canopy size of traditional air sprayers, underscores the need for research into more efficient and environmentally friendly spraying techniques. In this work, we tested the efficacy of two innovative spraying systems (namely, tunnel sprayer with recovery device and LiDAR-based sprayer) in reducing the impacts caused by *L. botrana* larvae. The study was conducted in a 9-ha vineyard in central-eastern Italy (Marche region), comparing three treatments (traditional sprayer, tunnel sprayer with recirculation, and LiDAR-based sprayer) with an

untreated control. Each treatment had eight replicates, with two rows of vines per replicate. Twenty-four clusters were inspected per replicate. We assessed *L. botrana* infestation at the bunch level (presence of at least one larval penetration hole or live 2nd generation larvae) and we measured the average number of infested berries per bunch. Statistical analysis was performed using chi-squared and Kruskal-Wallis tests with Bonferroni correction for multiple comparisons. The recovery system performed best in reducing both the number of infested bunches and the mean number of berries per bunch, followed by LiDAR, traditional, and control. Both LiDAR and tunnel sprayers significantly reduced the impacts of *L. botrana* larvae compared to the control. We demonstrated the efficacy of the tunnel sprayer and the LiDAR-based sprayer in reducing the impacts caused by *L. botrana* larvae. These innovative spraying systems might greatly reduce the environmental risks posed by insecticides through reducing spray drift, and optimizing the volumes applied based on canopy surface.

The work was carried out within the framework of the MISE-AGRIFOOD project (Project N. F/250006/02/X50)

SE38 006

HYPERSPECTRAL DETECTION OF SINGULAR/ INTERACTIVE EFFECTS OF SIMULATED TREE SHADING AND ALTERNARIA ALTERNATA INFECTION ON SORGHUM BICOLOR UNDER FIELD CONDITIONS

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The use of agroforestry has been extensively promoted worldwide, but these practices have often been applied without a clear understanding of their effects on plant diseases. Advancements in techniques to rapidly and non-destructively detect and monitor the impact(s) of agroforestry systems on crop health are thus recommended. This study examined the capability of full-range (350-2500 nm) reflectance spectroscopy to characterize the effects of simulated tree shading and *Alternaria alternata* infection on *Sorghum bicolor* under field conditions. By a plot trial, the cv. PR89Y79 was grown under full light (no shade) or a potential light availability reduced by 50% (shade), and then half of the plots (equally distributed between shaded and not shaded) were fully sprayed with *A. alternata* suspensions (strains A214 and A216) with final concentrations of 0 or 2×10^8 CFU mL⁻¹. After one month from inoculation, hyperspectral data were collected at both leaf and canopy levels (1 m above the canopy, nadir). Analyzing leaf spectral signatures (400-2400 nm) by permutational multivariate analysis of variance (PERMANOVA), a highly significant ($P < 0.001$) 'shade' effect was reported, and moderate significances ($P < 0.05$) were also found for 'Alternaria' and 'shade x Alternaria' ones. Differently, only the singular 'shade' and 'Alternaria' effects resulted significant on canopy hyperspectral profiles. According to PERMANOVA, partial least discriminant



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analysis showed an overall accuracy for the discrimination of the experimental theses ranging between 65 and 92%. Variations of spectral vegetation indices and leaf traits derived from spectra by developed partial least squares regression-models highlighted the influence of shading on *A. alternata* infection of sorghum at both physiochemical level. Overall, the present study highlights the potential of using hyperspectral data to improve the feasibility of assessing crop disturbances as a result of abiotic and/or biotic stress in a timely and cost-effective manner.

SE38 008

MACHINE LEARNING TECHNIQUES USING AIRBORNE MULTISPECTRAL DATA FOR DETECTION OF INFESTATION DETECTION BY XYLOTRECHUS CHINENSIS (CHEVROLAT) (COLEOPTERA: CERAMBYCIDAE) IN MULBERRIES

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The tiger longicorn beetle, *Xylotrechus chinensis* Chevrolat (Coleoptera: Cerambycidae), is an emerging enemy for the mulberries in Greece since its invasion in 2017. The detection of the pest is possible either by adults' emergence holes left on the bark, or by the appearance of the dried branches, when the insect has already done great harm on the tree. Remote sensing provides valuable information on the condition of vegetation health, which, in combination with machine learning techniques, can provide us with the ability to detect infestations by pests at an early stage before causing major damages on the plants. The objective of this study was the detection of infestation by *Xylotrechus chinensis* in mulberries through UAV remote sensing multispectral data, using six classification models. Mean NDVI, mean NDRE, mean EVI and tree crown area were calculated and used in machine learning models, along with data for adult emergence holes and temperature data. On-site observations by an entomologist were also taken place, classifying the trees in two different classes, depending on the presence of infestation by *X. chinensis* or not. Random Forest, Decision Trees, Gradient Boosting, Multi-layer Perceptron, K-Nearest Neighbors, and Naïve Bayes were the evaluated models. Random Forest emerged as the most effective predictive model, having the highest score in accuracy, precision, and recall. Further details about the results and extensive discussion are given in the presentation.



MC2

08.30-11.00 Concurrent Session **39**

Prevention and Sustainable Management of Transboundary Plant Pests

Chair: Fazil Dusunceli (Food and Agriculture Organization, Turkey)

SE39 C01

THE AFRICA PHYTOSANITARY PROGRAM: AN IPPC EXPERIENCE IN IMPLEMENTING STANDARDS TO MITIGATE THE SPREAD OF PLANT PESTS OF REGULATORY SIGNIFICANCE

Osama El-Lissy

International Plant Protection Convention (IPPC), Rome, Italy

Invasive plant pests can enter a country that lacks well-functioning national phytosanitary system without being detected. Once introduced, these pests cause significant damage to plant production, including food crops; negatively impacting the environment; and hindering regional and international trade of agriculture products. According to documented reports, globally, annual crop losses to plant pests are estimated to be between 20 to 40 percent of production. In Africa, crop yield losses due to pests is even higher, estimated between 30 to 60 percent. While plant pests continue to cause significant losses in food crops, the number of people affected by food shortages in the world continues to surge. According to FAO, the number of people affected by hunger increased from 811 million in 2020 to 828 million in 2021. In terms of economic impact, plant diseases alone cost the global economy around USD220 billion annually. The primary mission of IPPC is to protect plant resources from invasive pests and diseases and to facilitate safe trade of agricultural products. The IPPC fulfills its mission by working with its 185 contracting parties to develop and implement International Standards for Phytosanitary Measures (ISPMs). ISPMs provide the necessary foundational framework for phytosanitary management, ranging from pest risk assessment to risk mitigation. Based on the above context, the IPPC worked with cooperators at the national, regional, and global levels to develop and implement the first ever global phytosanitary program. The program is designed to enable the national competent authority and stakeholders to timely detect plant pests of regulatory, economic, and environmental significance. The program positions national governments and stakeholders to prepare for, respond to, and recover from plant pests and diseases in the most effective manner. Given the amplified challenge and lack of consistent phytosanitary system, Africa was chosen to be the region where the program begins, hence the Africa Phytosanitary Program or APP. This report provides an update on APP implementation strategy.

SE39 C02

REGIONAL APPROACH TO MANAGING WHEAT RUST DISEASES IN CENTRAL ASIA AND CAUCASUS THROUGH THE FAO-TURKIYE PARTNERSHIP PROGRAMME

Fazil Dusunceli^{1*}, Kumarse Nazari², Beyhan Akin³, Mesut Keser², Abdelfattah A. Dababat³, Veyssel Donderalp¹, Suat Kaymak⁴, Ayse Uysal Morca⁴, Gonca Savas¹, Rovshen Saparmammedov¹, Sebnem Gurbuz¹

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Wheat is a key crop for global food security and any threat to its productivity is a major concern particularly for the developing countries. It is an important commodity for national economies and rural livelihoods in Central Asia and Caucasus where its productivity is often challenged by the rust diseases (yellow, leaf, and stem rust), especially in years with high precipitation. These diseases are caused by the fungi belonging to the species of *Puccinia* spp. Their spores are airborne and transboundary moving across borders easily affecting multiple countries and regions. Their management requires integrated approaches including timely surveillance, monitoring of emerging races, speedy information sharing, contingency plans and rapid response. This initiative aims to strengthening of international collaboration and national capacities in prevention and management of wheat rust diseases at regional level in Central Asia and Caucasus. In this respect, an integrated project has been developed through the support of the FAO- Türkiye Partnership Programme. The five-year project is being implemented in collaboration with ICARDA, CIMMYT, and the RCRRC operated by ICARDA and Ministry of Agriculture and Forestry of Türkiye, as well as national institutions of Azerbaijan, Kazakhstan, Kyrgyzstan, Uzbekistan, Tajikistan, and Turkmenistan. Since 2020, The project has been supporting surveillance of the diseases in the countries, screening of wheat germplasm for resistance, national and regional trainings, contingency plan development, and regional collaborations and information sharing. As part of the research activities, a total of 433 wheat rust samples have been collected for race analysis, and 1150 breeding materials have been screened for resistance. Capacity building activities included training of over 173 technical personnel through the national trainings in Azerbaijan, Kyrgyzstan and Turkmenistan and regional trainings conducted by ICARDA at the regional Cereal Rust Research Center in Izmir, Türkiye.



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APPROACHES FOR RAPID RESPONSE TO CONTAIN TRANSBOUNDARY PEST OUTBREAKS: THE CASE FOR BANANA BUNCHY TOP DISEASE IN AFRICA

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Transboundary pests adaptable to new environments spread swiftly, disrupting agriculture and livelihoods. National plant quarantine and biosecurity regulations protect against their introduction via human and natural means, including seed and plant product transportation. These measures entail surveillance, early warnings, contingency measures, and rapid responses tailored to high-risk pests identified through pest risk assessments. However, these endeavors require substantial resources and constant adaptation. Many low and middle-income countries, particularly in sub-Saharan Africa, lack effective quarantine measures, leaving them vulnerable to invasive pests. Devastating epidemics caused by the Fall Armyworm, Banana Bunchy Top, Banana Fusarium Wilt Tropical Race 4, Maize Lethal Necrosis, and Papaya Mealybug, among others, exemplify this vulnerability. This presentation highlights the lessons from recent epidemics triggered by invasive pests, specifically focusing on the Banana Bunchy Top Disease (BBTD) case in Africa, stressing the urgency of strengthening biosecurity measures against invasive pests. BBTD caused by the banana bunchy top virus (BBTV), transmitted by the aphid vector *Pentalonia nigronervosa*, was first recorded in the 1920s in Egypt and later in the 1960s in the Democratic Republic of Congo. The virus spread was recorded so far in 16 countries. Notably, the virus has expanded its reach to 10 countries since 2008, highlighting the urgent need for enhanced biosecurity measures and collaborative efforts to mitigate BBTV. In this presentation, we will highlight the factors contributing to the uncontrolled spread of the virus based on the surveys and genomic surveillance data, the importance of international partnerships (ALLIANCE model), the role of data-driven tools, including remote sensing and artificial intelligence, rapid diagnostics, and integrated tactics to contain the virus in Africa.

SE39 C04

KEY ROLE OF MONITORING AND EARLY WARNING IN PREVENTION OF TRANSBOUNDARY PLANT PESTS: EXPERIENCES IN LOCUST MONITORING SYSTEMS

Alexandre V. Latchininsky

Senior Locust Management Expert, Plant Production and Protection Division, Food and Agriculture Organization of the United Nations (FAO)

The foundation of the preventive strategy for transboundary plant pests is monitoring and early warning. Food and Agriculture Organization of the United Nations (FAO) plays a key role in coordinating regular desert locust (*Schistocerca gregaria*) surveillance and monitoring in about 25 countries across Africa, Middle East and southwest Asia where permanent desert locust breeding areas exist plus another 25 countries that can have invasions. FAO developed eLocust3 –

a standardized locust field data collection system operational on mobile tools. With a press of a button, locust scouts send the data via satellite or the internet to National Locust Centres where the information is received and imported into a geographic information system (GIS) called RAMSES. Then the data are exported from RAMSES are exported to the Desert Locust Information Service (DLIS) operating at FAO Headquarters in Rome where it fully explores and analyses the data to forecast and provide early warning. The analysis takes place within a sophisticated Locust Data Cube system that assesses temporal and spatial trends. It also utilizes a GIS called SWARMS, which incorporates other data layers such as trajectory and dispersion models, temperature, rainfall and soil moisture predictions from meteorological satellites, models and remotely sensed vegetative cover information. Since 1996, SWARMS has been operationally used for locust early warning, and its databases contain historical data for nearly 100 years. As a result, monthly desert locust bulletins are produced and disseminated by the DLIS in three languages (English, French and Arabic). During periods of increased locust activity, the bulletins are supplemented by alerts, warnings and updates. The latest information about the locust situation can be found on FAO Locust Watch website <https://www.fao.org/locust-watch/en>. Similar locust monitoring set up is being developed for Central Asia in recent years through the Caucasus and Central Asia Locust Management System (CCALM).

SE39 C05

FAO-SADC-EU COLLABORATIVE EFFORTS TO DEAL WITH TRANSBOUNDARY PLANT PESTS IN THE SOUTHERN AFRICA REGION

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Transboundary plant pests are a major limitation to crop production and the trade of agricultural commodities in the Southern African Development Community (SADC) region. Outbreaks of pests threaten the food and nutrition security of countries in the region and impact negatively on both regional and international trade. The development of regional systems and capacities for early detection, accurate identification, sustainable, effective, and coordinated management of these pests in the 16 SADC countries was supported through the EU-funded project, "Support Towards the Operationalization of the SADC Regional Agricultural Policy (STOSAR)". Through strategic partnership anchored on the unique strengths and comparative advantages of each partner, FAO, SADC and EU successfully co-created innovative solutions to increase productivity and competitiveness of agriculture, while preventing the entry and spread of pests of economic importance in the SADC region. The following were achieved through STOSAR:

- Development of harmonized regional and national strategies for five transboundary pests (oriental fruit fly, fall armyworm, tomato leaf miner, banana Fusarium wilt TR4, and maize lethal necrosis disease);



- Strengthened capacity of 876 NPPO staff in pest surveillance, identification and management including pest risk analysis and compliance with other ISPMs;
 - Assessment of laboratories for accreditation readiness, and equipping of labs with equipment, reagents and consumables;
 - Knowledge and information sharing as well as policy advocacy through regional meetings of the SADC Plant Protection Technical Committee, production of four policy briefs, a peer-reviewed publication, two regional guidelines and two baseline reports;
 - Enhanced capacity of countries to access new markets for plants and plant products by developing national pest lists for priority trade commodities, improved understanding and implementation of phytosanitary measures, and improved pest surveillance, risk assessment, and management.
- Challenges were faced but important lessons were learned in developing and implementing a regionally coordinated approach for transboundary plant pest management.

SE39 C06

ADDRESSING TRANSBOUNDARY PLANT PEST AND DISEASE CHALLENGES IN TÜRKİYE

Suat Kaymak, Begül Kandiltaş, Ayşe Uysal

Department of Plant Health Research, General Directorate of Agricultural Research, Ministry of Agriculture and Forestry, Türkiye

Transboundary plant pests and invasive species are important species that are introduced from different geographical regions through diverse means. They spread and establish in new areas threatening crop production, economies, ecosystems and biodiversity. Increasing travel, trade, and tourism associated with globalization, economic development and population increase contribute to movement of species beyond their natural biogeographical boundaries. As a result, many alien species find their ways to new destinations across borders or through airborne movement becoming invasive and causing significant environmental and economic damages. To address the emerging challenges of the invasive pests, Türkiye is running national programmes and projects to minimize the spread of the introduced pests and prevent entry of other potential threats. For this purpose, regular and targeted surveillance programmes and strict border controls are employed to detect potential new cases. For the established pests, integrated pest management practices are promoted focusing on biological control methods and sustainable agronomy practices. Within the Ministry of Agriculture and Forestry, four specific plant protection research institutes (In Ankara, Adana, Diyarbakir and Izmir) and over thirty departments within other institutes carry out research programmes and projects on plant health. Among the recent critical incursion of pests for Türkiye, the most noteworthy examples include *Ricania japonica*, *Halyomorpha halys* thought to be introduced through eastern Black Sea border, and *Tuta absoluta* through coastal areas. Quarantine organisms red palm weevil (*Rhyncophorus ferrugineus*), and citrus long-horned beetle (*Anoplophora chinensis*), are known to be introduced through tree species imported from abroad. Recent interception of similar pests include mediterranean fruit fly (*Ceratitidis capitata*), corn borer (*Sesamia nonagrioides*) and *Fusarium oxysporum* f. sp. *cubense* (Foc) Tropical race 4 of Banana. So far, targeted survey activities indicate that *Xylella fastidiosa* and Citrus greening disease (*Candidatus liberibacter*) have not been intercepted in Türkiye.

SE39 001

THE POTENTIAL OF INTERCROPS AGAINST FALL ARMYWORM, *SPODOPTERA FRUGIPERDA* (J.E. SMITH) ON MAIZE UNDER FIELD CONDITIONS IN INDIA

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Fall armyworm (FAW), *Spodoptera frugiperda* (J.E. Smith) is a polyphagous and invasive insect pest that causes significant damage to maize threatening food and nutritional security. Extensive use of synthetic pesticides is not sustainable for managing the FAW in maize due to development of insecticide resistance, pest resurgence and toxicity to non target organisms. Climate-resilient intercropping systems enhances crop protection and reduces the carbon foot print of agriculture. Therefore, the present study aimed to identify suitable intercrops for the management of FAW in maize ecosystem. The experiment was conducted in the rainy season of 2023 at three different locations in India in a randomized block design with a plot size of 10 m x 10 m. The treatments were five and replicated four times. The different treatments were maize + cowpea (1:1), maize + groundnut/green gram (1:1), maize + fenugreek/amaranthus (2:2), maize alone and farmers' practice. Field observations on number of plants infested, leaf damage rating (1-9 scale), natural enemies, weed population and grain yield were recorded. The mean percent plant infestation and leaf damage rating on 1-9 scale was lowest in maize + cowpea (9.83, 2.64) followed by maize + groundnut (11.22, 2.86) compared to sole maize (19.85, 4.22). The overall mean population of coccinellids, spiders, earwigs per plant was maximum in maize intercropped with Cowpea (0.89, 0.96, 0.43) followed by intercropping with groundnut (0.87, 0.88, 0.39) compared to sole maize (0.78, 0.66, 0.28) and farmers practice (0.32, 0.38, 0.23), respectively. The weed suppression was observed in maize intercropped with cowpea and groundnut compared to sole maize. Maximum grain yield and the highest cost-benefit ratio was obtained in maize intercropped with cowpea (62.61 q/ha, 1:3.0) followed by groundnut (59.55q/ha, 1:2.71) in comparison to sole maize (46.13 q/ha, 1:1.84), respectively. Location specific intercropping had shown positive influence with cowpea, groundnut in maize with reduction in FAW damage, increased natural enemy population, weed suppression, higher yields and economic returns.

MC3

08.30-11.00 Concurrent Session **40A**

Recent Advances in Vegetable IPM

Chair: Ricardo Oliva (World Vegetable center, Taiwan)

SE40 001

EFFICACY OF SOIL BIODISINFECTION WITH BRASSICACEAE SPECIES FOR CONTROLLING FUSARIUM OXYSPORUM F. SP. LACTUCAE IN LETTUCE CROPS

Palmero D.

1Universidad Politécnica de Madrid, Madrid, Spain

In Spain, a prominent European lettuce producer, the emergence of *Fusarium oxysporum* f. sp. *lactucae* (FOLac) race 4 poses new challenges, demanding innovative control strategies. This study evaluates the efficacy of biosolarization using Brassicaceae species to control FOLac, using naturally infected soil from lettuce commercial fields. Test involved mesocosm with FOLac-infested soil, assessing five Brassicaceae species as biofumigants. Using a randomized block design, 3-liter containers were prepared for each biofumigant species, and the experiment was replicated to analyze two different biofumigant doses. Brassica biomass was processed (sliced, crushed) and incorporated into the soil, followed by irrigation, and covering with transparent polyethylene film for small-scale biosolarization, lasting a minimum of 30 days. After this period, a susceptible lettuce variety was planted and monitored for 30 days. The analyses of glucosinolates were also conducted in the Brassicaceae species. Samples were frozen, lyophilized, and then subjected to glucosinolates extraction, purification, desulfatation, and analysis by HPLC. This information was critical as glucosinolates, upon hydrolysis, transform into biocidal isothiocyanates, which are the active compounds in biofumigation. The results indicated a significant reduction in *Fusarium* populations and disease severity index in the treated soils compared to untreated controls. Biosolarization notably enhanced disease control, primarily attributed to sustained anoxia and elevated temperatures (Fig 1).

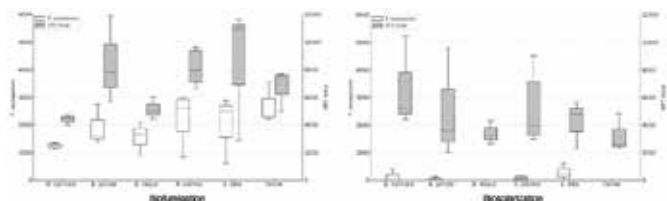


Fig 1.- Fungal population after soil treatments

The analysis of glucosinolates revealed distinct profiles among the Brassicaceae species, which could influence the efficacy of biofumigation. Although biofumigation alone also reduced pathogen density (Fig 2), its effect was less pronounced than that of biosolarization.

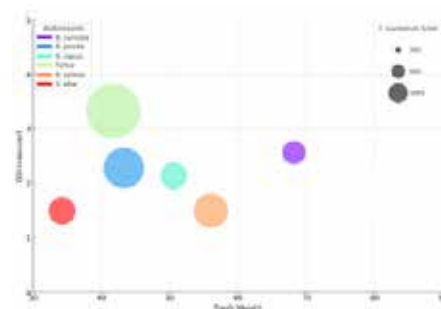


Fig 2.- Effects of biofumigation on Fusarium wilt severity

This study demonstrates that biosolarization using Brassicaceae residues provides substantial protection against *Fusarium* vascular wilt in lettuce, positioning it as a potential alternative to chemical fumigants. Funding by Plan Estatal de Investigación Científica, Técnica y de Innovación 2021-2023 (PID2021-125545OR-C22).

SE40 C01

CHALLENGES AND PROSPECTS FOR IMPLEMENTING VEGETABLE IPM IN ASIA

Ramasamy Srinivasan

World Vegetable Center, Shanhua, Tainan, Taiwan

Asia stands as the global leader in vegetable production, commanding approximately 72% of the acreage and yielding an impressive 927 million tons annually. Despite this remarkable feat, there remains untapped potential, as evidenced by the steady increase in productivity from 20 t/ha in 2013 to 22 t/ha in 2022. However, this figure falls short of the region's actual capability, highlighted by the notable yield differentials across Asian sub-regions. While Eastern and Western Asia boast yields ranging from 26-32 t/ha, South- and Southeast Asia struggle, achieving only 12-16 t/ha. These disparities stem not only from variations in production systems but also from persistent challenges in pest and disease management. Consequently, many vegetable growers use indiscriminate chemical pesticides to mitigate yield losses. However, heightened concerns for human and environmental well-being have prompted Asian governments to pivot towards safer agricultural practices. Integrated Pest Management (IPM) has emerged as a cornerstone of this shift, although its adoption varies significantly among countries and commodities. Institutional inertia, inadequate infrastructure, limited access to IPM resources, and a lack of market incentives for sustainably produced goods hinder widespread adoption. Collaborative efforts led by the World Vegetable Center, alongside national agricultural research



systems and the private sector, have been pivotal in developing, piloting, and scaling IPM technologies for diverse vegetable crops in the region. This presentation will delve into the key challenges and potential pathways for promoting IPM in Asian vegetable production systems to catalyze sustainable agricultural practices across the continent.

SE40 C02

VEGETABLE GRAFTING, A POTENTIAL IPM COMPONENT FOR THE MANAGEMENT OF SOIL BORNE DISEASES OF TOMATO IN ETHIOPIA

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1. World Vegetable Center, Addis Ababa, Ethiopia, 2. World Vegetable Center, Taiwan

The study was conducted to evaluate the graft compatibility, performance, and potential for usage in integrated fusarium and bacterial wilt management (IPM) of nine eggplant and one tomato rootstock varieties grafted on hybrid tomato cv. Gelila 39. Rootstock and scion seedlings were grown under controlled conditions in the greenhouse. Grafting was conducted when seedlings reached a 3-4 leaf stage. RCBD with three replications were used including un-grafted control. There was high compatibility between eggplant rootstocks and hybrid tomato cultivars with graft success ranging from 80 to 100% for most of the rootstock scion combination except VIO 41945 and VIO 41809-A. VIO46103/Gelila 39 and VIO43614/Gelila 39 demonstrated good graft compatibility, higher yield, and resistance to Fusarium and bacterial wilt complex. VIO41945/Gelila 39, VIO41979-A, and VIO41809-A also exhibited good graft compatibility and high yield, but susceptible to the wilt disease complex. Grafting of tomato is a viable and effective way to improve tomato production and productivity in Ethiopia. To ensure that Ethiopia has access to the best disease-resistant cultivar across all areas, some of the imported rootstocks that were resistant to bacterial wilt and fusarium diseases should be tested in many places. Future considerations should include expanding the use of tomato grafting activities and popularizing and demonstrating them as an IPM component.

SE40 C03

TRACKING SOIL-BORNE PATHOGENS IN THE VEGETABLE AGROECOSYSTEM

Ian Quibod¹, Jaw-rong Chen², Hsiao-ping Huang^{2,3}, Ya-ping Lin², Lourena Maxwell², Alvaro Perez-Quintero¹, Ricardo Oliva²

1. Health Institute of Montpellier (PHIM), Université Montpellier, IRD, CIRAD, INRAE, Institut Agro, Montpellier, France, 2. The World Vegetable Center, Shanhua, Taiwan, 3. National Taiwan University, Taipei, Taiwan

The emergence of highly aggressive clones of plant pathogens in vegetable agroecosystems is a significant threat to the global supply of nutrients. However, the response to contain fast-evolving pathogens is usually slow and not

geographically targeted. Preventing disease epidemics will require fast-tracking the distribution of pathogens' variants and customizing the deployment of resistance in a way that aligns with consumer demands. Bacterial wilt, caused by *Ralstonia solanacearum* species complex (RSSC), is one of the most devastating soil-borne diseases of tomato, but it also affects other important solanaceous crops such as potato, pepper, and eggplant. The disease is difficult to control due to its persistence in the soil and the lack of genetic resistance in commercial varieties. We used this disease as a key study since there is no systematic information on the distribution of the different variants of the pathogen. Through comparative genomics on 450 strains, we identified seventeen RSSC populations in the global context. We used highly conserved genomic regions to develop DNA markers for effective surveillance of RSSC in Asia and Africa. In addition, we are validating the resistance profile of several rootstocks and tomato lines carrying different R genes in different regions. In this context, we developed an interactive platform, called VeggieMon, which integrates early-season diagnostics and disease resistance profiles for surveillance purposes. We hope the platform will allow us to manage bacterial wilt epidemics in real time and to define breeding priorities for the region.

SE40 C04

ENHANCING PLANT DEFENSE MECHANISMS: INTEGRATING RESISTANT VARIETIES, BIOCONTROL AGENTS, AND PLANT ACTIVATORS FOR SUSTAINABLE SOUTHERN BLIGHT MANAGEMENT

Lourena Maxwell¹, Chien-hua Chen¹, Francely Flores², Jaw-rong Chen¹, Ming-Ren Yen³, Yu-Shin Nai³, Assaf Eybishitz¹, Derek Barchenger¹, Ricardo Oliva¹, Srinivasan Ramasamy¹

1. World Vegetable Center, Shanhua, Taiwan, 2. Zamorano University, San Antonio de Oriente, Tegucigalpa, Honduras, 3. National Chung Hsing University, Taichung, Taiwan

Southern blight, caused by the soil-borne fungus *Sclerotium rolfsii*, is among the most challenging soilborne diseases for farmers of various crops, including tomato and pepper, leading to significant yield and economic losses. Chemical fungicides offer limited effectiveness against this persistent pathogen, highlighting the need to enhance plant defense mechanisms for sustainable disease management. The study aimed to assess the effectiveness of resistant varieties, biocontrol agents, and plant activators in managing Southern blight in tomato and pepper crops. Greenhouse experiments were conducted to examine the tolerance of different tomato and pepper breeding lines and accessions to Southern blight. The effectiveness of the plant activator (Neutralized phosphoric salt) and biocontrol agents (*Trichoderma* sp. and *Bacillus* sp.), alone or combined with tolerances, was assessed in controlling Southern blight in tomato. RNA-Seq analysis was performed to identify differentially expressed genes triggered by these interventions. For pepper, the breeding line AVPP9813 exhibited higher seedling survival within the first 72 hours. Transcriptome analysis revealed 72



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differentially expressed genes, including receptor-like kinases, antioxidants, transcription factors, and pathogenesis-related genes associated with defense and signaling responses during infection. For tomato, integrating resistant tomato accession VIO09462 (*Solanum lycopersicum* L), biocontrol agents, and plant activators significantly reduced (80%) the incidence of Southern blight compared to a single or no intervention. In addition, transcriptome analysis also showed the expression of several plant resistance-induced genes, which are candidate genes to be further explored in developing host resistance to Southern blight. These findings highlight the potential of integrating disease management intervention, including resistant varieties, biocontrol agents, and plant activators, to enhance plant defense mechanisms for effective and sustainable Southern blight management.

BANQUET

11.30-14.00 Concurrent Session **37B**

Recent Advances in Plant Virology

Chair: Xueping Zhou (Institute Of Plant Protection, Chinese Academy Of Agricultural Sciences, China)

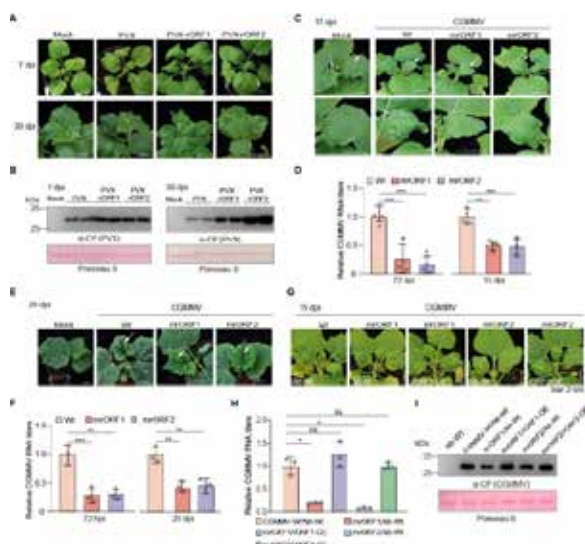
SE37 002

THE -RNA OF TOBAMOVIRUSES ENCODED ADDITIONAL VIRAL PROTEINS

Li F.

Institute Of Plant Protection, China Academy of Agricultural Sciences, Beijing, China

Tobamoviruses with a positive-sense single-stranded RNA (+ssRNA) as viral genome are one group of the most destructive plant viruses. Just like other +ssRNA viruses, it is generally thought that tobamoviruses possess the ability to encode proteins solely on their positive strand (+RNA). However, here we identified small open-reading frames (ORFs) on the negative strand (-RNA) of tobamoviruses with potential coding capabilities. Using cucumber green mottle mosaic virus (CGMMV) as a model, we identified and characterized the corresponding peptides encoded by the viral -RNA (rORF1 and rORF2) through mass spectrometry analysis and ribosome-profiling. We observed that the protein encoded by CGMMV rORF1 localized to peroxisomes, while the protein encoded by rORF2 localized to the cell membrane and nucleus. CGMMV rORF1 could interact with the CGMMV replication-related 126-kDa protein in the peroxisomes. The overexpression of rORF1 and rORF2 could enhance the pathogenicity of potato virus X. Mutation of rORF1 and rORF2 significantly impaired CGMMV virulence. Transgenic overexpression of rORF1 and rORF2 could rescue the pathogenicity of CGMMV rORF1 and rORF2 mutants. Furthermore, we showed CGMMV may employ internal ribosome entry site to translate these rORFs. This study reveals a novel arsenal of +ssRNA viruses: transient -RNA encodes functional proteins, sheds light on the presence and functions of small rORFs in peroxisomes encoded by the -RNA of CGMMV and other tobamoviruses, significantly expanding our knowledge of their protein-coding capacities.



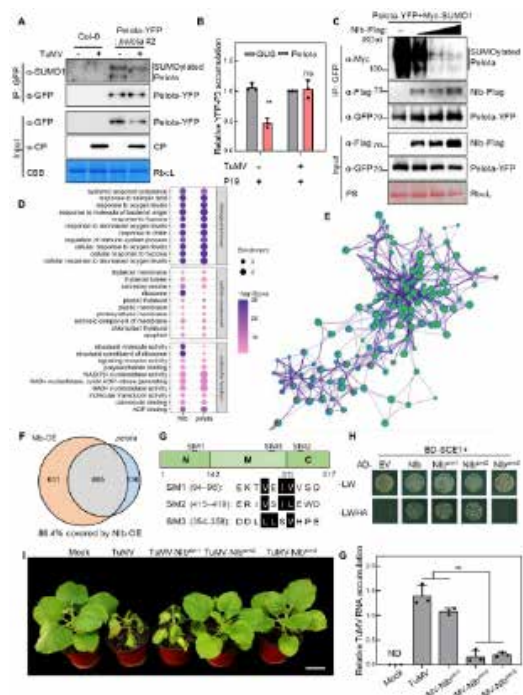
SE37 003

RNA POLYMERASE OF POTYVIRIDS INHIBIT PLANT RNA QUALITY CONTROL

Li F.

Institute Of Plant Protection, China Academy of Agricultural Sciences, Beijing, China

RNA-targeting regulation mechanism is emerging as a critical regulator in plant virus infection. The Pelota-Hbs1 complex, a key component of RNA quality control (RQC), promotes the degradation of Potyvirids RNA by recognizing a specific G1-2A6-7 motif within the P3 cistron. Despite this, the extent to which Potyvirids have evolved mechanisms to evade the Pelota-mediated RQC remains unclear. In this study, using turnip mosaic virus as a model, we demonstrated that virus infection inhibits the RQC activities of Pelota. We subsequently identified the viral-encoded protein responsible for this inhibition, revealing the essential involvement of NIb in this process. NIb can effectively abolish the SUMO modification of Pelota by competing with SCE1, SUMO E2 conjugating enzyme, which ultimately leads to the inhibition of Pelota-mediated RQC. This strategy of RQC evasion appears to be a general event among Potyvirids. Our findings illuminate the dynamic interplays between post-translational modifications, plant defense mechanisms, and viral counter-strategies during virus infection.





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SE37 004

WISTERIA VEIN MOSAIC VIRUS: AN EVOLUTIONARY-BASED CASE STUDY APPROACH TO TRACE THE EMERGENCE OF NEW VIRUS THREATS

Morelli M., D'Attoma G., Saldarelli P., Minafra A.

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Wisteria mosaic disease (WMD) severely affects *Wisteria* spp. and was originally reported in the US. After the identification of its aetiological agent, Wisteria vein mosaic virus (WVMV), a species of the genus *Potyvirus*, numerous reports followed worldwide. In 2023 we first identified an Italian isolate of the virus, named WVMV Bari, and more recently completed its full-length sequence, relying on high-throughput barcoding technology. PhyML-SMS phylogenetic inference placed WVMV Bari in the broad cluster known as the 'bean common mosaic virus supergroup'. In-depth analyses using two-dimensional diversity matrices, built on the coding regions of the ten available WVMV isolates, identified interesting evolutionary traits at the intraspecific level and with close species. A clustering algorithm analysis confirmed the existence of two phylogenetic groups, a major clade comprising WVMV isolates from *Wisteria* spp. and a minor clade at the species boundary comprising two divergent WVMV isolates and one isolate of kudzu chlorotic ring blotch virus, all found in non-*Wisteria*, Fabaceae, hosts. A reconstruction of the evolutionary ties that led to this differentiation was attempted. Potential recombination events were found only between non-*Wisteria* hosts. The divergence time between the two clades was estimated around the 18th century, based on relaxed molecular clock dating. Median Joining network analysis substantiated the existence of the two host-related groups, by highlighting the number of mutations leading to the separation. K_{xy} , D_{xy} and D_a inter-population parameters and permutation-based tests revealed a high level of genetic differentiation between the two phylogroups. The fixation index (F_{ST}) and migrant value (N_m) showed an infrequent gene flow. Negative neutrality tests in *Wisteria*-affecting variants may prove a recent population expansion or the occurrence of purifying selection. Evidence of negative selection was also found in both phylogroups, based on dN/dS ratio. SLAC analysis identified sites under selection in the taxonomically relevant CP region. Our results highlighted that WVMV is a culpably neglected species whose emergence dynamics raise interesting questions about its taxonomic distinctiveness and its potential to spread in other economically relevant crops, such as legumes. The main intention of our study is to propose an evolution-based analysis approach that serves as a case study to investigate how other emerging viruses may spread.

difficulty in acquiring and the high cost of maintenance pose significant obstacles. Our innovative solution to this problem is the introduction of pathogen-mimicking artificial positive controls (PaMAPCs), which are applicable to viruses fungi and nematodes. This approach significantly broadens the applicability of assays, including quantitative PCR, by offering controls that can be designed and implemented in just three days. This rapid turnaround is in sharp contrast to artificial controls, such as plasmids and gBlocks, which often take several weeks for procurement and use. PaMAPCs provide a highly accurate simulation of natural infections, thus ensuring the easy detection of lab-based contamination. PaMAPCs markedly speed up the development of detection assays while simultaneously boosting their reliability and efficiency in identifying pathogens. This makes PaMAPCs an invaluable asset not only in plant pathology but also in veterinary and human pathology, enhancing infectious disease management across the board. By bridging the gap between the need for rapid, accurate detection and the challenges posed by traditional artificial controls, PaMAPCs represent a significant advancement in the field of diagnostics.

SE37 006

EMERGING VIRUSES INFECTING PEPPER IN OKLAHOMA

Ali A., Pasley C.

The University of Tulsa, Tulsa, United States

Pepper (*Capsicum* spp.) is an economically valuable crop in the world including the United States of America (USA) due to its several benefits to human health in the form of spice or vegetable. Although several viruses have been reported infecting pepper from other states in the USA, but little is known about the type and distribution of viruses infecting pepper in Oklahoma. Therefore, we conducted a comprehensive pepper virome study to identify plant viruses and their subsequent occurrence in six different counties across Oklahoma. We collected >310 samples from pepper and weeds during the 2021 and 2022 growing seasons which were analyzed by high-throughput sequencing (HTS) and reverse transcription (RT) PCR assays. Viral contigs obtained by HTS were further validated using RT-PCR or PCR assays followed by Sanger sequencing. In total, we detected 17 viruses (15 RNA and two DNA viruses) and one viroid. Among the identified viruses, the most dominant virus was beet curly top virus (BCTV), followed by tomato yellow leaf curl virus (TYLCV), potato yellow dwarf virus (PYDV), and pepper mild mottle virus (PMMoV). The viral incidence varied from season to season, and in some cases, constituted >80% during some surveys. Taken together, this study contributes to a greater understanding of the pepper virome in Oklahoma which could assist local growers in recognizing the possible viral agents capable of causing disease in pepper. This knowledge will further assist growers in developing more informed management strategies.

SE37 005

DEVELOPMENT OF PATHOGEN-MIMICKING ARTIFICIAL POSITIVE CONTROLS (PaMAPCs) AND THEIR APPLICATION IN DIAGNOSTICS

Tzanetakos I., Stainton D., Singh S., Sierra Mejia A.

Department of Entomology and Plant Pathology, University of Arkansas, Fayetteville, United States

Detection assays frequently face the challenge of securing positive controls, which are essential for validation. The



SE37 007

PLANT VIRUSES OF THE FAMILY POTYVIRIDAE ENCODE VIROPORIN

Chai M., Li L., Xiang W., Wu X., Cheng X.

Northeast Agricultural University, Harbin, China

Potyviridae is the largest family of plant RNA viruses, members of which cause significant yield losses of many crops worldwide. The 6-kilodalton peptide 1 (6K1) is one of the least characterized potyviral proteins, and its exact biological function has remained elusive. Dissection the biological function of 6K1. Confocal microscopy and subcellular fractionation were performed to study the subcellular localization of 6K1. Bioinformatic approaches such as AlphaFold2 and AlphaFold-Multimer were performed to predict the structure of 6K1. Bimolecular fluorescent complementation, coimmunoprecipitation, and gel filtration were performed to confirm the multimeric state of 6K1. Membrane permeabilization and yeast complementation assays were performed to analyze the viroporin activity. Mutagenesis and infectivity assay were performed to confirm the function of 6K1 in viral proliferation. 6K1 is an endoplasmic reticulum-localized protein and forms multimers. 6K1 forms pentamers with a central hydrophobic tunnel, can increase the cell membrane permeability of *Escherichia coli* and *Nicotiana benthamiana*, and can conduct potassium in *Saccharomyces cerevisiae*. Mutations affect the multimerization of 6K1 attenuates or completely abolish the proliferation of turnip mosaic virus. The 6K1 or its homologous 7K proteins in potyvirids (viruses of the family *Potyviridae*) also have the ability to increase cell membrane permeability and transmembrane potassium conductance. Taken together, these data suggest that 6K1 and its homologous 7K proteins are functional viroporins. The results advance our knowledge on the function of 6K1 and the pathogenicity of potyvirids, which may lead to the development of 6K1 inhibitors for disease management. Hitherto, viroporins were exclusively found in enveloped clinical animal viruses. Thus, our results also shed insight into the distribution and diversity of viroporin. 6K1 and its homologous 7K protein in potyvirids are functional viroporins.

136448, PI 442312, PI 458731, and PI 420328, displayed delayed and less severe symptoms. Nevertheless, CCYV RNA accumulated in all the PI accessions. Lower symptom severity while harboring a considerable amount of CCYV indicates their inherent tolerance to the yellowing disease induced by CCYV. When comparing CCYV RNA accumulation in PI accessions with the commercial cultivar 'Gentry', lower virus titers were observed across all tested accessions. Specifically, PI 420328 and PI 458731 exhibited significantly reduced CCYV titers compared to the susceptible cultivar in both mass exposure and clip cage experiments. PI 420328 was also found to be resistant to CYSDV, another closely related Crinivirus. Lower virus titers and elevated levels of 21- and 22-nucleotide (nt)-sized class CYSDV RNAs, the hallmarks of RNA silencing, were observed in this line, indicating more robust and efficient RNA silencing. These accessions, displaying reduced symptoms and lower virus titers, hold promise as sources of resistance to CCYV and CYSDV in breeding programs.

SE37 008

EVALUATION OF SUMMER SQUASH (*CUCURBITA PEPO* L) FOR RESISTANCE AGAINST EMERGING CRINIVIRUSES

Bag S., Kavalappara S.

University Of Georgia, Tifton, United States

Emerging criniviruses cucurbit chlorotic yellows virus (CCYV), and cucurbit yellows disorder virus (CYSDV) transmitted by whiteflies, poses a significant threat to cucurbit crops globally, particularly summer squash (*Cucurbita pepo* L.) in the Southeastern United States. Due to the absence of resistant commercial cultivars, exploring resistant germplasms becomes imperative. Cucurbita germplasms with potential resistance to CCYV and CYSDV were previously identified through field screening. Controlled greenhouse screening of these germplasm lines was performed with aim to validate their resistance to CCYV infection. The susceptible cultivar Gentry used as control exhibited early and severe symptoms in response to CCYV infection. In contrast, all the PI accessions tested, including PI 512749, PI 615141, PI

SKALKOTAS

11.30-14.00 Concurrent Session **41**

Advances in Nematode Research and Plant Protection

Chair: Thomais Duarte (South East Technological University, Ireland)

Alexandros Dritsoulas (Agricultural University of Athens, Greece)

Giannis Giannakou (Agricultural University of Athens, Greece)

SE41 C01

AN INVESTIGATION ON THE DEVELOPMENT OF A COMBINED PLANT HEALTH BIO-AGENT USING CALCIUM ALGINATE FORMULATION

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The use of conventional chemical pesticides in agriculture poses severe threats on human health and the environment, with soil biodiversity also being negatively affected. Entomopathogenic nematodes (EPN) and plant growth promoting rhizobacteria (PGPR) have considerable potential as biological control agents. Formulations of EPN can be enhanced by PGPR and provide good protection to plants against insects promoting plant health. Two research studies have demonstrated that the PGPR, *Pseudomonas* spp., are associated with the EPN *Steinernema feltiae*, and they play a role in the parasitic lifecycle of these nematodes (Ruii et al., 2022; Ogier et al., 2020). In addition, these two groups of beneficial organisms have been shown to tolerate each other well in a previous enviroCORE project (Hurley, 2018). The current study aims to formulate a novel biological product consisting of a combination of Irish strains of *S. feltiae* (SB 12[1]) and of *Pseudomonas ogarae* F113 GFP. In this study, a novel calcium alginate bead formulation is being developed as a suitable medium to encapsulate EPN and PGPR simultaneously. Both *S. feltiae* (SB 12[1]) and *P. ogarae* F113 GFP were cultured prior to their incorporation into the formulation of calcium alginate bead. 18% glycerol is included in the calcium alginate bead formula to improve the retention of the EPN while being stored and until they are needed for application. Preliminary results showed that a 1.5% sodium alginate-glycerol concentration was the best to generate well shaped calcium alginate beads. Currently work is ongoing to ensure nematode dormancy as well as good organismal viability and infectivity within the beads after short- and long-term storage. Following these experiments, molecular communication analyses and modelling will be carried out to better understand how those biological agents interact in their novel formulation to improve their efficacy in the final biological product.

SE41 C02

METABARCODING SURVEY SUPPORTS SPECIFICITY OF EPN-PAENIBACILLUS SP. ASSOCIATION AND IDENTIFIES POTENTIAL BACTERIAL ANTAGONISTS OF DIAPREPES ROOT WEEVIL IN A FLORIDA CITRUS ORCHARD

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Diaprepes root weevil (DRW, *Diaprepes abbreviatus*) is a major economic pest of citrus trees in Florida and the Caribbean basin. Biological control by native entomopathogenic nematodes (EPN) has been proposed as a driver of DRW abundance across Florida's ecoregions. Weevils also typically occupy specific locations within orchards for unknown reasons. To identify potential causes of local patterns of weevil abundance and tree condition, we measured relationships between DRW and edaphic properties (biotic and abiotic) within a central Florida orchard. Adult DRW were trapped and monitored weekly for two years in 94 plots arranged in a grid pattern within a 2.5 ha area. One year after monitoring began, soil in each plot was sampled and DNA extracted from organisms recovered by sieving-sucrose centrifugation. Soil subsamples were processed for physicochemical properties and DNA was subjected to metabarcoding (Illumina NovaSeq) for three gene regions (ITS2 rDNA, 16S rDNA, and COI mtDNA). Species-specific qPCR primer-probe sets were also used to measure *Steinernema diaprepesi* and *Heterorhabditis indica*. Here we focus on a restricted set of 124 amplicon sequence variants (ASV, comprising 55 identified *Paenibacillus* species) because of the known entomopathogens in this group and the two species reported to be ectoparasites of EPNs. Soil pH was strongly associated with *Paenibacillus* ASVs ($P < 0.001$). Fourteen bacterial ASVs were dissociated with DRW ($P < 0.05$), whereas none were positively associated with the weevil according to Spatial Analysis by Distance Indices (SADIE). Several *Paenibacillus* species, elevation, coarse sand particles, and combined ASVs of all identified nematophagous fungi (but no EPN) were significant variables explaining 36% of DRW and tree condition variability in a redundancy analysis. Of 123 ASVs, only *Paenibacillus* sp. JF317562, an ectoparasite of *S. diaprepesi*, was highly correlated ($r = 0.82$, $P < 0.0001$) to that nematode measured by barcode. The ectoparasite was unrelated ($r = 0.12$, NS) to *S. diaprepesi* measured by qPCR.



SE41 C03

NEW ADVANCEMENTS IN ROOT-KNOT NEMATODES MANAGEMENT USING PLANT GROWTH PROMOTING RHIZOBACTERIA

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The root-knot nematodes (RKN; *Meloidogyne* spp.) are among the most damaging pests, affecting both the quantity and quality of many annual and perennial crops, and causing major yield losses. *Meloidogyne* species have an extremely broad host range, affecting over 2000 cultivated plant species and over 5500 plant species in total, causing severe damage through root gall formation and root dysfunction. The number of active substances allowed for plant protection decreased under Directive 91/414/EEC (followed by Regulation 1107/2009/EC), resulting in the ban and withdrawal of many pesticides from the EU market. Bacteria of the genera *Bacillus* and *Pseudomonas*, many of which belong to the group of Plant Growth Promoting Rhizobacteria (PGPR), dominate many soil ecosystems and are considered effective competitors against plant-parasitic nematodes. Due to their facultative parasitic nature, they can establish themselves or remain dormant in the soil even without the presence of the nematodes they combat. These bacteria can reduce nematode populations through various mechanisms such as: a) direct parasitism of nematodes, b) antibiosis along with a reduction in root parasitism, c) induction of systemic resistance in plants, d) formation of biofilm on the root surface, and e) competition for nutrients. Also, the addition of certain bacterial species to the soil can, through the induction of systemic resistance, cause changes in biochemical pathways within the plant, ultimately creating conditions that deter the development of nematodes. In the present work, different genera of PGPR were used against RKN. Flat-bottom 24-well plates were used for motility *in vitro* bioassays using second-stage juveniles and metabolites. The efficacy of certain bacterial strains was tested in soil infested with root-knot nematodes. Efforts were made to test the induction of resistance using the split-root system of tomato plants. The research is in progress, utilizing both commercially available strains and those isolated from indigenous plant species. The results obtained using commercial products containing PGPR are discussed.

SE41 C04

INVESTIGATION OF FLUAZAINDOLIZINE AS A POTENTIAL NOVEL TOOL TO MANAGE XIPHINEMA INDEX

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Salibro™ (500 SC) is a novel nematicide containing the active ingredient (a.i.) fluaizindolizine (Reklemel™ active). In *in vitro*

tests, its effect on the motility and the mobility of *Xiphinema index* were evaluated at 0.5 to 100 ppm a.i. in comparison to Vydate® 10L at 12.5 ppm a.i. (a.i. oxamyl) and Velum PRIME® SC at 1 ppm a.i. (a.i. fluopyram). Salibro at 5 to 100 ppm a.i. as well as both other nematicides strongly reduced the motility of *X. index* compared to the control at all incubation times (24 to 72 hours). However, if the pre-exposed nematodes were rinsed and challenged to migrate through a 1 cm layer of coarse sand (mobility test) only the longest pre-exposure period of 72 hours caused a significant effect at 25-100 ppm of Salibro a.i. This indicates some level of reversibility. In addition to the *in vitro* experiments, pot tests were run on fig trees with various pot sizes (0.250 to 1.5 L), different timings of inoculations as well as different environmental conditions (indoors vs outdoors) to evaluate the impact of Salibro on *X. index* reproduction. Throughout the course of those experiments, it became clear that only the higher rates of Salibro (with 1 to 2 kg ai/ha) would qualify for sufficient and reliable control of *X. index*. Still Salibro performed significantly better and more consistently, than both of the other nematicide standards. Currently long fallow periods are recommended after grape vine uprooting and before replanting to eliminate *X. index*. In such replanting programs, one or more applications of Salibro around the newly planted vines may have the potential to prevent the subsequent build-up of any residual nematode population.

SE41 001

POTENTIAL OF BIOCIDES IN THE MANAGEMENT OF MELOIDOGYNE INCOGNITA PARASITIZING BEETROOTS

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Beetroot an important vegetable is threatened by various field and postharvest pests. Plant parasitic nematodes are important pests of beetroot. *Meloidogyne incognita*, a dominant member of the taxon induces root-knot diseases which thwart the translocation of nutrients and water, invariably reducing yield of beetroots on the field. The spate of *M. incognita* infestation of beetroots has demanded recurrent use of nematicides, and are widely used by farmers and non-farmers alike. Systematic exposure to pesticides leads to environmental pollution and severe health threats. This necessitated development of biocides in the management of *M. incognita*. This study aims to evaluate the probable effect of essential oil and powdered material from *Blumea aurita* on *M. incognita* infected beetroot plants. Whole plant of *B. aurita* was air dried at ambient temperature and reduced to fine particles by pounding. Leaves of *B. aurita* were hydro distilled to extract the essential oil (EO). Constituents of essential oil were analysed by gas chromatography mass spectroscopy (GCMS). The field experiment was a randomised complete block design (RCBD) with five replicates. Beetroot seedlings were inoculated with 2000 Juveniles of *M. incognita*. EO (50, 75, 100 mg/ml) and powdered material (250, 275 and 300 g) were compared with furadan (0.5, 1.0 and 1.5 kg/ai/ha). GCMS results indicated cavacrol, borneol, caryophyllene epoxide, δ -elemene, and α -copaene as major compounds. Gall index and *M. incognita* population was significantly low in treated plants at harvest. A significant ($p < 0.05$) reduction in growth of beetroots was observed in untreated inoculated plants, while plants treated with EO had higher yields



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compared to powdered material treatments. *B. aurita* has potential in the management of *M. incognita* on beetroots, thus ensuring abundance of safe and nutritious vegetable to maintain a healthy life. Active compounds in EO could be formulated into commercial products to minimize challenges of *M. incognita* management.

SE41 002

ROOT-GALL NEMATODE MANAGEMENT STRATEGY ON OKRA (*ABELMOSCHUS ESCULENTUS*) WITH PHYSICNUT [*JATROPHA CURCAS* (L)] TANNINS

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Among natural plant products phytochemical based strategy provides a potential alternative to synthetic chemical control of root-gall nematodes. *In vitro* and screen house experiments were carried out to evaluate the efficacy of *J. curcas* Tannins and plant parts on okra infected with root-gall nematode with the aim of developing a plant based nematicide. Efficacy of Tannins against nematode (J2s) mortality was evaluated *in vitro*. A 3x6 factorial experiment fitted in Completely Randomized Design was conducted to control root-gall infection on Okra *in vivo*. Data obtained were subjected to Correlation analysis to determine the relationship between root-gall index and yield using SPSS and GENSTAT. Results: Tannins evaluated against nematode juveniles (J2) in *in vitro* conditions revealed that nematode mortality was highest (94 %) after 72 h incubation period at 10 ml rate of application. Results showed that Tannins applied at 5 ml /pot significantly ($P=0.05$) reduced root-galls and increased plant growth parameters on application of leaf and seed tannins than the untreated control in 2019 and 2020 years of study respectively. Leaf and seed Tannins also increased Okra pod weight by 67 % (4.88 g) and 61 % (5.11 g) over the control in both years. The study shows the scientific validation of Physic nut Tannins as a potential alternative to synthetic chemical control of root-knot nematodes for the development of an environmentally friendly Biopesticide. However, studies conducted did not investigate the effect of tannins in other types of soil and nematode species. Field evaluation is therefore suggested to consolidate on the gains of this study.



MC2

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Biological Plant Protection

Chair: Massimo Pugliese (University of Torino, Italy)
Danai Gkizi (University of West Attica, Greece)

SE42 001

INTEGRATED DISEASE MANAGEMENT OF POWDERY AND DOWNY MILDEWS ON GRAPE: RECENT RESULTS IN ITALY

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Powdery (causal agent *Erysiphe necatrix*) and downy (causal agent *Plasmopara viticola*) mildews are the most important grape diseases, causing relevant losses both in terms of quality and quantity. Climate change may further favour the development of these diseases in the future, under policies limiting the use of chemical fungicides, copper included. The purpose of this study was to evaluate the efficacy of products with low environmental impact on the cultivars "Arneis" and "Nebbiolo" both on potted plants and under field conditions during the years 2020-2022. The antifungal products were applied according to the manufacturer's instructions, with a hand-pulled 2-stroke engine sprayer at a pressure of 15 bar, distributing 400-600 L/ha of water. A standard strategy based on sulphur + copper was used as control. For each cultivar, the experimental scheme included four randomized blocks per treatment, each containing 10 plants. *Ampelomyces quisqualis*, *chitoooligosaccharides* and *oligogalacturonides* (COS-OGA), and essential oil of sweet orange were as effective as sulphur in controlling powdery mildew on potted plants. *Bacillus pumilus* was also effective compared to the untreated control. Potassium phosphate was the most effective, among the other tested products, in controlling downy mildew on potted grapevines. The integrated disease management strategies applied in field and based on COS-OGA, essential oil of sweet orange and potassium phosphate significantly reduced the two diseases to a level similar to that observed by the standard strategy based on sulphur + copper. Furthermore, the amount of copper was reduced by 1.2-1.3 kg/ha applying the integrated strategies compared to the standard one. Low environmental impact products were effective in trials carried out on "Arneis" and "Nebbiolo", showing to be applicable in vineyard to reduce, at least partially, the use of traditional fungicides and to comply recent restrictions in the use of copper.

SE42 002

BIOLOGICAL CONTROL OF BOTRYTIS CINEREA USING COMMERCIAL WINE YEASTS

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Botrytis cinerea causes significant yield losses every year globally in viticulture. Grapes are very prone to *Botrytis* rot when they reach maturity, but at this point the use of chemical products to control the disease is not an option since chemical residues will contaminate the production. *Botrytis* is also notorious for easily developing resistance in chemical fungicides. To overcome the above problems, research is focused on the biological control of the disease. The present study focuses on the use of 5 commercial wine yeasts belonging to the species *Saccharomyces cerevisiae*, *S. pastorianus*, *S. bayanus*, *Lancacea thermotolerans* and *Tolulaspora delbrueckii*, to control the growth of *B. cinerea*. *In vitro* assays showed that although the mycelial growth was not reduced in the presence of yeast cells, metabolites or volatiles at 3 or 6 days post inoculation, at the end of the *in vitro* experiment the production of *Botrytis* conidia was significantly reduced in the presence of yeast metabolites or volatiles. *Ex vivo* assays in detached berries immersed in the yeast cultures showed significantly reduced growth of *B. cinerea* at 15 days post inoculation with the pathogen combined with reduced conidia production. In order to study further the mode of action of the *S. bayanus* yeast strain which presented the most promising results *in vitro* and *ex vivo*, Real-time PCR gene expression analysis was performed in PR3, PR4 and PR5 grapevine defense genes, at 1 and 3 days post inoculation with *Botrytis* on detached berries. The result showed no significant defense induction by the yeast leading to the assumption that its mode of action against *Botrytis* is the antagonism for space and nutrients. Our results indicate that the use of yeast strains already available on the market for wine making may also be used for the biological control of the disease in vineyards.

SE42 003

PROSPECTIVES OF CATENARIA ANGUILLULAE SOROKIN FOR BIOLOGICAL CONTROL OF PLANT PARASITIC NEMATODES

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Since agricultural production around the world has considerably been threatened by diseases caused by plant parasitic nematodes owing to modernization in agriculture for satisfying the need of an alarmingly growing population and their ill consequences

on the existing natural biological equilibrium, the development of ecofriendly methods of disease management involving use of biological control agents is of paramount significance for sustainable agriculture with ultimate goal of safe food security for good human health. *Catenaria anguillulae* is a ubiquitous, facultative endozoic zoosporic blastocladian fungal parasite of nematodes that has received a good attention of a large number of researchers after it was first reported by Sorokin in 1876 causing epidemics in nematode population. This fungus has almost all the attributes what an effective biological control agent must have such as fast multiplication, compatibility with wide range of pH and temperature, salt concentrations and soil additives commonly used for raising crops, particularly oilcakes, fertilizers and agrochemicals. The fungus also serves as the best educational tool to demonstrate biological control in class room without any chemical input in water only. A detailed work was done on refinement of media for its mass production. The OS1 and M2 media were found suitable for mass culture. The experiments on performance of *C. anguillulae* following its mass culture against *Heterodera cajani* and *Xiphinema basiri* revealed the reduction in population of the test nematodes. The aforesaid facts about the fungus clearly reflect its potential as an appropriate biological control agent against plant parasitic nematodes. However, an extensive work is still required on formulation development, delivery system and performance against a wide range of plant parasitic nematodes.

SE42 004

EXPLORING THE TERMICIDAL PROPERTIES OF CLOVE (*SYZYGIUM AROMATICUM*) AND GARLIC (*ALLIUM SATIVUM*) AGAINST THE SUBTERRANEAN TERMITE, *HETEROTERMES INDICOLA* (ISOPTERA: RHINOTERMITIDAE)

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Heterotermes indicola (Wasmann), a subterranean termite belonging to the Isoptera: Rhinotermitidae family represents a severe threat to wooden structures in Pakistan. Traditional termite control methods primarily involve the use of synthetic insecticides, which not only pose hazards to the environment but also adversely impact non-target organisms, including humans. To address the ecological concerns associated with synthetic chemicals, we conducted a comprehensive study to investigate the termiticidal properties of clove (*Syzygium aromaticum*) and garlic (*Allium sativum*) against subterranean termites. This research was carried out at the "Termite Laboratory" of the Plant Protection Division (PPD), Nuclear Institute for Food and Agriculture (NIFA), Peshawar, over the period of 2022-2023. The laboratory maintained a consistent temperature of $25 \pm 2^\circ\text{C}$ and a relative humidity of $65 \pm 5\%$. Our study examined the toxicity and deterrence capacities of two botanicals, namely clove (*S. aromaticum*) and garlic (*A. sativum*). We prepared aqueous and alcoholic extracts of clove at concentrations of 0.25%, 0.5%, and 1%, in addition to a 1% garlic solution. Notably, clove exhibited remarkable toxicity and deterrence, even at a concentration as low as 1%, while garlic displayed comparatively limited efficacy.

We also observed the persistence of residual toxicity, with clove-treated arenas retaining their toxic effects for more than 60 days, whereas garlic lost its potency and deterrence over time. To evaluate the synergistic effects, we combined clove and garlic extracts at ratios of 1:1, 1:2, and 1:4, finding that ratios of 1:4 and below demonstrated effective deterrence. In summary, our findings highlight the exceptional efficacy of a 1% clove extract, which resulted in the highest mortality among worker termites. Ethanol extracts of clove consistently outperformed their aqueous counterparts. Similarly, the aqueous extract of garlic proved less effective when compared to its ethanol counterpart. Most notably, the combination of both clove and garlic extracts exhibited a synergistic effect, offering robust termite deterrence. Ultimately, all concentrations of clove and garlic, particularly in ethanol extracts, emerged as potent repellents against subterranean termites.

SE42 005

USE OF ENTOMOPATHOGENS AND NANO-PARTICLES FOR THE SUSTAINABLE MANAGEMENT OF RED FLOUR BEETLE, *TRIBOLIUM CASTANEUM* (TENEBRIONIDAE: COLEOPTERA)

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Tribolium castaneum, is a prevalent agricultural pest that invades stored grains. The aim of bioassay to compare the nanoparticles and entomopathogenic fungi to control *Tribolium castaneum*. The complete randomized data method was applied to analyze all the data, and all pairwise comparison tests used Tukey's HSD at 5% significance. The use of entomopathogenic *Beauveria bassiana*, *Metarhizium anisopliae* and *Aspergillus flavus* is the first step in our research. All of the fungi showed detrimental effect on *T. castaneum*, in the results. The concentrations of *B. bassiana* with 81.69% and 62.94% were very applicable than *A. flavus* and *M. anisopliae*. The toxic effect of nanoparticles comprising Ag and ZnO assessed and examined for adult *Tribolium castaneum*. Results revealed that Silver showed more effective treatment other than zinc. Different concentrations of Silver triggered mortality rates ranging from 55.26% to 90.52%. The possibility for ecological treatment of *Tribolium castaneum* can be attained through the collective use of nanoparticles and entomopathogens. Results showing that silver and strains of *B. bassiana* showed operative result compared to other combinations. The mortality of 90.16% was detected on the 9th day for the concentration of $350 \pm 1 \times 10^9$, when strain *B. bassiana* combined with varying concentrations of Ag. The conclusion of the study that *Tribolium castaneum* is susceptible to commercial *B. bassiana*, *M. anisopliae*, and *A. flavus* formulations. The investigated fungal strains and Nanoparticles may handle *T. castaneum* and other stored-product pests.



SE42 006

ELICITOR APPLICATION IN STRAWBERRY IPM RESULTS IN LONG-TERM INCREASE OF PLANT RESILIENCE WITHOUT YIELD LOSS

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As a first step to integrate elicitor applications into current IPM strategy increasing plant resilience against pests, we investigated repeated elicitor treatments in a strawberry everbearer nursery and cropping cycle under glass. During nursery methyl-jasmonate (MeJA) was applied every three weeks, testing induction of defenses with plant bioassays. Thrips damage and reproduction by spider mites, whitefly and aphids were strongly reduced upon elicitor treatment. Subsequently, we applied MeJA every three weeks, or based on scouting of pests, during a whole cropping cycle. Thrips leaf bioassays and LC-MS leaf metabolomics were applied to investigate induction of defenses. Leaf damage by trips was lower for both MeJA application schemes compared to the control except the last weeks. Thrips damage decreased from vegetative to mature plants during the cropping cycle. Surprisingly, growth during production was not affected by MeJA application as were fruit yield and quality. LC-MS leaf metabolomics showed a strong induction of vegetative plants decreasing during maturation of plants towards the end of cultivation. Mostly flavonoid and phenolic glycosides known as plant defense compounds were induced. While induced defense decreased with maturation of plants constitutive defense increased as measured in the leaf metabolome of control plants. Our data propose that young relatively small plant stages lack constitutive defense necessitating an active JA defense response. As plants mature constitutive defense metabolites seem to accumulate providing a higher level of basal resistance. The results obtained have important implications for cultivation of strawberry showing that both vertical integration of different pest management tactics as well as horizontal integration controlling several different pests concurrently, can be realized. Regular application of elicitors during the crop cycle forms a promising pest management tool as part of a sustainable IPM strategy not only in strawberry and other long-term cultivation crops but in production greenhouses in general.

SE42 007

SUCCESSFUL CASES OF ORGANIC CROP PROTECTION INTERVENTIONS DEMONSTRATED THROUGH FARM SCIENCE CENTERS IN SOUTHERN INDIA

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Increasing realization on the unsustainability of high input inorganic agriculture coupled with growing demand among consumers for residue free food led to a shift in the thinking of policy makers and scientists from productivity to income centric approaches in agriculture integrating inorganic and organic methods of crop protection. The Krishi Vigyan Kendras (KVKs) translated as Farm Science Centers, have been popularizing

such organic crop protection interventions through assessment and demonstration in the southern states of India. Few such successful cases are presented in this paper. The organic package for tomato comprising of bio-fertilizers and bio-intensive strategies for pests and diseases recorded 15-35% and 20-25 % lower incidence of diseases and pests respectively while ensuring higher benefit cost ratio of 3.18 as against 2.17 in inorganic method in Chittoor district of Andhra Pradesh state. Farm Science Center of Medak district in Telangana state demonstrated successful management of fall army worm in maize utilizing neem oil, pheromone traps, application of soil, charcoal or entomopathogenic nematodes in leaf whorls which ensured higher net returns of Rs.7915 compared to chemical control. Rhizome treatment and soil application with *Trichoderma harzianum* brought down rhizome rot incidence in turmeric to 6.75% compared to 23.25% in chemical control and recorded 16.75 % higher yields as demonstrated by the Farm Science Centre of Erode district in Tamil Nadu state. The KVKs have also been facilitating aggregation of organic growers and linking them to markets to raise their bargaining power and to enable them to realize premium price for their specialized products. Formation of organic producer clusters, creating effective market linkages, value addition and creation of demand among consumers would be the future strategy of the Farm Science Centers of the region to take the organic movement forward.

SE42 008

TRICHODERMA AFROHARZIANUM: FROM BIOCONTROL CHAMPION TO EMERGING MAIZE PATHOGEN IN EUROPE

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Trichoderma afroharzianum is a ubiquitous species occurring worldwide. Due to its mycoparasitic and endophytic properties, certain *Trichoderma* isolates are used in agriculture as biological plant protection and biocontrol agents. However, in 2018, a massive occurrence of *T. afroharzianum* on maize cob was observed for the first time in Germany. Since then, *Trichoderma* ear rot has been observed at several locations in Germany, France, and Italy, especially after dry and hot vegetations periods. Symptoms of *Trichoderma* ear rot are shown as massive production of green to gray-green conidia on the kernels and husk leaves. Moreover, affected crops are soft rotten characterized by excessive moisture and low starch content due to the production of alpha-amylase by the fungi. This results in a significant reduction in cob weight and quality with a reduced germination rate. Furthermore, it was observed that not only *T. afroharzianum* can infect maize but also certain *Trichoderma* strains from approved biological fungicides and soil additives have demonstrated pathogenicity after artificial inoculation. Out of the 14 tested products, three strains of biofertilizer and one strain from an approved biofungicide exhibited medium to high pathogenicity, similar to pathogenic reference strain. These findings suggest that certain strains commonly used in agricultural products for their beneficial properties may also possess pathogenic characteristics. In additions, greenhouse and field experiments has shown that *Trichoderma* is capable of infecting other cereals



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beyond maize like wheat and barley leading to discolorations and reduction of thousand-kernel weight. This expanded host range underscores the potential threat posed by *Trichoderma* infections and highlighting the importance of assessment and monitoring in the selection and application of microbial agents in agriculture.

SE42 009

TAXONOMIC STUDY OF ASSASSIN BUGS (REDUVIIDAE: HOMOPTERA) FROM PUNJAB, PAKISTAN

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Insects are elongated narrow head-like structures belonging to the family Reduviidae. They are known as Assassin bugs. Their tubular mouthparts are most commonly called the proboscis, but some authors use the term rostrum. They feed on a diverse variety of insects including flies' mosquitoes roaches' beetles aphids and caterpillars. They can also feed on beneficial insects like the Ladybird beetle. A bug improves its hunting success by slathering itself in the sticky resin of grass, in a rare example of tool use by insects. A total of nine Assassin bug species viz., *Phymata americana*, *Emesaya brevipennis*, *Triatoma infestans*, *Arlus cristatus*, *Lopodytes Rondani*, *Apiomerus flaviventris*, *Platyeris rhadamanthus*, *Stenolemus bituberus* and *Z. tetracanthus*, were recorded from the adult specimens collected from different plants. Assassin bugs can occur on almost any terrestrial plant including tree crops gardens and landscapes. These specimens were collected from Faisalabad, Multan, Rahim Yar Khan, Sargodha, Lahore, Rawalpindi, Mianwali, Layyah, and Dera Ghazi Khan respectively. These species differ in antennae, color, size, margin, submarginal, dorsal disk, transverse moulting suture, caudal furrow, and vasiform orifice. Shannon-Weiner Index value for richness is a maximum of 1.13 at Faisalabad followed by Sargodha, Rahim Yar Khan, Rawalpindi, Multan, Mianwali, Lahore, Dera Ghazi Khan, and Layyah respectively. A key for the Assassin bug species which are parasitizing different insects in Punjab is also prepared.

SE42 010

OZONATED WATER APPLICATION AS PROMISING TOOL FOR SUSTAINABLE PLANT PATHOGENS MANAGEMENT IN (EDIBLE) FLOWER PRODUCTION

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Ozonated water (OW) is regarded as an innovative approach in managing plant pathogens in pre- and post-harvest. Ozone dissolves in aqueous solution generating reactive oxygen species, allows to produce safe food-products and it is particular suitable where zero chemicals residues are necessary, as in the case of edible flowers. Here, OW was tested on two pathosystems, i.e. *Botrytis cinerea*/*Begonia hybrida* and *Erysiphe buhrii*/*Dianthus chinensis*, in order to evaluate the *in vitro* and *in vivo* antifungal effects on both fungal species, and

screen plant defence responses in terms of phytohormones/ signalling molecules [i.e. ethylene, salicylic, jasmonic and abscisic acids (Et, SA, JA and ABA)]. *In vitro* tests revealed a consistent inhibitory effect of OW on mycelium growth of *B. cinerea* on potato dextrose agar (~50%, in comparison to controls) and a high reduction of conidia germination of *E. buhrii* (~60%). The *in vivo* experimental trials lasted two and four weeks for *B. cinerea*/*B. hybrida* and *E. buhrii*/*D. chinensis*, respectively; pathogens were inoculated through foliar spraying of conidia suspensions (10⁵ conidia mL⁻¹) and a pot irrigation supplemented with OW (400 ppb) was provided. Few hours after the inoculation, *B. hybrida* OW-irrigated plants showed the early activation of the Et-JA mediated response (+33 and +47%, respectively), while SA and ABA increased in *D. chinensis* ones (+70 and +80%, respectively). At the end of the experiments, OW resulted effective in reducing the intensity/incidence of symptoms (brown spots and soft tissues) associated to *B. cinerea* in *B. hybrida* petals (~30%, in comparison to controls) and the *E. buhrii* mycelium spots on leaves and floral calyxes of *D. chinensis* (~45%). Additionally, no colour or morphology alterations due to OW application were observed. The obtained data indicate the potential of this sustainable technique in fungal pathogens containment in edible flowers.

SE42 011

CHARACTERIZATION AND MANAGEMENT OF ROOT DISEASES OF GREENHOUSE CUCUMBERS USING CULTURAL AND BIOLOGICAL METHODS

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Cucumber a widely grown vegetable crop in Oman. It is mainly cultivated in greenhouses under soil-based systems. Damping-off and wilt symptoms are among the most common diseases of cucumber in greenhouses. This study focused on identifying the causal agents of cucumber damping-off and wilt diseases and testing the effect of cultural and biological methods in reducing disease levels. *Pythium aphanidermatum* was the most common pathogen associated with cucumber damping-off and wilt symptoms, followed by *Rhizoctonia* and *Fusarium*. Solarization in summer helped reduce disease levels significantly. Screening fungal and bacterial isolates against *Pythium*, *Fusarium* and *Rhizoctonia* species showed the efficacy of some fungal isolates and bacterial strains in reducing pathogen growth and disease levels. Fungal species belonging to *Talaromyces* and *Trichoderma* and bacterial strains belonging to *Pseudomonas* and *Bacillus* species were found effective in inhibiting growth of the soil-borne pathogens in culture. The antagonistic microorganisms affected hyphae morphology and spore production of the pathogenic species. The antagonistic microorganisms also suppressed damping-off and wilt symptoms and resulted in improved growth and yield of cucumber. The study shows that several pathogenic species can be associated with cucumber damping-off and wilt symptoms. Some fungal isolates and bacterial strains from the Omani environment were found effective in reducing fungal growth and disease levels.

SE42 012

THE IMPACT AND DISSIPATION OF BIOCONTROL PRODUCTS IN SOIL MICROCOSMS USING AN UNTARGETED METABOLOMIC APPROACH

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Over the last three decades, the imperative to increase global agricultural production has precipitated significant escalation in chemical pesticide use. Nonetheless, these pesticides engender myriad adverse effects on health and environment. Consequently, biological products have emerged as promising alternatives for reducing application of chemical pesticides. Generally, biopesticides exhibit chemical complexity and frequently lack comprehensive characterization, rendering conventional methodologies (e.g., radiolabeling) unsuitable for monitoring biopesticide compounds and by-products. In this context, how persistence and impact of biopesticides could be evaluated? In recent years, the untargeted metabolomics approach has surfaced as a valuable tool for detecting and identifying low molecular weight metabolites (<1500 Daltons). Based on these developments, a new approach was developed, the Environmental Metabolic Footprint (EMF). The EMF is devised to explore, throughout kinetics, the comprehensive meta-metabolome, encompassing both the endometabolome (soil micro-organisms) and xenometabolome (products and by-products). This method is well-suited for investigating chemically complex biocontrol compounds, even those lacking characterization. Indeed, the untargeted approach enables the detection of unknown compounds. On another hand, investigating the endometabolome permits acquiring information regarding the environmental impact of the biopesticide. Monitoring the meta-metabolome through the time will allow obtaining a new proxy called the resilience time that corresponds to time needed for the biopesticide and its by-products dissipation as well as its impact on the matrix. Another objective of this approach lies in segregating the endometabolome and xenometabolome through bioinformatic processing that is a real challenge. It will permit, by targeting only the biopesticide product, evaluating its overall dissipation over the kinetics (dissipation time). To exemplify this approach, we propose to study the impact and fate in soil microcosms of a biofungicide extracted from a plant, *Inula viscosa*. Notably, the EMF approach can be applied to various biological samples, including soil, plants, fruit, sediments, to monitor resilience time and dissipation time.

Environmental Metabolic Footprinting



SE42 013

INCORPORATING OMICS AND ARTIFICIAL INTELLIGENCE TO EXPLORE AGRICULTURAL POTENTIAL OF LACTOCOCCUS LACTIS AS BIOCONTROL AGENT FOR SUSTAINABLE RICE FARMING

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Lactococcus lactis (*L. lactis*) is a prominent lactic acid bacterium in the food industry, known for its safety and diverse applications. Interestingly, it harbors untapped agricultural potential, yet a detailed understanding of its plant growth-promoting functions is scarce. This study aimed to isolate *L. lactis* from the rhizosphere of *Ocimum basilicum*, and explored its genomic and metabolomic profiles within a rice pathosystem. Among all strains isolated, RBX7 displayed plant growth-promoting (PGP) properties such as phosphate solubilization and indole-3-acetic acid (IAA) synthesis, but it lacked nitrogen fixation ability. This strain were inhibitory key rice pathogens *in vitro*, namely *Xanthomonas oryzae*, *Pantoea ananatis*, and *Curvularia lunata*, and maintained rice leaf health despite pathogen challenge. Scanning Electron Microscopy analysis showed disrupted pathogens' cell membrane integrity following RBX7 treatment. Metabolomics revealed differentially abundant metabolites likely contributing to the inhibitory effects against the pathogens. Whole Genome Sequencing of RBX7 resulted in a nearly complete genome, which was then subjected to in-house machine-learning (ML) models based on Protein Families (PFAM) features for predicting PGP and pathogenic traits. The ML models predicted RBX7 to have PGP characteristics and lack pathogenic property. Phylogenomic analysis distinguished the rhizosphere-derived RBX7 from its dairy counterparts, implying specialized adaptations for thriving in soil and plant environments. Genome mining results aligned with the *in vitro* findings, yet it could not pinpoint the operon accountable for IAA production. Conversely, the detection of an intact Nisin Z operon, coupled with the lack of virulence or antibiotic resistance markers, emphasized RBX7's suitability for agricultural use. In conclusion, the multi-omics strategies integrated with artificial intelligence in this research highlighted the potential of *Lactococcus lactis* RBX7 as a bio-effective agent for sustainable rice agriculture. Future efforts will focus on meta-transcriptomic and proteomic analyses to decipher the mechanisms and identify specific proteins responsible for the antimicrobial actions.

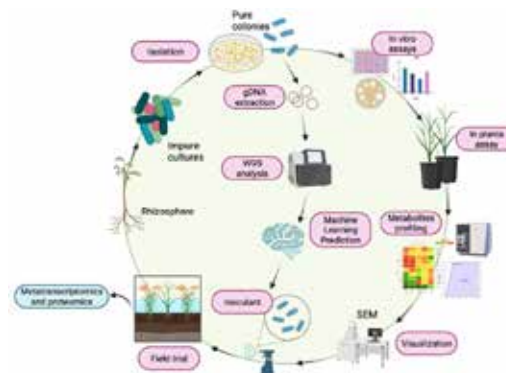


Figure 1: Exploring the potential of *Lactococcus lactis* as biocontrol agent for controlling rice pathogens using genomics, metabolomics and artificial intelligence



MC3

11.30-14.00 Concurrent Session 408

Recent Advances in Vegetable IPM

Chair: Ricardo Oliva (World Vegetable Center, Taiwan)

SE40 C04

THE POTENTIAL OF TRAP CROPS AGAINST LEAFHOPPER (*AMRASCA DEVASTANS*) AND WHITEFLY (*BEMISIA TABACI*) ON EGGPLANT UNDER FIELD CONDITION IN THAILANDSopana Yule¹, Chuanpit Khumsuwan¹,Paola Sotelo-Cardona², Ramasamy Srinivasan²

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Leafhopper (*Amrasca devastans*) and whitefly (*Bemisia tabaci*) cause severe loss on eggplants through hopper burn and eggplant yellow mosaic virus (EYMV), respectively. Eggplants are commonly used as a trap crop against both pests, whereas limited research has revealed efficient trap crops against these pests when eggplants are grown as cash crops. This study aimed to validate the trap crops (okra and sunflower) combined with sticky traps and insecticides against these pests under the field in two seasons. Seven treatments, including T1 - trap crop (TC) alone; T2 - yellow sticky traps (YST) alone; T3 - TC + YST; T4 - TC + chemical pesticide (Ch); T5 - YST + Ch; T6 - chemical pesticide ; T7 - untreated control (C) were tested in on station experiment at Nakhon Pathom, Thailand, following the Randomized Complete Block Design (RCBD) with three replications. In the wet season, the results showed that the leafhopper density on leaves treated with was lower than the control. Still, hopper burn intensity (HBI) was similar among all treatments. Low whitefly densities (egg and pupa) were detected in YST, and pupa and adult in TC, and TC + YST. The lowest EYMV infection was in YST, followed by TC + YST, YST + Ch, TC + Ch, and Ch. The greater marketable yields were detected in Ch, followed by YST + Ch and TC + Ch. In the hot season, low leafhopper density on leaves in all treatments was shown compared with control, while reduction of HBI was found in TC + Ch, YST + Ch, and Ch. Whitefly populations during the nymphal-adult and nymphal-pupa stages were significantly low in YST, TC, and TC + YST, respectively. This study revealed that TC and YST have the potential to decrease pest density, damage of HBI and EYMV, and increase yield.

parts of Himalayan states in the country. The basic aims of the studies were effective management of various pests and diseases of tomato and reduce the number of application of chemical pesticides. The major pest and disease cause serious economic damage; are : early and late blight disease, fruit rot and fruit borer. Based on the investigations, it revealed that these pests and diseases can be managed effectively by adopting pest and disease monitoring, their proper identification, level of incidence, selection of appropriate chemical pesticides, preparation of solution and dosage and right time of application. The results of the field trials envisaged that application of Azoxystrobin + Tebuconazole @ 1 ml/l water has been found very promising against early and late blight disease. Similarly, Chlorothalonil @ 1 gm/l water showed excellent result in management of fruit rot disease. Application of Emamectin benzoate (Biological insecticides) @ 0.5 gm/l water was considered as most promising insecticide against fruit borer. Implementation and demonstration of these integrated pest management (IPM) practices in tomato enhanced the yield by 98.12% and numbers of application of chemical pesticides have been reduced by 60%. The findings of the field trials also advocate that quality of the tomato also improved significantly due to healthy plants because of effective management of different pests/diseases and very less use of chemical pesticides. The tomato fruits harvested from the field trials received premium price in the markets due to high quality which is also beneficial for human health and their welfare. Considering the significance of tomato in economic empowerment; it is strongly recommended to the farmers for large scale adoption of above IPM practices.

SE40 003

NUTRITIONAL MANAGEMENT AND PREDICTION OF TOMATO LEAF CURL VIRUS DISEASE

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Tomato leaf curl virus (TLCV) is a notable constraint for the cultivation of tomato worldwide. It causes 70-100% crop losses in case of severe attacks. Whitefly *Bemisia tabaci* (Genn.) transmits TLCV persistently and circulative. As no viricides are available, different insecticides are commonly used for the management of TLCV disease. Frequent use of insecticides creates serious environmental issues and resistance in insects. In present study, plants were sprayed with aqueous solution of nutrients and salicylic acid as an eco-friendly approach for the management of TLCV. Furthermore, a disease predictive model was developed based upon environmental factors to find out the possibility of disease outbreaks. Predictive modelling is an efficient tool for accurate TLCV disease management. TLCV disease predictive model was formulated on the basis of weather

SE40 002

PEST AND PESTICIDE MANAGEMENT IN TOMATO UNDER MOUNTAIN REGION OF INDIA

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Tomato is commercially important vegetable crops extensively grown in mountain region of India as an off season. According to an estimate 18-70% loss in yield has been recorded in different

variables through regression analysis. Nutrients and salicylic acid gave significant reduction in TLCV disease incidence i.e. 49% and 51%, respectively. A disease predictive model based on 2 years of ecological and meteorological data was formulated $y = 0.541 + 0.049x_1 + 0.91x_2 - 0.079x_3 + 0.11x_4$ $R^2 = 0.85$. This would aid the farmers to identify, assess and select appropriate disease management methods.

SE40 004

INTEGRATED PEST MANAGEMENT OF BEMISIA TABACI ON TOMATO IN CULTIVATION FACILITIES IN TAIWAN

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The whitefly, *Bemisia tabaci*, is a worldwide pest which damage tomato by transmitting viruses and causing tomato yellow leaf curl disease. *B. tabaci* is regarded a key pest affecting the quality and yield of tomato in facility in Taiwan and the incidence of virus of tomato plants will reach more than 70% if the vectors are not controlled. In this study, we recommended to hang 10 yellow sticky traps on plants per acre to monitor the densities of whiteflies, and control measures should be applied when 50 adult whiteflies in average were caught per sticky trap per week. Furthermore, the parasitoids, *Eretmocerus orientalis* of *B. tabaci* were regularly released to control whiteflies. The results showed that the incidence of viral disease of tomato plants were less than 10% in the two experimental locations. A strategy of *B. tabaci* IPM has been proposed that whiteflies should be thoroughly controlled to prevent them from spreading the viruses in the early stages of tomato cultivation. In addition, eradicating plants infected with viruses and releasing natural enemies can suppress the occurrence of whiteflies. This approach helps reducing pesticide usage while maintaining tomato yield and quality, thereby achieving the goal of safe production.

SE40 005

NICOTIANA BENTHAMIANA AS DEAD-END TRAP PLANT FOR WHITEFLY CONTROL

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1. Zhejiang University, Hangzhou, China

The whitefly, *Bemisia tabaci* is destructive for the production of several important greenhouse vegetables including tomato. It is hard to control this tiny insect pest as its high resistance to most insecticides. Thus alternative methods such as biological control and the application of functional plants may play vital roles in integrated pest management of whitefly. In the laboratory, we observed LAB strain of *Nicotiana benthamiana*, a commonly employed laboratory model plant, demonstrated remarkable lethality and attractiveness towards Hemiptera insect *B. tabaci*. In this study, through various laboratory and field assays, we explored the potential of using *N. benthamiana* as dead-end trap plant for whitefly control. We found *N. benthamiana* demonstrated

nearly 100% lethality to whiteflies, with emitted volatiles attracting them. Moreover, field trials showed that potted and planted *N. benthamiana* blocked and trapped whiteflies from several Solanaceae crops effectively, comparable to common commercial yellow sticky traps. No negatively impacts were found on the natural predator *Nesidiocoris tenuis* which implied the good compatibility of this companion plant with biological control agents. This study unearths a new benefit of *N. benthamiana* and develops an easy-to-apply and sustainable strategy for field whitefly control.

SE40 006

DEVELOPMENT AND VALIDATION OF A PLANT DISEASE MODEL FOR VENTURIA OLEICOLA IN THE FRAMEWORK OF INTEGRATED PEST MANAGEMENT

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1. Horta Srl., Spain

Olive scab is the main disease affecting olives worldwide. It is caused by the fungus *Venturia oleagina* (synonym *Spilocaea oleagina*), which can infect over a wide range of temperatures and long wet periods. Symptoms appear after a latency period that can vary from 10-14 days up to months, depending on host and environmental conditions, and this makes disease management challenging. Traditional olive orchards are commonly treated on spring and autumn, whereas intensive olive productions, with a higher disease pressure, apply fungicides on a calendar basis. Both disease control strategies are not compliant with concepts and principles of integrated pest management (IPM), which promotes reasoned interventions based real infection risk. In this work, we show a process-based model for *V. oleagina*, which considers the current scientific knowledge on the epidemiology of the disease and predicts the sporulation, dispersal of conidia and infection based on environmental conditions. We also show some model validations, even though validation of *V. oleagina* models is challenging due to the uncertainty concerning the length of latency period; the validation was carried out in Italy, in 2020, by using 32 batches of trap plants which were exposed to natural inoculum in an affected orchard and replaced after each rain event. Results indicated that the model was able to correctly predict the infections events with 85% accuracy. The model has been implemented in the decision support system Agrigenius Olive, a digital tool designed to support the holistic and sustainable management of the olive grove, having a multi-modelling approach covering all the key cultivation aspects (plant growth, pests and diseases, fungicide protection dynamics, abiotic stresses, nutrition, weeds).

SE40 007

THE SPRING WHEAT IN QINGHAI SERVES AS AN IMPORTANT OVER-SUMMERING LOCATION FOR THE STRIPE RUST IN CHINA

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Wheat yellow rust, caused by *Puccinia striiformis* f. sp. tritici (PST), is one of the biotrophic pathogen. The occurrence can



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lead to devastating consequences for wheat. Gansu serves as a reservoir during non-cropping periods and provides PST inoculum to wheat both in fall and spring, while Qinghai serves as a region providing huge amounts of over-summering inoculum. The inoculum exchange of yellow rust and migration between these two regions is essential to ensure their perpetual existence. To understand this relationship, we systematically studied the genetic diversities, seasonal population dynamics, recombination, and gene flow of wheat yellow rust populations in different over-summering areas of Gansu and Qinghai using molecular markers combined with a spatiotemporal sampling strategy. Shared genotypes provide molecular evidence of migration between the pathogen populations in Gansu and Qinghai. Genotypic frequencies show that the pathogen mainly flows from Qinghai to Gansu in autumn, while the reverse movement from Gansu to Qinghai occurs in spring. The inoculum source on spring wheat can be directly transmitted to autumn seedlings, not necessarily through volunteer wheat. Therefore, the bridging effect of spring wheat may play a more important role than the off-season pathogen surviving on volunteer wheat. Furthermore, linkage disequilibrium tests indicate that sexual recombination continues throughout the year in the Tianshui and Dingxi regions of Gansu.



CONFERENCE I

11.30-14.00 Concurrent Session **43**

Approach to Integrated Soil and Plant Health Assessments in IPM Systems

Chair: Dima Alnajar (Syngenta Group, Switzerland)

SE43 001

ON-FARM EVALUATION OF CROP DIVERSIFICATION ON PEST MANAGEMENT AND SOIL FERTILITY IN MAIZE-BASED FARMING SYSTEM

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Pest outbreaks are rare in polycultures due to the ability of the diverse plant culture to self-sustain through natural pest control by increasing the occurrence of natural enemies. A study was conducted with the aim of investigating the effect of maize farm diversified with bean and desmodium on pest protection and soil fertility improvements. A field trial was conducted in Ethiopia to compare the intercropping performances of maize-common bean-desmodium; maize-desmodium; maize-common bean and sole maize/control. Maize stem borers and fall armyworm infestation ranged from 3.9 to 40.3% with the highest infestation in the control, followed by the common bean and desmodium intercrops. The lowest maize infestation (3.9%) was recorded in the maize intercropped with common bean and desmodium. The highest yield was recorded in the maize-common bean-desmodium intercropping (8.06 t ha⁻¹) followed by the maize-desmodium intercropping (7.1 t ha⁻¹), while the lowest yield occurred in the sole maize/control (5.43 t ha⁻¹). Crop diversification significantly altered the physicochemical properties of the soil. The soil organic carbon, organic matter, total nitrogen, available phosphate, and potassium in all maize-common bean and desmodium intercropping was significantly higher than that of the control (sole maize). This study demonstrated that diversified farming systems use synergies from mixed farming systems that improve soil fertility and pest control.

SE43 002

SYNGENTA CROPWISE® PROTECTOR DISEASE RISK ASSESSMENT MODEL FOR CEREAL CROPS

Alnajar D, Crockford A, Blanc F, Godet V, Humphrey J, Amokrane Y, Varraillon T

1. Syngenta Group, Basel, Switzerland

Fungal diseases in cereal crops lead to significant reductions in both yield and grain quality. Informed disease management decisions, supported by data and digital models, are essential components in the implementation of Integrated Pest Management (IPM) strategies. Predicting cereal diseases is challenging and complicated by several factors, such as the erosion of genetic resistance in varieties, the rise of

pathogen resistance to fungicides' various modes of action, climate conditions conducive to pathogen proliferation, and suboptimal fungicide application practices. To address this complexity, the process-based model in Syngenta Cropwise® Protector synergizes a crop phenological model with disease progression and fungicide efficacy models. The aim of this study is to showcase how the Cropwise® Protector disease model functions to predict yellow rust, brown rust, septoria leaf blotch, powdery mildew, eyespot, and fusarium head blight of wheat. Growers provide the planting date, crop variety, previous crops, and agronomic practices, namely tillage and nitrogen fertilization, while the weather conditions are fed to the model by in-field weather stations or from a gridded weather source. The model predicts the daily disease risk on a per-leaf-layer basis starting from the growth stage BBCH30. Based on a weekly data collection from BBCH30 to BBCH65, 7-year results indicated that the Cropwise® Protector disease model predicted disease risk with over 85% accuracy in France in the disease complex of wheat. Fungicide application timing was recommended with no more than 5-10% risk of delay by targeting the incubation phase of the disease development. Consequently, the Cropwise® Protector disease model empowers growers to make timely and informed decisions for proactive disease management. Overall, this disease model stands as a cutting-edge digital agronomy tool that not only aids growers in making better decisions but also promotes environmental stewardship and sustainable practices in agriculture.

SE43 003

ANALYSIS OF UTILIZATION OF CROP ROTATION IN MAIZE AND BEANS FOR HEALTHY PLANTS AMONG FARMERS IN NORTH CENTRAL NIGERIA

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Practice of maize and beans rotation among farmers in North Central Nigeria with an estimated population of over 22 million people. The study specifically sought to describe the socio-economic characteristic of beneficiaries of indigenous knowledge of maize and beans among farmers in North Central Nigeria, determine knowledge utilization of crop rotation practice in North Central Nigeria. A sample of 300 respondents was selected using stratified, purposive and simple random sampling technique. Two hundred and ninety nine (299) out of 300 questionnaires were retrieved and used for analysis. Descriptive statistics such as frequency, percentage and mean score were used. The pooled results showed the age of the respondents were sixty year and below



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(95.0%), married with household to be between 1-8 persons. Most (87.0%) had farm size range 0.1 - 4.99 ha (small size). 94.3% had farming experience 1 - 10 years, 38.1% had tertiary education while 10.9% had no formal education, 79.6% had income N51,000 - N100,000 per annum described as poor income. 78.6% that had extension visit of previous year was as low as 1 - 5 times. More than half of the respondents resided in rural areas, mean score was highest than cut-off mean for knowledge utilization process by the respondents. The study concluded that most farmers utilized the indigenous knowledge practice of maize - bean rotation. It was recommended that stakeholders in knowledge creation guarantee involvement of more farmers for ultimate utilization of indigenous knowledge to protect crop achieve higher leaf area index to enhance the welfare of farmers in North Central Nigeria.

BANQUET

15.00-17.30 Concurrent Session **44**

Plant Protection Products

Chair: Richard Peters (Agriculture and Agri - Food Canada)

Giulia Scimone (Department of Agriculture, Food and Environment, University of Pisa, Italy)

SE44 001

A NEW YEAST-DERIVED PRODUCT IN PLANT PROTECTION TRADE: BOOSTING GRAPEVINE ECO-FRIENDLY WEAPONS AGAINST BOTRYTIS CINEREA AND PLASMOPARA VITICOLA INFECTION

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Necrotrophic and biotrophic pathogens represent a persistent challenge for plant development/growth. To counteract their impact, plants have evolved a fine defense system, which takes advantage of a complex compounds network. This study investigated the alerting mechanisms induced by the application of a new yeast extract (YE; supported by Kwizda Agro GmbH, Wien) formulate on *Vitis vinifera* cv. Sangiovese plants inoculated with *Botrytis cinerea* (Bc) or *Plasmopara viticola* (Pv). The YE was applied once a week for three consecutive weeks on grapevines (YE⁺), while control plants were treated with sterile water (YE⁻). Eight plants per set were foliar sprayed with Bc or Pv (1×10^5 conidia mL⁻¹; Bc⁺ or Pv⁺), while the remaining ones were inoculated with sterile medium (Bc⁻ and Pv⁻). Three fully-expanded leaves per plant were sampled from 3 h post inoculation (hpi), used fresh for ethylene (Et) determination, and immediately frozen for jasmonic and salicylic acid (JA and SA), hydrogen peroxide (H₂O₂) and malondialdehyde by-products (MDA) analyses. Results showed an activation of Et and JA signalling pathways in both YE⁻/Bc⁺-YE⁻/Pv⁺ leaves (more than 4-fold higher and +45% as average, in comparison to controls) and YE⁺/Bc⁺ (9-fold higher and +35%) at 24 hpi, while unchanged values resulted in YE⁺/Pv⁺ ones. Salicylic acid content decreased in both YE⁻/Bc⁺ and YE⁺/Bc⁺ leaves at 3 hpi (-33 and -58%), did not show any changes in YE⁻/Pv⁺, while reached a maximum in YE⁺/Pv⁺ ones (+79% at 72 hpi). The YE application per se (YE⁺/Bc⁻ and YE⁺/Pv⁻) induced a production of JA, SA and H₂O₂. Unchanged MDA values (except in the case of YE⁻/Bc⁺) suggested the activation of antioxidative mechanisms able to counteract oxidative pressure in all YE⁺ plants. These results confirm the potential of this new eco-friendly product to enable grapevines to better invest resources and protect themselves against important pathogens.

SE44 002

TANYMECUS DILATICOLLIS, A MAJOR PEST OF THE MAIZE CROPS IN ROMANIA

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Maize leaf weevil (*Tanymecus dilaticollis* Gyll) is one of the most dangerous pests of the maize crops in Romania. Every year, more than one million hectares cultivated with maize were attacked, especially in the south and southeast of Romania. Weevil's attack occurred at the beginning of the maize vegetation period. In the absence of adequate control measures, this pest can destroy plants. After the ban of the neonicotinoids in the European Union, no alternatives remain available in Romania for maize seed treatment to control this pest. This study aims to find alternatives to neonicotinoids for controlling maize leaf weevil attacks. The experience was carried out from 2021 to 2023 in the most favorable area for this pest, located in the southeast of Romania (Mihail Kogălniceanu, Ialomița County). This study has tested foliar treatment with acetamiprid and lambda-cyhalothrin active ingredients, seeds treatment with cyantraniliprole, and granules application with lambda-cyhalothrin. Foliar treatments were applied after maize emergence when plants were in BBCH 11-12 stages, while treatment with granules was applied at the same time as the sowing of maize. It assessed plant density and pest attack when maize was in the BBCH 14 stage. In 2021, a lower maize leaf weevil attack was on plants with cyantraniliprole seeds treatment, but the maize had leaves consumed in the proportion of 50-75 %. Foliar or granule treatments didn't reduce the attack. In 2022, there was a high pest pressure, and all plants from this study were destroyed. In 2023, seed treatment with cyantraniliprole as an active ingredient reduced the attack compared with control. However, the plants have leaves consumed in the proportion of 75-90 %, while most of the plants were destroyed at other treatment types. In the case of high pest pressure, no alternatives for seed treatment with neonicotinoids were found.





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SE44 003

MANAGEMENT OF WHITE MOLD OF POTATO WITH FOLIAR FUNGICIDES

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White mold of potato, caused by *Sclerotinia sclerotiorum*, can cause significant yield reductions in commercial crops. Typically, infection of potato flower petals leads to infection of foliage when infected petals fall into the canopy and lodge there. Infection of leaves and stems leads to cracking and breaking of stems with resultant yield loss. Protecting flower petals from infection therefore, becomes a reasonable management option for growers. Various foliar fungicides (and a water control) were applied at 10% crop flowering and again 2 weeks later in a trial established in a field with a known history of white mold over two field seasons in Prince Edward Island, Canada. Plots were evaluated for the number of stems showing symptoms of white mold at various points during the growing season, until harvest. Plots were harvested and tubers graded for yield and quality. During the growing season, the applied foliar treatments did not cause phytotoxicity or affect plants adversely. White mold was prevalent in the trial which allowed excellent evaluation of foliar treatments for disease control. All fungicides provided significant disease suppression compared to the untreated control. In general, combinations of fluopyram/pyrimethanil, pydiflumetofen/fludioxonil and fludioxonil alone provided excellent disease suppression, while a combination of pydiflumetofen/difenoconazole and fluazinam alone offered somewhat lesser but similar significant disease suppression compared to the control, with close to 50% reduction in number of infected stems. A combination of benzovindiflupyr/difenoconazole was generally less effective than the other fungicides at managing disease in the crop. White mold, although prevalent, was not severe enough to affect yields in the trials, and no significant yield differences were found among treatments. Foliar fungicides, when applied at the proper timing at early crop flowering, have the potential to significantly reduce white mold in potato crops.

SE44 004

EFFECT OF PEST CONTROL STRATEGIES ON EUROPEAN ARTHROPOD PESTS OF APPLE: A META-ANALYSIS

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About 60 arthropod species are considered to be economically important pests in European apple orchards. Pest management has become an important issue in apple production due to increasing consumer demand for environmentally friendly food products with high aesthetic standards and ongoing policies to make EU fruit production more sustainable. The aim of these meta-analyses was to determine the most effective management strategy against arthropod pests of apples in Europe. We conducted Europe-wide meta-analyses based

on a systematic literature review to determine how effective Integrated Pest Management (IPM) and organic management are in reducing arthropod pest numbers in apple orchards. We also looked at the differences between synthetic and organic pesticides in terms of pest control as a key difference between IPM and organic apple production systems in Europe. The overall mean effect size for each comparison was estimated using random effects models. IPM did not significantly reduce pest numbers if compared to organic management. Organic apple production systems partly depend on alternative approaches to pest management and these can be comparable in effectiveness to IPM strategies. Synthetic pesticides significantly reduced pest numbers compared to organic pesticides, although there was significant heterogeneity in the effect sizes calculated. For example, the organic pesticide Spinosad performed better than some synthetic pesticides in the studies reviewed. Organic pesticides significantly reduced pest numbers compared to no pesticide treatments. The meta-analyses show the relative efficacy of organic pesticides, taking into account the inherent heterogeneity within each dataset. The results highlight the importance of alternative pest control approaches to synthetic pesticides, but also document the relatively high efficacy of these pesticides. Decisions regarding pesticide use, whether based on monitoring and thresholds under IPM regulations or alternative approaches under organic certification, should not be based primarily on pest control concerns.

SE44 005

RNA INTERFERENCE TECHNOLOGY: A NEW PATH FOR THE RESEARCH AND MANAGEMENT OF THE OBLIGATE BIOTROPHIC PHYTOPATHOGENIC FUNGUS *PODOSPHAERA XANTHII*

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The cucurbit powdery mildew disease, caused by *Podosphaera xanthii*, represents some of the most destructive plant diseases worldwide. Currently, chemical control and resistant cultivars are the primary approaches for disease management. However, fungicide-resistant isolates and new races of the pathogen make its control even more challenging, being necessary the development of innovative strategies. Chitin is a crucial component of fungal cell walls and an effective inducer for plant immunity, thus, phytopathogenic fungi have developed virulence factors to suppress this defensive response. In this study, the molecular machinery of chitin-triggered suppression through the effectors involved in the modification of chitin immunogenic oligomers (CDA) and in their degradation (EWCAs), were used as targets for the design of new antifungal strategies. To test this, an

RNA interference (RNAi) technology, which consists of the application of double-stranded RNA (dsRNA) designed to suppress the expression of the corresponding target genes, was used. Initially, the uptake and processing of dsRNAs targeting the genes previously mentioned by RNAi machinery of melon plants through small RNA sequencing (sRNA-seq) was studied. Then, the *P. xanthii* uptake was explored through confocal microscopy visualization. Subsequently, these dsRNAs were infiltrated and sprayed on melon cotyledons and leaves, respectively, to evaluate fungal development and induce genes silencing. The results from sRNA-seq demonstrated that melon plants could process these dsRNAs at 24 hours post-spraying, while confocal images revealed the presence of fluorescent dsRNAs inside fungal spores after overnight application. Moreover, the RNAi strategy reduced by approximately 50% powdery mildew disease symptoms. Furthermore, by protecting these dsRNAs with “carbon dots” nanoparticles in greenhouse conditions, a significant prolongation of powdery mildew disease inhibition was observed over time, indicating its potential as an innovative and precise method for managing cucurbit powdery mildew.

This work is part of the R+D+i projects PDC2021-121373-C21 and PID2022-1362400B-C21, funded by MCIN/AEI/10.13039/501100011033/FEDER, UE.

SE44 006

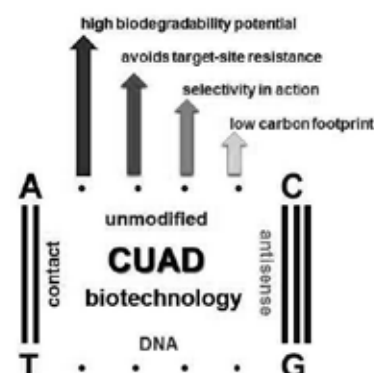
CONTACT UNMODIFIED ANTISENSE DNA (CUAD) BIOTECHNOLOGY: OLIGONUCLEOTIDE INSECTICIDES AS THE NEXT-GENERATION PLANT PROTECTION PRODUCT FOR GREEN AGRICULTURE

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In 2008, it was shown for the first time that antisense oligodeoxyribonucleotides can be used as contact insecticides. Oligonucleotide insecticides (briefly, olinscides, or DNA insecticides) as a new class of insecticides has its own peculiar characteristics. Olinscides are short unmodified antisense DNA fragments that use rRNAs of pests as target and act through DNA containment mechanism ('arrest' of target rRNA, block of functioning of ribosomes accompanied with hypercompensation of expression of rRNA and degradation of target rRNA by RNase H). Today, CUAD (contact unmodified antisense DNA) biotechnology is the only biotechnology that uses short unmodified antisense DNA oligonucleotides in plant protection. Unlike modern chemical insecticides, oligonucleotide insecticides use natural mechanism of action and are part of this mechanism. In addition to the high biodegradability potential, low carbon footprint, and selectivity in action, the opportunity of solving the problem of target-site resistance using oligonucleotide insecticides based on conservative gene sequences is expected. As a new class of insecticides, oligonucleotide insecticides can reduce the concentration of modern chemical insecticides (organic xenobiotics) in ecosystems. CUAD biotechnology is a very efficient eco-friendly platform against hemipteran insect

pests from Sternorrhyncha suborder (aphids, whiteflies, mealybugs, psyllids, etc.). Biodiversity decrease, climate change, high toxicity load in ecosystems dictate new rules for plant protection. It is obvious that oligonucleotide insecticides will be brought to market in abundance and will complement current classes of chemical insecticides. We envision that two next-generation classes of insecticides, oligonucleotide insecticides (CUAD biotechnology) and RNA biocontrols (double-stranded RNA technology), can be used solely and also can come into place in complex formulations for selective control of a wide range of insect pests and will mark a new marvelous period in efficient and green plant protection.



SE44 007

STUDIES ON THE PATHOGENICITY OF FUNGAL BASED BIO-PESTICIDE METARHIZIUM ANISOPLIAE VAR. ACRIDUM AGAINST DESERT LOCUST, SCHISTOCERCA GREGARIA

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The current study was carried out to test the efficacy of different entomopathogenic fungi (EPF) formulations i.e. *Metarhizium anisopliae* against the nymphs and adults of the desert locust, *Schistocerca gregaria* Forskål under field as well as laboratory conditions. Four different concentrations were applied against different instars of the nymphs and adults of desert locust. Susceptibility was found to be greatest in 3rd-instar nymphs, followed by 5th instars, and then adults. Along with lethal effects, sublethal doses of EPF reduced the number of egg pods/female, total eggs/pod, and egg hatching, while extending nymphal developmental time and reducing adult longevity; again, *Metarhizium anisopliae* performed well. Sublethal doses not only retarded reproduction, but also caused behavioral changes, like reduction in food consumption, fecal production, and weight gain. All EPF formulations not only showed significant mortality in laboratory conditions, but also performed very well under the field conditions. The maximum mortality against 3rd-instar (81.7% and 74.0%), 5th-instar (73.3% and 65.1%), and adult locusts (67.5% and 58.9%) was observed when using *Metarhizium anisopliae* under greenhouse and field trials, respectively. The current study showed that all of the EPF formulations have the potential to reduce pest populations, and could be used in the integrated pest management program.

MC2

15.00-17.30 Concurrent Session **45**

Xylella Fastidiosa

Chair: Anna Maria D'Onghia (Plant Protection, Ciheam Bari, Italy)

Maria Holeva (Benaki Phytopathological Institute, Greece)

SE45 C01

PRECISION SYSTEMS FOR MONITORING XYLELLA FASTIDIOSA IN OLIVE GROVES

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Monitoring *Xylella fastidiosa* (Xf) is a difficult task for any NPPO because this bacterium has different behaviors depending on the subspecies and strains involved, the number and type of host species and vectors, the latency period, and different environmental conditions. New frontiers of technology offer several smart solutions in the field of plant health, facilitating the task of operators in implementing more efficient, accurate, timely, and cost-effective large-scale pest monitoring programs. The challenge is therefore to develop integrated and multidisciplinary monitoring systems (remote sensing, metabolomics, GIS, information technology, statistics, modeling, forecasting, biotechnology, etc.), both at the land and grove levels, to: (i) rapidly identify Xf outbreaks, thereby guiding inspections and field sampling; and (ii) assess the rate of infection and its spatial and temporal spread, in order to take precision phytosanitary measures to control it and evaluate its effects in eradicating or containing the infection. An overview on RS, GIS, and IT applications in the Xf monitoring on olive trees is presented: (i) the photointerpretation of high-resolution aerial ortho-images for the identification of trees affected by the Olive Quick Decline Syndrome (OQDS); (ii) the Sentinel-2 images using the OSAVI index for the spatial & temporal evolution of the OQDS; (iii) the new spectranomic approach for early detection of the infection before symptoms development; (iv) the prediction maps at land scale; (v) the statistically-based sampling schemes at olive trees and olive groves; (vi) the applications for accurate field data acquisition by NPPOs (e.g., XylApp_{EU}, Ap³est) and citizens (XylApp_{citizens}); (vii) the rapid onsite diagnostic assays (real time LAMP, DTBIA); (viii) the precision monitoring systems.

SE45 C02

TEN YEARS OF IMPLEMENTATION OF LABORATORY TESTING TO PREVENT ENTRY OF XYLELLA FASTIDIOSA IN GREECE - RESULTS OF CURRENT DIAGNOSTIC EFFORTS

M. C. Holeva¹, C. Karafila¹, C. Reppa¹, I. Dervisi¹, P. Glynos¹, E. Siderea¹, A. Togias¹, G. Kolliopoulou¹, D. Perivolaris¹, L. Ntoulberis², S. Ioannidou², C. Arampatzis²

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Since the first detection of the devastating pathogen *Xylella fastidiosa* (Xf) in Europe (Italy) in 2013, annual surveys of domestic crops and phytosanitary border checks of plant material for import have been intensified in Greece regarding this pathogen. Such diagnostic checks are crucial for early detection of Xf in the country. To this end, human and technical resources have been allocated mainly via the official National Survey Program funded by the Hellenic Ministry of Rural Development and Food (HMRDF) and the European Union (EU). The Laboratory of Bacteriology of the Benaki Phytopathological Institute (LB-BPI), as the Reference Laboratory for bacterial phytopathogens in Greece (ISO-accredited for Xf detection), performs the official Xf diagnostic tests in plant material: a) collected by Phytosanitary Inspectors of the HMRDF in the framework of the surveys and phytosanitary border checks, and b) sent by growers and other citizens to the BPI plant clinic. The results are reported to the HMRDF and the EU, and after ten years of continuous diagnostic checks, Xf has not been found in Greece. Results of the current diagnostic efforts of the LB-BPI in the frame of the official testing and the European/national-funded projects: 'XF-ACTORS', 'CURE-Xf', 'EuroXanth', 'Olives Roads', are to be presented, with emphasis on: a) the use, in high-risk areas, of indicator plants exhibiting identifiable Xf symptoms at an early stage of infection, thus achieving a more targeted surveillance, b) optimization of Xf molecular detection by elimination of PCR inhibitors in plant DNA extracts, c) developing a multiplex real-time PCR protocol for detection of Xf and other quarantine prokaryotes in grapevine. Additionally, the LB-BPI participates in the European network of phytosanitary research programme (EUPHRESKO) aiming at developing new molecular Xf diagnostic methods. Such diagnostic approaches ensure the movement of Xf-free plant material and the early detection of an Xf outbreak.

**SE45 C03****THE CURRENT STATUS OF XYLELLA FASTIDIOSA IN THE EURO-MEDITERRANEAN REGION****M. Saponari¹, B.B. Landa²**

1. *Institute for Sustainable Plant Protection, National Research Council, Unit of Bari (Italy)*, 2. *Institute for Sustainable Agriculture, Spanish National Research Council, Cordoba (Spain)*

Since the first report of *Xylella fastidiosa* in the Old Continent, 11 years ago, the phytosanitary scenario has clearly evolved from a severe threat linked to a local outbreak emerged in southern Italy (Apulia region) on olives to the discovery of widespread endemic infections with apparent minor impact on landscape and crops. The epidemiological data so far built on the monitoring dataset collected in the Countries facing bacterial outbreaks show the relevance of the bacterial strains, crop susceptibility and landscape vulnerability in the evolution and severity of the infection outcomes. After the peak of alert raised by the case of *X. fastidiosa* subspecies *pauca* genotype ST53 in olives in Apulia region, numerous bacterial strains genetically distinct and with different host range have been reported in southern Europe, Israel, Lebanon and Iran. Most of these reports refer to old undiscovered introduction of the bacterium, from the American countries, whose impact has been neglected until a surveillance program was implemented in the last decade, leading to discover hitherto infections. Beside olive quick decline syndrome on olives, whose impact has been extensively documented, almond leaf scorch symptoms have been reported in Spain, Lebanon, Iran and Israel, while infections on grapes reported in Mallorca, Iran, Israel, Portugal and more recently in Italy, so far do not seem to cause severe impact as the notorious Pierce's disease in north America. Currently, numerous strains belonging to three major bacterial subspecies have been reported affecting more than 100 plant species in the EU and Mediterranean affected countries. The lack of effective control measures imposes to invest relevant resources on preventive strategies to protect *Xylella*-free areas from invasions, to early detect new outbreak, to ensure high quality standards of the propagating materials responsible for long distance dispersal of the bacterium and to reinforce phytosanitary barriers. On the other hand, the strategies for the management and containment of the bacterium in the infected areas should be revised considering the complexity of this bacterial species and the experience so far gained under the Mediterranean conditions.

SE45 C04**PHILAEUS SPUMARIUS: THE MOST POLYPHAGOUS INSECT HERBIVORE HAS BECOME AN UNFORESEEN VECTOR OF XYLELLA FASTIDIOSA IN EUROPE****Alberto Fereres**

Consejo Superior de Investigaciones Científicas. ICA-CSIC, Madrid, Spain

The meadow spittlebug *Philaenus spumarius* (L.) has become one of the best studied insect species in the last decade as it was reported to be the main vector in Europe of *Xylella fastidiosa*, a bacterium causing a devastating disease of olives (CoDiRO "Complesso del Disseccamento Rapido dell'Olivio") in southern Italy in 2013. This insect species was

very well known from centuries because of its unique color pattern polymorphism but never considered as a threat to agriculture before the outbreak of CoDiRO disease in Italy. Actually, the meadow spittlebug causes no direct damage to their host plants and is an extremely polyphagous species, being the species that exploits the greatest number of host plant species among any herbivorous insect. It feeds mainly in Asteraceae and Rosaceae but it can be found in at least 1,311 host plants belonging to 117 different plant families. During its juvenile stages it feeds on herbaceous dicots forming characteristic foams that can be easily found in many managed and unmanaged ecosystems. Adults, however may be present in both herbaceous and woody plants which facilitates the spread of *X. fastidiosa* among multiple hosts in any environment in which both the spittlebug and bacterium are present. In the last decade, an increasing number of scientific publications have concentrated on the biology, ecology and behavior of *P. spumarius*. New insights on the anatomy and physiology of the insect's mouthparts as well as on the transmission mechanisms of *X. fastidiosa* opens new opportunities to interfere with the transmission process. Furthermore, novel environmentally-friendly methods such as trap-plants to deploy push-pull strategies or biocontrol using endophytic entomopathogenic fungi are promising tools to manage meadow spittlebug populations and reduce the spread of *X. fastidiosa*-related diseases.

SE45 C05**INSIGHTS INTO THE MOLECULAR MECHANISMS OF RESISTANCE TO XYLELLA FASTIDIOSA IN OLIVES AND ADVANCES IN THE BREEDING PROGRAMS****P. Saldarelli, A. Giampetruzzi, M. Saponari, D. Boscia**

Institute for Sustainable Plant Protection, National Research Council, Unit of Bari (Italy)

In olive, resistance phenomena to the plant pathogenic bacterium *Xylella fastidiosa* (Xf) have been discovered in two cultivars, Leccino and FS17, whose infected trees show limited occurrence of desiccation phenomena on the canopies, together with low bacterial colonization of the trees. Similarly to what it is reported in other Xf-susceptible crop species, resistant phenotypes result from a complex network of host-pathogen interactions and factors. In this regard, despite the relevant research efforts, the genomic architecture and the genetic basis of the resistance to Xf remain largely unclear. Most recent studies include multi-omics approaches, i.e. physiological and anatomical observations, metabolomics, metagenomics and transcriptomics. Results converge on common features shared by the plant species/cultivars harboring resistant phenotypes. These include the size and the stem-average vessel diameters, pit membranes vulnerability to the Xf cell-wall degrading enzymes, ability to produce tyloses and a callose-like matrix to entrap the bacterium, resilience to the bacterium-imposed water stress, abundance of polyol compounds and common classes of differentially expressed genes (DEGs). In olive, initial gene expression studies indicate that Leccino response to Xf *pauca* involves Leucine Rich Repeat Receptor-like kinases and cell-wall receptors, both classes associated to Xf response in citrus or in grapevine, where orthologous genes are present in the Pierce's Disease resistant locus 1. Extending these molecular studies on progenies derived from the resistant



Concurrent Sessions

Friday, 5 July

cultivar Leccino which show resistant phenotypes, further confirmed that a plant defense response occurs in plant tissues infected with Xf. Of note is that the profiles of the DEGs differed among these Leccino progenies, confirming the complexity of the mechanisms underlying the phenomenon of resistance. On the other hand, several genes notoriously involved in the down regulation of the plant immune response have been also identified, i.e. "susceptibility genes", which are potential candidates for biotechnological application as demonstrated in grapevine and other crops.

SE45 C06

CAPACITY BUILDING AND COLLABORATION ON XYLELLA FASTIDIOSA THROUGH EUPHRESCO

B. Giovani

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The Euphresco network for phytosanitary research coordination and funding started in 2006 to support phytosanitary research programme owners and programme managers to develop and take advantage of synergies amongst national research programmes and activities in Europe. After receiving funding from the FP6 and FP7 EU Framework Programmes (EUPHRESCO I and EUPHRESCO II ERA-Net projects), Euphresco has become a self-sustained network (hereinafter referred to as Euphresco Net) hosted by the European and Mediterranean Plant Protection Organization (EPPO). *Xylella fastidiosa* has been a main concern for European and Mediterranean countries since it was first reported in Italy in 2013. Several projects were commissioned through Euphresco to support capacity building of European countries and to strengthen phytosanitary measures. The presentation will provide an overview of these projects and their main results and how Euphresco has been working, together with leading national and international organizations and initiatives on protecting plant health.

SE45 C07

CAPACITY BUILDING AND NETWORKING FOR XYLELLA FASTIDIOSA CONTAINMENT IN EUROPE AND IN THIRD COUNTRIES: THE IMPACT OF MSCA H2020 CURE-XF PROJECT

Maroun El Moujabber

CIHEAM Bari, Italy

The European Commission has published in October 2019 a list of 20 regulated quarantine priority pests. The list included *Xylella fastidiosa* whose economic, environmental and social impact on EU's territory is the most severe. Indeed, it has been shown that the bacterium *Xylella fastidiosa*, the pest with the highest impact on agricultural crops, could cause annual production losses of €5.5 billion, affecting 70% of the EU production value of older olive trees (over 30 years old) and 35% value of younger ones. In addition to direct impacts on production, *Xylella* has significant indirect effects on a wide range of upstream or downstream economic sectors. CURE-XF dealt with different aspects of the current re-emergence of diseases caused by *Xylella fastidiosa*: from the detection and characterization of the bacterial strain to the control and management of the diseases. The consortium relied on multidisciplinary expertise on genetics and bioinformatics,

pathogen-hosts interactions, diagnosis and monitoring, vector transmission and epidemiology, with the aim to raise awareness and strengthen actions for prevention. Main results and impacts:

- Improving scientific capacities and networking among the European Countries as well as between the EU and Third Countries, in particular involving high-risky neighbouring countries;
- Strengthening the knowledge and the know-how on Xf in Third Countries having intense exchange of plant material with Europe;
- Raising awareness in relation to Xf impacts and risks upon its establishment.
- Maximize the exploitation of results of the research program by facilitating networking and technology transfer to stakeholders and end users.

The main results of the project were the training of more than 1000 persons (120 in highly qualifying trainings) and the discovery of *Xylella fastidiosa* in Lebanon. In addition, the project helped Lebanon in establishing its national contingency plan and the National Action Plan for the monitoring and response to *Xylella fastidiosa*.

SE45 C08

ANCoSIX ITALIAN PROJECT: ADVANCING SUSTAINABLE AND INNOVATIVE CONTROL OF XYLELLA FASTIDIOSA

Franco Valentini

CIHEAM Centre International de Hautes Etudes Agronomiques Mediterraneennes, Via Ceglie 9, Valenzano, Bari, Italy

Given the threat posed by *Xylella fastidiosa* (Xf) on the Mediterranean cropping system, due to the numerous cultivated and wild hosts, the difficulties of eradicating the infections and controlling the vectors, pioneering plant protection tools are highly needed. Strategies aimed to control *Xylella-driven* diseases should rely on innovative approaches which could ensure effectiveness and environmental sustainability. ANCoSIX is a national research project funded by the Italian Ministry of Agriculture, which involves public research institutes and private companies to study several techniques for supplying active compounds in diseased and healthy olive trees, in order to study the curative and the preventive effects of phenolic extracts, peptides and phages on both susceptible and tolerant olive tree varieties. Nanotechnology represents a valid approach to overcome the most common issues of natural compounds, which are difficult to deliver in field conditions. In ANCoSIX the advantages of nanocarriers, such as cellulose nanocrystals and chitosan nanoparticles, are exploited for delivering highly performing nano-agrochemicals to plants by fertigation or trunk-injection systems. The results already obtained highlight the valid biological activity of the proposed compounds both in vitro and in plant model systems for the Xf multiplex and pauca subspecies, highlighting their potential in field applications, which are currently still being studied in experimental farms and greenhouses.



SE45 C09

DEFENSE PRIMING AND GRAPEVINE IMMUNE RESPONSES TO THE XYLEM-LIMITED BACTERIUM, XYLELLA FASTIDIOSA

M. C. Roper

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Priming improves plant defense by enhancing activation of induced defense responses to future pathogen challenge. Microbial signature molecules called pathogen or microbe-associated molecular patterns (PAMPs/MAMPs) can act as stimuli to induce the primed state. The Gram-negative bacterium, *Xylella fastidiosa*, is the causal agent of Pierce's disease (PD) of grapevine and is limited to the xylem, a tissue compartment that is primarily non-living at maturity, but mounts a robust defense response to xylem invading pathogens. *Vitis vinifera* grapevines pre-treated with the purified *X. fastidiosa* MAMP, lipopolysaccharide (LPS), as a priming stimulus had a significant decrease in both external and internal symptoms of PD as well as the rate of overall disease progression indicating that LPS primes the grapevine immune response to better defend itself against challenge with *X. fastidiosa*. Differential gene expression analysis revealed major transcriptomic reprogramming in primed vines in response to pathogen challenge at the point of inoculation and 20 nodes (1.5m) distal to the point of inoculation when compared to naive, untreated vines. Furthermore, a weighted gene co-expression analysis identified modules of co-expressed genes common to the point inoculation and 20 nodes above indicating that primed vines mount a temporally and spatially synchronous response to initial pathogen challenge. These responses included genes involved in signal perception, signal transduction, as well as auxin-related pathways.

SE45 001

POTENTIAL VECTORS OF XYLELLA FASTIDIOSA IN AUSTRALIA

Trebicki P.¹, Selleck C.², Hoskins J.³, Blacket M.²

1. Macquarie University, Sydney, Australia, 2. Agriculture Victoria, Bundoora, Australia, 3. NSW Department of Primary Industries, Yanco, Australia

Xylella fastidiosa although not present in Australia is a destructive plant pathogenic bacterium, with several subspecies affecting many plant species including grapevines, almonds, peaches, apricots, and olives in many countries worldwide. It is xylem limited, and transmitted mainly by spittlebugs, sharpshooters, leafhoppers and froghoppers. *Xylella* and its exotic vectors have been identified by the Plant Health Committee as the number one National Priority Plant Pest for Australia, by New Zealand MPI as an 'unwanted organism' and by the European Commission as one of most dangerous plant bacteria worldwide. Our research aims to provide biosecurity agencies with tools and knowledge which can be effectively implemented to rapidly eradicate *Xylella fastidiosa* or prevent and suppress its spread if there is an incursion in Australia. This project focuses on potential insect vectors of *Xylella*, biology, physiology, and ecology in targeted horticultural crops across three states of Australia. Efforts, methodology, results, and its significance to Australian food production will be communicated and discussed.



MC3

15.00-17.30 Concurrent Session **46**

Post-Harvest Pest and Disease Management

Chair: Christos Athanassiou (University of Thessaly, Greece)

SE46 C01

GRAIN PROTECTANTS: CHALLENGES AND OPPORTUNITIES

Christos G. Athanassiou^{1,2}, Manoj Nayak³, Frank H. Arthur⁴, Vaclav Stejskal⁵, Paraskevi Agrafioti^{1,2}, Maria Sakka¹, Christos I. Rumbos¹

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The term grain protectants refers to contact insecticides that are applied directly on grains, such as wheat, rice and maize, as they are loaded into a storage structure. Their use has certain incontestable advantages over the use of fumigants, which are: a) they have a long residual effect and can protect the grains for a long period of time, b) they can be partially applied on the grains and incorporated with the whole grain mass through admixture and c) they are applied once, during the initial loading and storage of grains. Moreover, despite the occurrence of resistance, grain protectants are effective for a wide range of insect and mite species that cannot be controlled easily with other methods. Although these insecticides have been extensively used to control insects in stored grains, their use has declined with the increasing use of fumigants, particularly phosphine. One of the main reasons for the decrease in their use is many active ingredients that were registered for direct application on grains have been withdrawn from the market, leaving a considerable gap in long-term stored grain protection strategies. Nevertheless, newer active ingredients with reduced mammalian toxicity have been registered as grain protectants in many parts of the world, and their use can be incorporated in Integrated Pest Management protocols. As such, grain protectants will continue to play a pivotal role in stored product protection, especially in developing countries where access to fumigants may not be always possible.

This research was carried out as part of the project «PrecisionFEEDProtect: Precision protection of stored feed from entomological infestations using innovative technologies» (Project code: KMP6-0077613) under the framework of the Action «Investment Plans of Innovation» of the Operational Program «Central Macedonia 2021-2027», that is co-funded by the European Regional Development Fund and Greece.

SE46 C02

USING BEHAVIOR AND MICROBIAL ECOLOGY TO HELP IMPROVE SURVEILLANCE AND MANAGEMENT PROGRAMS FOR STORED PRODUCT INSECTS IN A WARMING WORLD

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Understanding behavior is a central component to improve surveillance and integrated pest management programs (IPM). As climate change progresses, behavior of insects may be affected by temperature and other abiotic factors. One change may be in trapping efficiency for pests, while another may be in altered insect-microbe interactions after stored product insects disperse to a new environment (Figure 1). We tested these hypotheses over multiple years with different temperature profiles across a large monitoring dataset of transects in North America from collaborators ranging from South Dakota to Mexico. At each site, three habitats were surveyed (native, food facility, and field crop) with traps, and we linked captures to mean preceding weekly temperatures from local weather stations. In addition, we examined how the microbial growth on field-collected rice weevil, *Sitophilus oryzae* (Coleoptera: Curculionidae) varied over the season and with temperature preceding capture. We found the relationship between temperature and captures of stored product insects depended on species and included positive, negative, and parabolic relationships. Further, microbial growth and diversity initiated by *S. oryzae* in new environments and temperature in field from the preceding week to capture was positively correlated, suggesting a warming world will also exacerbate hotspots of microbial infestation, that may endanger human health. In certain areas, microbial growth corresponded to peak harvests for wheat and maize. Finally, we discuss implications of our research for quarantine species such as the khapra beetle, *Trogoderma granarium* (Coleoptera: Dermestidae) and the larger grain borer, *Prostephanus truncatus* (Coleoptera: Bostrychidae), which are threats to global food security. Our results suggest a complex interplay of temperature, stored product insects, and microbes that appears to be worsening with climate change.

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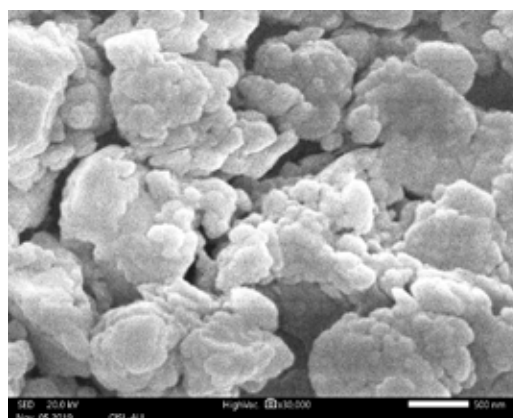
POSTHARVEST DISEASE MANAGEMENT OF FRESH PRODUCE WITH INNOVATIVE AND ECO-FRIENDLY SOLUTIONS

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Managing postharvest diseases in fruits and vegetables is crucial for global food security amid a growing population. Traditional methods like chemical fungicides are supplemented and substituted by innovative strategies utilizing natural compounds, such as essential oils and antifungal edible coatings, and biocontrol agents to curb losses. Biocontrol agents, with mechanisms like competition, biofilm formation, mycoparasitism, and resistance induction are extensively researched for various pathogens and crops. Endophytic microorganisms, found within plants, are emerging as a sustainable option for postharvest pathogen management. Fruit surfaces harbour resilient microbial communities, making it challenging for biocontrol agent the establishment. Integrating microbial communities to create a conducive environment for biocontrol agents shows promise in real-world conditions. Future strategies may involve timing biocontrol agent application already at flowering. Essential oils, valued for their safety, eco-friendliness, and antimicrobial properties, are gaining attention. Their phytochemicals disrupt pathogen metabolism, but challenges like low stability and water solubility require careful formulation. Essential oils can be applied as biofumigants in storage chambers or through coating and encapsulation techniques, enhancing antimicrobial and antioxidant activities. Combining biocontrol agents with nutrient additives, coatings, or essential oils is being explored for improved postharvest pathogen control, aiming for synergistic effects in disease management, in an integrated vision of disease management.

(L.). As EGEO is volatile, its efficacy can be enhanced by developing control-release formulations. There are limited number of studies on novel formulations and their efficiency in practical situations. In the present study the EGEO was formulated as nano-emulsion and nanocapsules by using natural polymers and their combinations. The developed formulations were bio-assayed under laboratory conditions against *S. oryzae*. The morphology, chemical structure and presence of active ingredients in the effective formulation were periodically characterized by FTIR, SEM, EDS and GC-MS. ai Entrapment Efficiency and release kinetics, storage stability and impact on germination were also studied. Chitosan-Sodium alginate nanocapsules (CSNC) recording maximum mortality in the lab was bio-assayed under field conditions. CSNC were effective even after 147 days while pure essential oil treatment was effective till 17 days. Chitosan and sodium alginate possess attractive applications because of their control release, non-toxic, and biodegradable nature. They offer ample scope in the development of novel stored product pest control formulations.



Chitosan-Sodium alginate nanocapsules (CSNC)

SE46 001

EUCALYPTUS GLOBULUS ESSENTIAL OIL-BASED CONTROL-RELEASE NANOFORMULATIONS AGAINST SITOPHILUS ORYZAE (L.): PROMISING NOVEL NANO-DELIVERY SYSTEMS FOR SUSTAINABLE GRAIN PROTECTION

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Among the post-harvest losses insects play a significant role. India's annual food grain storage loss due to insects is estimated to be INR 13 billion (IGMRI, 2022; TNAU, 2022). Stored pest management relies on using synthetic fumigants, causing toxic and environmental issues. Phyto-insecticides although promoted as alternatives need to be developed as commercial formulations with long residual efficacy. Control release formulations using natural polymers like chitosan and sodium alginate, improve stability and effectiveness. The *Eucalyptus globulus* (Labill) essential oil (EGEO) possessed anti-insect activities against Rice weevil, *Sitophilus oryzae*

SE46 002

ATTRACTIVENESS OF MALE AND FEMALE ADULTS OF CRYPTOLESTES FERRUGINEUS (COLEOPTERA: LAEMOPHLOEIDAE) IN GRAIN

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Whether stored-grain insects can communicate with each other inside stored-grain bulks is an important question for the development of pest management programs. Movements of the individual adults of *Cryptolestes ferrugineus* towards caged adult(s) were studied inside a tube (2.5 cm diameter and 10 cm length), using an infrared camera. The numbers of the caged adults were 1, 20, or 50 of females or males, and 100 or 200 mixed sex adults. Before insects were introduced, the cage was filled with 3 g of feed (whole wheat, wheat germ, and cracked wheat with a 16:1:1 ratio by weight), and the tube was filled with 14% moisture content of wheat. Unlike the trials conducted without grain in the tube for the single caged insects, when grain was present in the tube there appeared to be no major attraction. For the cases of 20 and 50 caged males and females with a single adult of the opposite sex introduced in the tube, there appeared to be more attraction of the single female to caged males. When single male and female adults were introduced to trials with 100 mixed sexed

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caged insects, they presented attraction, but with the females having a stronger response of movement towards the insect cages. When the number of mixed-sex caged adults was increased to 200, the single male and female introduced in the tube both moved towards the insect cages.

SE46 003

STUDIES ON BIOLOGY AT DIFFERENT TEMPERATURE REGIMES AND SCREENING OF INDIGENOUS STORED RICE CULTIVARS AGAINST RICE WEEVIL, SITOPHILUS ORYZAE (L.)

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Rice is one of the most widely grown cereals with high global recognition and food value. In stored conditions, the most notorious pest of rice is Rice weevil (*Sitophilus oryzae*). Therefore, the present experiments were conducted to study on biology of Rice weevil, *Sitophilus oryzae* (L.) at different temperature regimes i.e., $20 \pm 1^\circ\text{C}$, $25 \pm 1^\circ\text{C}$ and $30 \pm 1^\circ\text{C}$. and Varietal screening of different indigenous cultivars of stored rice grains against Rice weevil (*S. oryzae*). The experiments were conducted in Entomology laboratory of School of Crop Protection, CPGS-AS, CAU(I), Umiam, Meghalaya. The result of present study showed that the morphometrics of the eggs, larva and pupa were found to be smallest at $20 \pm 1^\circ\text{C}$ and largest at $30 \pm 1^\circ\text{C}$ with a range such as Egg: length (0.24 ± 0.03 to 0.47 ± 0.03) mm and breadth (0.09 ± 0.02 to 0.19 ± 0.03) mm, Larva: length (0.22 ± 0.03 to 6.37 ± 0.34) mm and breadth (0.08 ± 0.01 to 2.40 ± 0.03) mm, Pupa: length (2.56 ± 0.60 to 4.45 ± 0.43) mm and breadth (1.39 to 2.45 ± 0.51) mm at $20 \pm 1^\circ\text{C}$. The hatching percentage of eggs of *S. oryzae* was maximum (81.7 ± 12.6) per cent at $30 \pm 1^\circ\text{C}$ and minimum (25.0 ± 10.0) at $20 \pm 1^\circ\text{C}$. It is concluded that the variety Jowain white showed higher percent weight loss at 30 days (9.5 ± 0.2), 60 days (15.2 ± 0.1) and 90 days (20.4 ± 0.2). The variety Lakang baleigh showed lowest weight loss percent at 30 days (2.5 ± 0.3), 60 days (4.2 ± 0.1) and 90 days (8.3 ± 0.3). The variety Jowain white had highest susceptible index (13.9 ± 4.5) followed by Pnah sticky (13.6 ± 1.5), Jowain red (13.1 ± 1.9) and the variety Lakang baleigh showed least susceptible index (6.6 ± 1.9). Growth index was found to be highest in the cultivar Mynri umiam (1.90 ± 0.08). Considering the findings of the present study may be concluded that Prolonged storage of rice in lower temperature is beneficial and among the local varieties evaluated Lakang baleigh was found to be relatively resistant to the pest *S. oryzae*.

SE46 004

THE USE OF SIGS (SPRAY-INDUCED GENE SILENCING) STRATEGY IN THE CONTROL OF BOTRYTIS CINEREA IN HORTICULTURAL CROPS

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The causal agent of the gray mold disease, *Botrytis cinerea*, is one of the main limiting factors of horticultural crops production worldwide. Its control is very dependent on the

use of fungicides; however, due to the ability of this pathogen to develop resistance and the new European legislation regarding the reduction of fungicide diversity available for grower in the next years, new advances and technologies are needed to control this important harvest and postharvest disease. In this study, the potential of the RNA interference (RNAi) technology known as "spray-induced gene silencing" (SIGS) as an alternative for the management of *B. cinerea* was explored. For this purpose, a double-stranded RNA (dsRNA) molecule was designed to target the transcription factor *mrr1*, which is involved in the overexpression of the ABC transporters (drug efflux transporters) and the resistance to several commercial fungicides. To test the efficacy of *mrr1*-dsRNA, dsRNA uptake by the fungus, sensitivity assays (effect on conidia germination, detached leaf, and fruit assays), fungal biomass analysis and gene expression studies were carried out. The results showed that the fungus was able to uptake it, reducing the conidia germination of the pathogen. Subsequently, it was shown, in in vivo assays performed on tomato leaves and apple fruit, that the application of *mrr1*-dsRNA downregulated *mrr1* gene expression and its silencing produced a significant reduction of *B. cinerea* development, resulting in a reduction of the fungal biomass. These results demonstrated the potential of the SIGS strategy for the control of *B. cinerea* and *mrr1*-dsRNA as a new molecule with fungicide activity that could be included into the several strategies carried out for sustainable plant protection control programs in the field.

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SE46 005

SEED STORAGE FUNGI AND AFLATOXIN CONTAMINATION IN SESAME IN BENUE STATE, NORTH CENTRAL NIGERIA

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Storage of Sesame (*Sesamum indicum*, L.) is gaining more ground among the farmers and users of the crop in Benue state, North central Nigeria in order to maximize profits or for evacuation for exports and seed for planting. These expected benefits usually fail due to fungi infestation and their attendant aflatoxin contamination. Using direct plating technique; the study identified twelve species of fungi genera and various concentrations of aflatoxin contamination. The percentage occurrence of fungi isolates shows *Aspergillus flavus* and *A. niger* being dominant and significantly higher than ($P \leq 0.05$). High power liquid chromatography (HPLC) analysis revealed four (4) types of aflatoxin namely: AFB1, AFB2 AFG1 AND AFG2 with $3.95 - 11.75 \mu\text{g/kg}$, $0.00 - 2.35 \mu\text{g/kg}$, $0.00 - 2.06 \mu\text{g/kg}$; $0.00 - 1.47 \mu\text{g/kg}$ respectively showing AFB1 with the highest frequency occurrence of $3.95 - 11.75 \mu\text{g/kg}$. All the values were above the Maximum Residual limits (MRL) of

2 µg/kg for aflatoxin B1 and 4 µg/kg for total aflatoxin. In conclusion, it is recommended that several treatments should be applied to reduce the levels of contamination in sesame before its utilization.

SE46 006

INTEGRATED PRE- AND POSTHARVEST PRACTICES FOR MANAGING BROWN ROT OF CITRUS FRUIT TO MEET QUARANTINES FOR PHYTOPHTHORA SPECIES

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Brown rot of citrus fruit can be a devastating disease worldwide resulting in significant crop loss especially in harvest seasons with high rainfall. Several species of *Phytophthora* have been reported to cause brown rot in California, and *P. syringae* and *P. hibernalis* are quarantine pathogens in some export markets for California citrus fruit. These quarantines aim to prevent pathogen dissemination, but they have caused market closures if the pathogens are detected at arrival ports resulting in significant economic hardship and barriers to trade. With use restrictions on copper fungicides, new preharvest treatments were developed using the new foliar-applied fungicides oxathiapiprolin and mandipropamid that have different modes of action. These fungicides were registered and provide persistence for 2 to 3 months with high performance in reducing brown rot decay and direct crop loss in the field. To meet export demands, however, postharvest washing removes residues of these fungicides to non-detectable levels. With the occurrence of symptomless (latent) brown rot infections, postharvest treatments are needed to obtain the highest level of control for export markets, and these also need to target other common fruit pathogens. Potassium phosphite (KPO₃) is registered in the United States for pre- and postharvest use on citrus and is highly effective against brown rot in heated applications but is only suppressive against *Penicillium* decays. Trials were done on the positioning of the acidic KPO₃ in the packinghouse fruit processing sequence so it would not interfere with the efficacy of alkaline treatments that are used for green mold and sour rot control. We determined that the KPO₃ treatment after the sodium carbonate dip and before the fungicide-fruit coating treatment provides the highest efficacy for brown rot while the activity of treatments against other postharvest decays of citrus is maintained. This ensures that fruit arrivals are of highest quality.

SE46 007

EFFICACY OF EXTREME TEMPERATURES ON ALL LIFE STAGES OF *ORYZAEPHILUS SURINAMENSIS* (L.) AND *EPHESTIA KUEHNIELLA* ZELLER

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The escalating resistance observed in diverse contact and fumigant insecticides underscores the necessity for seeking alternative pest control management approaches. Utilizing extreme temperatures presents a hopeful alternative to chemicals for effectively disinfecting a range of stored

commodities like dried fruits and grains. In this study, a series of laboratory bioassays were carried out to assess the insecticidal efficacy of heat and cold treatment on two major pests of dried fruits. All life stages of the saw-toothed grain beetle, *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae), and the Mediterranean flour moth, *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) were exposed to cold or heat treatment at temperatures ranging from -18°C to 5°C and 30°C to 55°C, respectively, for various durations ranging from 1 to 168 hours. Furthermore, a second series of bioassays was conducted to address the disinfection of artificially infested dried fruits such as currant, figs, and dates. Our study findings reveal varying susceptibility across different life stages, emphasizing the significance of exposure duration as a critical factor. Generally, temperatures below -10°C and above 45°C have shown effectiveness in controlling all tested species across all life stages. In conclusion, our results suggest that extreme temperatures show promise in controlling *O. surinamensis* and *E. kuehniella*, as well as for disinfecting dried fruits.

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SE46 008

DIATOMACEOUS EARTHS ARE PROMISING ALTERNATIVES FOR THE CONTROL OF STORED PRODUCT INSECTS

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Diatomaceous earths (DEs) are promising alternatives over the use of traditional insecticides. In a laboratory screening, ten diatomaceous earth formulations (namely as AGR, DD, ELC, SLEC, D3, D5, D6, TRH, D7, D8) were evaluated, for the control of a wide range of stored product insect species at different dose rates. The most effective diatomaceous earth formulation was the DD-Silicid formulation, with high mortality levels for most of the stored-product insect species tested. This formulation was further utilized in semi-field conditions. The objective of this application was the evaluation of the insecticidal activity of Silicid against the rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae) and the confused flour beetle, *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) under "real-world" conditions in a feed mill facility in Greece. Different dose rates (i.e. control, 500, 1000 and 2000 ppm) of the DE formulation were applied in a series of lots of 20 kg of hard wheat, stored and kept from November to February in the facility. Three hundred (300) adults of each species were introduced into each lot and one probe trap was inserted in the middle of each grain quantity. Based on weekly monitoring via the probe traps and



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grain samples, the results showed that the DE application can effectively control the tested populations in semi-field conditions, even in the lowest DE dose. The present study provides data that encourage the utilization of diatomaceous earths for the control of stored product beetles in bulked grains, and the incorporation of this technique in integrated pest management strategies.

This research is part of the project «Integrated management of insect infestations in stored animal feed: Feed without pesticides» (Project code: KMP6-0088130) that is co-funded by Greece and European Union by the Action «Investment Plans of Innovation» in Central Macedonia under the framework of the Operational Program «Central Macedonia 2021-2027».



CONFERENCE I

15.00-17.30 Concurrent Session 47

Molecular Plant Microbe Interactions

Chair: Nikolaos Mastrodimos (School of Agriculture and Food Science, University College Dublin, Ireland)
Anastasia Venieraki (Agricultural University of Athens, Greece)

SE47 001

CHARACTERISING A SMALL, SECRETED, PROTEIN FROM THE WHEAT PATHOGEN ZYMOTSEPTORIA TRITICI BY ECTOPIC EXPRESSION IN ARABIDOPSIS

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Zymoseptoria tritici causes the devastating disease Septoria Tritici Blotch (STB) in wheat. This fungal pathogen is of high economic importance and can reduce yields by up to 50%. During infection, *Z. tritici* employs effector proteins to suppress host immunity and facilitate infection. In this study, we identified a novel small, secreted protein from *Z. tritici*, *ZtHRLP1*, which has structural homology with hormone receptors from *Arabidopsis thaliana* and wheat. The expression of *ZtHRLP1* peaks at the transition from the biotrophic to the necrotrophic stage of *Z. tritici* growth. To investigate a potential role in hormone signaling, *ZtHRLP1* was ectopically expressed in *Arabidopsis*. The impact on hormone signaling in these *Arabidopsis* lines is currently under investigation via marker gene expression analysis and susceptibility to pathogens such as *Botrytis cinerea*. In addition, the impact of hormone manipulation on STB disease was investigated. Exogenous hormone application promotes STB symptoms, inducing the earlier appearance of chlorosis, yellowing, and necrosis, while significantly increasing pycnidia and pycnidiospore production. These results suggest that *Z. tritici* may target plant hormone signalling to promote disease in wheat and employ the candidate effector *ZtHRLP1* to hijack this. Future analysis of the *ZtHRLP1* *Arabidopsis* ectopic expression lines will support such a role.

SE47 002

DIVERSITY OF BACILLUS AMYLOLIQUEFACIENS GROUP SPECIES SECONDARY METABOLITES BIOSYNTHETIC GENE CLUSTERS INVOLVED IN PLANT PROTECTION - AN EVOLUTIONARY PERSPECTIVE

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Different *Bacillus* endophytic bacterial strains isolated from Greek indigenous medicinal plants or olive trees, after numerous experiments emerged remarkable beneficial characteristics and biological activity against soilborne phytopathogens. We investigated their plant growth-

promoting effect and their ability to induce expression of defense-related genes, resulting in the emergence of promising bioproducts for sustainable horticulture as they combine direct antifungal activity against plant pathogens and the ability to prime plant immunity and enhance plant growth. We continued our research to identify their mode of action using Genome mining through ClusterBlast. Two groups of *Bacillus* biocontrol agents are drawing the attention of researchers: the *B. subtilis* and the *B. amyloliquefaciens* groups. Both of these show high phenotypic similarities and are commonly used for the control of fungal pathogens. The agricultural uses of the *B. amyloliquefaciens* group, which consists of *B. amyloliquefaciens*, *B. velezensis*, *B. siamensis* and *B. nakamurai* species, have recently emerged as an interesting source of biocontrol agents for the management of pathogenic fungi and bacteria. *B. amyloliquefaciens* species have been reported to synthesize and secrete secondary metabolites with strong antifungal and antibacterial activities with some of them being able to induce the plant innate defence systems. *B. velezensis* showed superior potential for the production of these secondary metabolites compared to *B. amyloliquefaciens*, *B. siamensis* and *B. nakamurai* species. Indeed, recent comparative genomic analysis indicated that *B. velezensis* harbour a much higher diversity of secondary metabolites biosynthetic gene clusters involved in plant protection. In this study, we present comparative data on the in silico characterization of secondary metabolites biosynthetic gene clusters involved in plant protection from endophytic *B. velezensis* and *B. amyloliquefaciens* strains isolated from medicinal plants and olive trees. Furthermore, we present evolutionary patterns of secondary metabolites biosynthetic gene clusters in *B. amyloliquefaciens* group species.

SE47 003

ALL PATHWAYS LEAD TO DEGRADATION: THE UBIQUITINATED RELATIONSHIP OF HOST-PATHOGEN INTERACTION AND INDUCED RESISTANCE

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Biotrophic phytopathogenic fungi have a dynamic interaction both with the host and the changing environment causing serious diseases. Plant defense inducers (PDIs) is a group of various substances that could be used to manage



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such pathogens. Here, we discuss the early stages of *Podosphaera xanthii* infection on *Arabidopsis thaliana* plants with and without the application of a nano-chitosan PDI, using transcriptomics, proteomics, and protein structure prediction with the use of an artificial intelligence software. *Arabidopsis* seedlings grown in growth media supplemented with Salicylic acid loaded chitosan nanoparticles (SA-CNPs), showed enhanced ROS production and *PR1* expression along with significant reduction of *P. xanthii* conidial germination. Inoculation with the pathogen did not cause any significant differentiation in *Arabidopsis* proteome, while SA-CNPs application highly upregulated both transcript and protein expressions. Functional analysis of the upregulated genes revealed enrichment in the integration of ROS production, Salicylic acid and glutathione activity, key defense pathways. Interestingly, SA-CNPs application upregulated the expression of various proteasome-, ubiquitin- and ribosome-related proteins, indicating a critical integration of the degradation machinery with induced defense. On the other hand, *P. xanthii* transcriptomics identified 78 secreted proteins (SPs) that were greatly downregulated after SA-CNPs application. Differentially expressed SPs of interest were subjected to tertiary structure prediction by the AlphaFold2 server. Through structural comparisons using the DALI server, we identified within the secreted proteins the distinct functional domains of an E3 ubiquitin-protein ligase RNF8 (E3/RNF8), a N-terminus ubiquitin-like structure with C-terminal S27a ribosomal subunit domain (UBS27), and a 5'-AMP-activated protein kinase catalytic subunit alpha-1. Protein-protein interaction (PPI) prediction showed that *Arabidopsis* OTU1, an endoplasmic reticulum-associated linkage-specific deubiquitinase with high expression after SA-SNPs application, is highly associated to effector UBS27 with interface template modeling score (ipTM) = 0.671. The results suggest a dynamic interplay between pathogen invasion and hosts's defence in balancing protein homeostasis.

SE47 004

PHYTOPHTHORA SOJAE BOOSTS HOST TREHALOSE ACCUMULATION TO ACQUIRE CARBON AND INITIATE INFECTION

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Successful infection by pathogenic microbes requires effective acquisition of nutrients from their hosts. Root and stem rot (RSR) caused by *Phytophthora sojae* is one of the most important diseases of soybean (*Glycine max*). However, the specific form and regulatory mechanisms of carbon acquired by *P. sojae* during infection remain unknown. Here, we show that *P. sojae* boosts trehalose biosynthesis in soybean through the virulence activity of an effector PsAvh413. PsAvh413 interacts with soybean trehalose-6-phosphate synthase 6 (GmTPS6) and increases its enzymatic activity to promote trehalose accumulation. *P. sojae* directly acquires trehalose from the host and exploits it as a carbon source to support primary infection and development in plant tissue. Importantly, *GmTPS6* overexpression promoted *P. sojae* infection, whereas its knockdown inhibited the disease, suggesting that trehalose biosynthesis is a susceptibility factor that can be engineered to manage RSR in soybean.

SE47 005

VELVET FAMILY PROTEIN FPVELB AFFECTS VIRULENCE IN ASSOCIATION WITH SECONDARY METABOLISM IN FUSARIUM PSEUDOGRAINEARUM

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Fusarium crown rot, primarily caused by the soil-borne fungal pathogen *Fusarium pseudograminearum*, is a widespread and destructive disease that affects cereal crops, particularly wheat and barley. *F. pseudograminearum* successfully infects and colonises host tissues by producing certain secondary metabolites (SMs). The velvet protein family is a crucial regulator in development, virulence, and secondary metabolism of fungi. In this study, we investigated the function of a velvet protein family member, VelB orthologous gene, FpVelB, in the development, virulence, and expression of SM genes in *F. pseudograminearum*. We conducted a functional analysis of FpVelB, a member of the velvet family proteins in *Fusarium pseudograminearum* and which causes destructive crown disease in wheat, using a gene replacement strategy to enhance our understanding of its physiological functions. Deletion of FpVelB resulted in decreased radial growth compared to that of *F. pseudograminearum* and enhanced conidial production. Furthermore, FpVelB modulates the fungal responses to abiotic stress through diverse mechanisms. Significantly, virulence decreased after deletion of FpVelB in both the wheat stem base and head. Genome-wide gene expression profiling revealed that the regulation of genes by FpVelB is associated with several processes related to the aforementioned phenotype, including "immune", "membrane", and "antioxidant activity", particularly with regard to secondary metabolites. Most importantly, we demonstrated that FpVelB regulates pathogen virulence by influencing deoxynivalenol production and modulating the expression of the *PKS11* gene. FpVelB is crucial for plant growth, asexual development, and abiotic stress response and is essential for full virulence via secondary metabolism in *F. pseudograminearum*.

SE47 006

XOPG2 EFFECTOR OF XANTHOMONAS CAMPESTRIS PV. CAMPESTRIS IS A MAJOR RACE 5 DETERMINANT

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Black rot caused by *Xanthomonas campestris* pv. *campestris* (Xcc) poses a significant risk to brassicas, such as cabbage, cauliflower, and Brussels sprouts. Xcc has 11 known races that are differentiated on a set of brassica cultivars. Yet, our understanding of the role of race-specific effectors in Xcc pathogenicity is still limited. Using comparative genomics, we identified the *XopG2* effector (a metalloprotease) as the Xcc Race 5 determinant. Expression of *XopG2* in the highly virulent Xcc Race 6 compromised its pathogenicity in the Xcc R5-resistant Brassica oleracea line SxD1, while in planta expression of *XopG2* resulted in cell death formation. Furthermore, mutations on the metalloprotease active site of



XopG2 and XopG2 deletion mutants avoid recognition by the resistant SxD1 line. Consistent with this result, we identified naturally occurring allelic variants of XopG2 with a truncated metalloprotease active site that have evolved to evade recognition. Using proteomics and split luciferase assays, we identified EDR2 (enhanced disease resistance 2) as the interacting partner of XopG2. Currently, we are working to identify resistant proteins guarding EDR2.

SE47 007

BENEFICIAL PSEUDOMONAS HIJACKS PLANT-WIDE TRANSCRIPTION TO INDUCE COUMARIN-DEPENDENT SYSTEMIC RESISTANCE

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Induced systemic resistance (ISR) is a defense mechanism expressed in leaves upon root colonization by beneficial microbes. Root-specific transcription factor MYB72 is essential for ISR in the *Arabidopsis-Pseudomonas simiae* WCS417 (WCS417) model system. It is also important, together with downstream components F6'H1 and BGLU42, in the *Arabidopsis* response to iron (Fe) deficiency and the production of Fe-mobilizing coumarins. While coumarin-related genes are essential for ISR establishment, the detailed mechanism describing how plants integrate microbially-induced coumarin accumulation into plant resistance remains unknown. To elucidate the role of coumarins in ISR activation, we investigated the transcriptomic response of coumarin-related genes and determined coumarin profiles within ISR-expressing *Arabidopsis* in a temporospatial manner. For this, we performed a time-series RNA-seq experiment (1 to 7 days) in roots and shoots following WCS417 colonization. Root inoculation with WCS417 activated a large set of Fe deficiency-related genes and coumarin biosynthesis-associated genes, first in roots within 2 days after treatment, and later in shoots within 5 days after treatment. Multispectral imaging of coumarins in the roots showed that WCS417 triggered a different coumarin distribution pattern from that in Fe-starved plants. We further monitored WCS417-mediated coumarin accumulation across the whole plant via liquid chromatography-mass spectrometry. We found altered coumarin levels with a transient peak both locally in the root and systemically in the shoot, indicating that systemic coumarin accumulation could play (in)direct role in ISR. To finally examine whether systemic coumarin accumulation is linked to ISR, we performed ISR bioassays with coumarin biosynthesis mutant *f6'h1* and an *F6'H1* overexpressing transgenic line. The *f6'h1* mutant failed to mount ISR against *Pst* DC3000, while the *F6'H1* overexpressing line showed constitutive resistance, indicating that functional coumarin biosynthesis is essential for ISR triggered by WCS417. Together, our findings display how beneficial bacteria WCS417 hijacks plant-wide transcription to induce coumarin-dependent systemic resistance.

SE47 008

INVESTIGATING THE EARLY RESPONSE OF PEPPER AGAINST POTATO VIRUS Y

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Potato virus Y (PVY) is enlisted among the most important viruses that mainly affect the cultivation of Solanaceous plants. The management of viral diseases is based on prophylaxis mainly through the use of genetically resistant cultivars and the application of insecticides for the elimination of virus insect vectors. The latter has great environmental and economic impact. Therefore, the identification of novel sources of genetic resistance is of high importance. In this context, this study aims to unravel the early response of pepper plants (*Capsicum annuum* cv. Yolo Wonder) against PVY for the identification of novel candidate resistance genes. The transcriptome related to the early response of plants against three distinct PVY isolates was assessed with a high throughput sequencing method (RNA-Seq). The obtained data were analyzed with standard *in silico* pipelines for the identification of differentially expressed genes (DEGs), gene ontology (GO) terms and pathways in the PVY inoculated plants compared to mock inoculated ones. The bioinformatics analysis identified 1,788 DEGs, with the majority being up-regulated in the PVY inoculated plants compared to the mock ones. The GO terms "*immune system process*", "*detoxification*" and "*signaling*" were over represented upon PVY inoculation. Furthermore, PVY inoculation resulted in the alteration of the cell wall modification pathway, with an increased expression of genes involved in the biosynthesis of celluloses and hemicelluloses. The effect of the cell wall modification process in the PVY infection was further investigated through the quantification of cellulose content with Calcofluor White staining protocol in a SP8 X confocal microscope. This study constitutes the first analysis of the early response of pepper against PVY that depicts the involvement of known and novel pathways and genes, while the data obtained can be used for the identification of new candidate resistance genes.

SE47 009

MINING THE EFFECT OF BIOSTIMULANTS AND PLANT PROTECTION PRODUCTS (PPPS) ON CANNABIS (CANNABIS SATIVA) METABOLISM APPLYING METABOLOMICS

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Cannabis cultivation is considered of high potential due to the production of a vast array of bioactive metabolites

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(e.g. cannabinoids, terpenoids), which have high economical and scientific value. Nonetheless, until recently, there was no regulatory framework for its cultivation and research. This resulted in the lack of knowledge on its cultivation and plant protection, particularly in cases where the crop is intended for medicinal purposes. The above-mentioned hindered the progress of further development of Cannabis cultivation. Recently, there has been increasing interest in the biostimulant-related research, driven by their numerous advantages over traditional plant protection methods. Thus, the aim of the present study was to mine the effect of selected biostimulants and plant protection products (PPPs) on the metabolism and cannabinoid content of Cannabis. The newly introduced, high in cannabigerol hemp variety “Enectarol”, was cultivated in a greenhouse. Four biostimulant products [amino acids, chitosan, harpin proteins ($\alpha\beta$), seaweed extract] and a PPP, potassium phosphonates, were applied as recommended. Throughout the course of the cultivation, samples were collected for metabolomics. Two different platforms were employed; GC/MS and a ^1H NMR spectrometer and global metabolomics analysis was performed. Bioinformatics analysis highlighted the differences among the metabolomic profiles of the plants following the biostimulant and PPPs applications and corresponding metabolites-biomarkers of their effect on the physiology and defense mechanisms of the plant (e.g., linolenic acid) were discovered the and on its bioactivity (e.g. cannabinoids). Treatments substantially affected the levels of metabolites such as the cannabinoid cannabidiol, amino acids such as isoleucine and valine, and several Krebs Cycle intermediates, which signify an alteration of plants' energy equilibrium. The findings provide insights into the potential application of biostimulants for enhancing cannabis plant protection, as well as for the qualitative and quantitative improvement of the final product(s).



Figure 1 Pipeline of the experimentation

SE47 010

IN PLANTA INVESTIGATION OF THE MECHANISM OF ACTION OF A BACILLUS SP. ENDOPHYTE APPLYING METABOLOMICS

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The issues that the plant protection sector is facing dictate the need for the exploitation of new sources of bioactivity and the implementation of alternative strategies. Recently, endophytic microorganisms have become the focus of research, mainly due to the multifaceted interactions they develop with their hosts, including the production of a vast array of bioactive metabolites, and their capacity to elicit the plants' defense mechanisms. Within this context, here, an olive tree endophytic *Bacillus* sp., previously isolated and assessed regarding its metabolite producing capacity and bioactivity against olive tree pathogens, was applied to olive trees. The aim of the study was to investigate in planta its mechanism of action and its potential to be developed as a biostimulant or biopesticide (Figure 1). Large-scale cultivation of the bacterium was performed in liquid cultures. A bacterial preparation (10^6 cfu·mL⁻¹) was obtained during its exponential phase of growth, which was applied to young olive trees via foliar spraying and by drenching. Leaf sampling was performed two- and five-days post-treatment. The effect of the endophyte on olive tree metabolism was studied employing a combination of GC/MS, ^1H NMR, and LC/Orbitrap platforms for metabolomics analysis. Performing bioinformatics analysis, several metabolites-biomarkers were discovered. Based on the findings, it seems that the isolate being studied causes plants' priming, and also changes in their energy equilibrium and overall metabolism. Such results confirm the potential of this olive tree isolate for further development as an alternative plant protection agent to the currently applied biological control agents and conventional plant protection products. Furthermore, as a proof-of-concept, the proposed pipeline is highly foreseen to become a primary plant protection strategy, integrating well with the concept of sustainability and the European Green Deal roadmap.

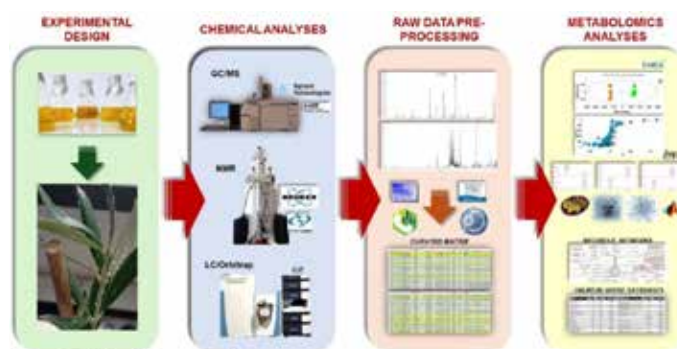


Figure 1. Pipeline of the experimentation

BANQUET

18.00-20.30 Concurrent Session **48**

Phytoplasmas and Viroids in World Agriculture

Chair: Assunta Bertaccini (*Alma Mater Studiorum University of Bologna, Italy*)

SE48 C01

CITRUS VIROIDS: FRIENDS OR FOES?

Georgios Vidalakis¹, Irene Lavagi-Craddock¹, Tyler Dang¹, Sohrab Bodaghi¹, Fatima Osman¹, Stacey Comstock¹, Michelle Ortiz¹, Paulina Quijia-Lamina¹, Deborah Pagliaccia¹, Ashraf El-Kereamy¹, Subhas Hajeri², Valeria Lavagi¹, Jonathan Kaplan³, Andres Espindola⁴, Kitty Cardwell, Grant Chambers⁵, Nerida Donovan⁵

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Since the discovery of the first citrus viroid in the early 1970s by Semancik and Weathers, research on viroids at the University of California, Riverside has continued unabated, focusing on their detection, biology, and potential beneficial uses in citriculture. To date, eight viroid species have been identified in citrus. Of these, only two are associated with the severe, economically significant diseases of exocortis and cachexia, which affect sensitive citrus hosts such as trifoliate and mandarins, respectively. All other viroid-citrus host combinations appear to be harmonious, echoing the original 1946 idea by Altenburg, who introduced the concept of "viroids" as "symbionts akin to viruses". Certain viroid-host combinations may prompt citrus trees to undergo dwarfing and reduced canopy volume, which can be deemed beneficial from a citricultural and economic standpoint, especially since dwarfed trees are suitable for high-density plantings and citrus under protective screens (CUPS). Such agricultural innovations are more crucial now than ever due to threats of catastrophic diseases like Huanglongbing (HLB) and the scarcity of agricultural land, water, and labor, which demand increased production per unit of land area. Our laboratory has conducted field, molecular (miRNA and transcriptome), and economic studies to link observed viroid-induced dwarfing phenotypes to their underlying molecular mechanisms and potential economic benefits for commercial application. Whether studying citrus viroids as pathogens or dwarfing agents, developing technologies for viroid RNA preparation and detection remains critical. Such research supports the production of viroid-free citrus nursery stock and the successful deployment of any beneficial viroid-RNA technology at a large scale. In our laboratory, the pursuit of advanced viroid detection technologies has pushed the boundaries of biological indexing and quantitative polymerase chain reaction (qPCR) assays and has produced high-throughput sequencing (HTS) based platforms for the routine detection of citrus viroids within metagenomic datasets.

SE48 C02

PHYTOPLASMA ASSOCIATED DISEASES: THE TIP OR THE BOTTOM OF PLANT PATHOLOGY ICEBERG?

Assunta Bertaccini

Alma Mater Studiorum University of Bologna, Bologna, Italy

Phytoplasmas associated with hundred of plant diseases and strongly reducing agricultural production in several areas of the world. They are transmitted by insects, propagation materials and seeds and very often their presence is not recognized and allows plants to be more prone to other pathogens or diseases. The identification of transmission routes and phytoplasma strains is achieved by a provisional molecular taxonomy distinguishing them into different '*Candidatus Phytoplasmas*'. Generally diverse phytoplasmas are associated with the different diseases, however in several cases the same disease is associated with different phytoplasmas. The "flavescence dorée", apple proliferation ('*Candidatus Phytoplasma mali*') are only reported in Europe and associated with one phytoplasma having different strains with diverse geographical distribution and variable pathogenicity. On the contrary the coconut lethal yellowing is associated with phytoplasmas that are different according to the geographic distribution. '*Ca. P. palmicola*' is present in Africa, '*Ca. P. palmae*' in America and '*Ca. P. novoguineense*' in Oceania and there are also other phytoplasmas infecting palms. The citrus and alfalfa witches' broom diseases are mainly present in Asian countries associated with '*Ca. P. aurantifolia=citri*' and with a number of diverse phytoplasmas, respectively. From these quick overview it appears clear as the management of these diseases must be based on knowledge of phytoplasma identity, insect vectors and dissemination ways. The use of environmentally friendly advanced tools that enable to enhance the plant resistance to pathogens such as plasma activated water (PAW) together with the increased knowledge about microbiomes of healthy and phytoplasma-infected plants are among the possible pathways to help in mitigating the impact of phytoplasma-associated diseases in a sustainable and ecofriendly manner.





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SE48 C03

DECIPHERING THE INTERACTIONS BETWEEN POSPIVIROIDS AND THEIR HOSTS

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Viroids are minimal, single-stranded circular naked RNA pathogens that infect numerous plant species. They do not encode any proteins, therefore they completely rely on their host machinery to move and replicate. We have studied extensively the cross-talk between viroids and the host's RNA silencing responses. Further we have identified VIRP1 in tomato as a strongly interacting host partner for Potato Spindle Tuber Viroid (PSTVd), specifically binding to PSTVd RNA through an atypical RNA-binding proline-rich domain. The resistance of *VIRP1*-suppressed tobacco plants to PSTVd mechanical inoculation suggested that this protein is essential for viroid infectivity and there are indications that VIRP1 could mediate viroid nuclear import. VIRP1 localizes to the nucleus and carries a bromodomain, which recognizes acetylated histones and translates the "open chromatin" signal to regulation of gene expression. However, its endogenous role still remains unknown. We investigated the effect of different VIRP1 mutations in PSTVd infectivity. In order to further understand the endogenous role of VIRP1, we looked for its interactors, as well as how VIRP1 affects gene expression. Our results indicate that VIRP1 acts as a transcriptional activator and is implicated in responses related to hormone signaling and development. In addition, we have studied the interaction of PSTVd with a group of host non-DICER dsRNases known as RTLs. Results from this work will be presented at the meeting.

of the Class II (TB/CYC) which controls shoot branching and phytochrome. Sodium was found to accumulate in the symptomatic leaves, four times compared to its levels in the asymptomatic leaves. The main findings and the influence of these diseases on acid lime production will be presented.

SE48 001

PHYTOPLASMA-INDUCED WITCHES' BROOM DISEASE OF LIME: CHARACTERIZATION AND MANAGEMENT

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Sultan Qaboos University, Seeb, Oman

Acid lime (*Citrus aurantifolia* L.) is an important crop in different countries, including China, India, Oman and Iran. Witches' broom disease of lime, caused by '*Candidatus* Phytoplasma aurantifolia', is the most serious disease of acid lime in Oman. The disease was observed in Oman in the 1970s. However, due to the delay in diagnosing the causal agent and taking necessary actions to eradicate the disease, witches' broom disease spread to other areas in Oman and was reported in the UAE in the late 1980s. This resulted in the death of about one million lime trees in Oman and the UAE. Our investigations showed that the disease is widespread in most lime growing areas. Disease levels and symptoms were found to vary according to climatic conditions. Investigating phytoplasma transmission showed that leaf hoppers are effective in transmitting phytoplasmas from one tree to the other, while psyllids were less effective. Yeast two-hybrid (Y2H) assay was conducted to study the interaction of SAP11_{WBDL} phytoplasma with acid Lime TCP domains and Arabidopsis TCP transcription factors. Findings from Y2H study showed that SAP11_{WBDL} mediated destabilization



SKALKOTAS

18.00-20.30 Concurrent Session **49**

Endophytes as Bioinsecticides

Chair: Spiridon Mantzoukas (*Department of Agriculture, University of Ioannina, Greece*)

SE49 C01

ENDOPHYTIC ENTOMOPATHOGENIC FUNGI: PAST, PRESENT AND FUTURE PROSPECTS

Spiridon Mantzoukas

SE49 C02

SEARCH OF ENDOPHYTIC ENTOMOPATHOGENIC FUNGI FROM SUBURBAN GREEN SPACES IN ACHAIA

Ioannis Lagogiannis

SE49 C03

ENTOMOPATHOGENIC FUNGAL ENDOPHYTES. THEIR DIVERSITY IN GREECE

Z. Gonou-Zagou

SE49 C04

EFFECTS OF THE ENTOMOPATHOGENIC ENDOPHYTE BEAUVERIA BASSIANA ON GROWTH AND PHOTOSYNTHETIC PERFORMANCE OF THREE BRASSICA OLERACEA VARIETIES

Foteini Kitsiou

SE49 C05

ENDOPHYTES: ENTOMOPATHOGENIC FUNGI AS AN EMERGING BIOLOGICAL CONTROL AGENTS OF FOREST AND AGRICULTURAL PLANT

Panos V. Petrakis

SE49 C06

ENDOPHYTIC ENDOMOPATHOGENIC FUNGI: WHAT COMPARATIVE GENOMICS OF METARHIZIUM BRUNNEUM REVEAL

Vassili Kouvelis

MC3

18.00-20.30 Concurrent Session **51**

Interactions between Plant Protection and Pollinators

Chair: Fani Hatjina (Department of Apiculture, ELGO DIMITRA, Greece)

Antonios Tsagkarakis (Agricultural University of Athens, Greece)

SE51 C01

CHRONIC EXPOSURE EFFECTS OF SUFLOXAFLOR AND IMIDACLOPRID UNDER LABORATORY AND FIELD CONDITIONS

Fani Hatjina, Leonidas Charistos and collaborators

Department of Apiculture, Ellinikos Georgikos Organismos 'DIMITRA', Nea Moudania, Greece

Neonicotinoids are globally the most widely used insecticides. Sufloxaflor is one of the sulfoximines which constitute a highly effective group of pesticides against a wide range of sap-feeding insect. Imidacloprid is also one of the most used insecticides belonging to the family 'neonicotinoids', at least before the European ban. However, both pesticides also affect the non-target insects such as the honey bee. In Department of Apiculture- ELGO DIMITRA, several laboratory and a field experiments have been conducted, where sufloxaflor and imidacloprid, independently, were administered via the food. Sufloxaflor was fed to the bees at a concentration of ng/kg in sugar solution or in both sugar solution and pollen patties but by using a 10 times higher concentration in pollen. Imidacloprid was fed to bees at a concentration of 2 ng/kg sugar solution and 3 ng/kg in pollen paste during one month. For the laboratory experiment, bee survival, rate of mortality, amount of consumed food, the size of the HPGs and the amount of fat body was assessed. For the field experiment, colony population, amount of brood and dead bees in front of the colonies was assessed. For sufloxaflor, the preliminary results have shown no difference in any variable between treated and control bees in the laboratory but a significant difference was detected between treated and control colonies in terms of amount of brood. Most of treated colonies did not manage to overwinter successfully. Fat body mass measurements showed that the actual mass of fat was not significantly different between the groups, however, the dry weight of the abdomens before and after the removal of the fat was higher in the control group than in the treated groups. For imidacloprid the results showed that the observed changes were influenced by the mode of intoxication (chronic or acute), meaning that the chronic exposure to imidacloprid decreased more the diameter of the acini, than the acute exposure always compared to control bees. The exposure to imidacloprid resulted also to significant alterations in the expression profile of the isoenzymes of GST group. The results demonstrate the adverse sublethal effects of both substances, which depend on the mode of action. Especially for sufloxaflor, further experiments are needed to demonstrate the real impact on honey bee colonies and bee's physiology and behaviour.

SE51 C02

HONEYBEES AS MULTIROLE AGENTS IN ORANGE TREE ORCHARDS

Melina Georgitsi, Antonios Tsagkarakis

Laboratory of Sericulture and Apiculture, Agricultural University of Athens, 75 Iera Odos str., 118 55 Athens, Greece

Apis mellifera L., the European honeybees, are important pollinators, as they are responsible for the reproduction of more than 300 different plant species. The present study aimed to investigate the role of honeybees in quality and quantity of the citrus production, as well as their potential impact to the population of crop pests. For this reason, 100 beehives were placed in a 10 ha orange orchard (cv Navellina, 30 years old) at Koutsopodi, Argolis. The beehives remained there during the whole tree blossom. Afterwards, all of them were removed except of 10, which remained for the whole sampling season, in order to investigate the impact of the honeybee presence on insect-pest population. To observe the impact of honeybees on orange pollination, fruit samplings were performed after the ripening, in which 100 fruits (10 fruits from each of 10 trees) were collected and measured for weight and diameter. The same sampling procedure was performed in another orange orchard (5ha, cv Navellina, 25 years old), which was located at Dalamanara, Argolis, 5 km SW in distance from the first orchard, close to which no beehives had been installed, and which was serving as untreated control. Both orchards received the same cultivation techniques during the whole sampling season. For the estimation of the insect pest population, fortnightly leaf samplings were performed from both orchards. In each sampling, 4 sprouts (10 leaves in average) were collected from each of 10 trees (40 sprouts - 400 leaves in total) of each orchard and the population of the insects was recorded. Results showed that fruits harvested from the orchard with honeybee presence were significantly bigger in weight and diameter, compared with those harvested from the untreated control. Furthermore, significant differences found in the insect pest populations between the honeybee-treated orchards, which will be presented and discussed in details during the presentation.

SE51 C03

EXPLORING POLLINATION SERVICES THROUGH A SUPPLY-DEMAND APPROACH

Giorgio Sperandio^{1,2}, Anna Simonetto¹, Andrea De Francesco¹, Paola Gervasio¹, Paola Riolo², Fani Hatjina³, Gianni Gilioli¹

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Pollination services are crucial for agriculture, biodiversity, and food security worldwide. Declining insect populations pose a threat to this essential service. While honey bees are important pollinators for many crops, studies emphasize the significant contribution of wild pollinators to effective pollination and crop yield. In this work we developed an Individual-Based Model (IBM) that allows exploring pollination services provision by honey bees and wild pollinators in realistic in-silico landscapes. The IBM is composed by several modules (Figure 1) simulating i) the stage-specific physiological responses of honey bees and the population dynamics of the hive, ii) the phenology, the dynamics of food provision and the pollination requirements of the landscape, and iii) the distribution, phenology and dynamics of wild pollinators. The landscape is considered as a composition of food patches characterised by different vegetation types. Based on literature data, vegetation types were further characterised by specific flowering period(s), food production and supply, and pollination requirements. Wild pollinators are considered as a diffuse individual, whose site-specific density is given by the vegetation type and the season. Different groups of wild pollinators are considered in the model based on body size and foraging range. The model allows exploring different management scenarios, identifying the number of honey bee colonies that can be economically viable to place in a given area, ensuring the presence and the diversity of wild pollinators communities and the effective delivery of pollination services. The model can be implemented as a decision-support tool for identifying optimal trade-offs between agricultural production, preservation of biodiversity and ecosystem services, while ensuring economic viability for beekeepers and farmers.



Figure 1: conceptual scheme showing the main modules of the IBM and its potential uses. This work has been carried out within the PRIMA Project SafeAgroBee

SE51 C04

A MODELLING TOOL SUPPORTING SUSTAINABLE BEEKEEPING MANAGEMENT PRACTICES

Andrea De Francesco¹, Giorgio Sperandio^{1,2}, Anna Simonetto¹, Paola Gervasio¹, Fani Hatjina³, Gianni Gilioli¹

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Apiculture provides essential ecosystem services through honey and hive products production, along with pollination of crops and wild plants. Sustainable beekeeping requires consideration of the interplay between the pollination demand of entomophilous species and the availability of nectar and pollen in landscapes comprising both crops and natural habitats. While resource availability is time-limited for crops, natural habitats tend to represent a more stable and diverse source of food. In cases of resource limitations, competition between honeybees and wild pollinators may lead to shifts in floristic composition and biodiversity loss in natural habitats due to a decline in abundance and functional diversity of specialized wild pollinators that are crucial for the reproduction of plant species that have coevolved with them. To address these complexities, we developed an innovative Individual-Based Model capable of solving the supply and demand dynamics of hives, ultimately determining the carrying capacity of a landscape based on its composition. The model assesses the optimal number of hives needed to fulfil pollination demands for both cultivated crop and natural habitat patches while accounting for the presence of wild pollinators. Results from the model runs demonstrate the feasibility of achieving sustainable beekeeping. Honeybees and wild pollinators participate to the satisfaction of the pollination demands of both cultivated and wild plants, ensuring agricultural production without compromising plant biodiversity in natural habitats. Additionally, the model's ability to compute in-hive product dynamics ensures the economic viability of beekeeping operations while sustaining hive health. In conclusion, this research contributes to the ongoing dialogue on sustainable beekeeping practices that balance economic viability with the preservation of the wild pollinators' functionality. The innovative modelling approach presented here provides a valuable framework for evaluating management strategies that optimize ecosystem services provided by honeybees while safeguarding the vital contributions of wild pollinators.



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SE51 C05

THE HEALTH STATUS INDEX: A HOLISTIC TOOL FOR THE SUSTAINABLE BEEKEEPING

Anna Simonetto¹, Giorgio Sperandio^{1,2}, Andrea De Francesco¹, Paola Gervasio¹, Fani Hatjina³, Gianni Gilioli¹

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Honeybees play a crucial role in providing ecosystem services like food (e.g., honey, beebread, and royal jelly) and pollination essential for maintaining biodiversity and food security. The provision of these services is heavily influenced by the health status of the bees. The health status of a colony of bees is highly impacted by heterogeneous factors including environmental conditions, chemical and biological stressors, beekeeper conditions and practices, and agricultural practices. This complexity makes it challenging to implement management strategies that both preserve honeybee health and ensure the productivity and economic viability of beekeeping. In this work, we developed a Health Status Index (HSI) that allows the exploration of relationships between external drivers (environmental conditions and beekeeping practices) and six health dimensions. The HSI constitutes a Partial Least Squares Path Modeling (PLS-PM) framework incorporating a second-order structural equation model (SEM). We defined five core dimensions of colony health, constituting the first level of the model: Queen, Inhive Product, Disease and Infections, Demography, and Behaviour. Honeybee health is conceptualized as a second-order construct. Additionally, the model encompasses the assessment of three latent dimensions associated with external drivers: the Resource Providing Unit (RSU), the Environment (mainly related to climate), and Beekeeping Management Practices. The model has been calibrated on data produced throughout the SafeAgroBee PRIMA project, integrated with literature. The model was able to discriminate the health status of the honeybee colonies according to the characteristics of each scenario. The HSI provided a good capacity to integrate different types of data.



CONFERENCE I

18.00-20.30 Concurrent Session 52

Frost Damage Mitigation Strategies for Crops, Organized by Project LIFE-FROSTDEFEND

Chair: Dimitris Georgakopoulos (Department of Crop Science Laboratory of General and Agricultural Microbiology, Agricultural University of Athens, Greece)

SE52 C01

ANOTHER WAY TO REDUCE THE RISK OF FROST BY MONITORING AND MAINTAINING MOIST SOILS CLOSE TO FIELD CAPACITY

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The Association Climatologique de la Moyenne Garonne et du Sud-Ouest (ACMG) is an association of farmers started in the south-west of France in 1959 for fighting climate scourges as frost, hail, drought, and now global heating. Over the last ten years, the earlier start of vegetation, caused by global warming, has caused serious frost as in 2021 and 2022 on vineyards and orchards in Western Europe. At the request of the wine unions of Saint-Emilion, Lustrac and Moulis, we carried out two aerial remote sensing missions by plane with an infrared camera over vine production areas during two very frosty nights. The images of a 2 m pixel were analyzed in the field with the producers in order to understand the temperature differences on the ground which vary by several degrees. We have observed that for a same management with permanent grass in the middle of the row, there is a difference of up to more than 3°C between moist soil, close to field capacity, and soil dried out superficially by the grass. As part of the An-Gel Sudoe (Interreg) and ADRC (New Aquitaine Region) projects, we are testing the hypothesis that a moist soil, due to better thermal conductivity, makes it possible to store more calories during the day which rise to the surface the following night, which further reduces the risk of frost. If the hypothesis is confirmed, this means that it is possible to reduce the risk of frost in a semi-passive manner by monitoring the humidity of the soil and regularly applying sprinklers in addition to the rain for maintaining the higher possible the thermal conductivity of the soil.

SE52 C02

THE LIFE-FROSTDEFEND PROJECT: CREATION OF A FORECASTING TOOL FOR FROST DAMAGE RISK AND MITIGATION ACTIONS FOR TREE CROPS

Dimitrios G. Georgakopoulos¹, Khalil Geballa-Koukoulas¹, Maria Gini², Prodromos Fetfatzis², Michalis Karvelas⁴, Vaïos T. Karathanos^{4,5}, Panagiotis Gkionis⁴, Dimitrios Kontogiannatos¹, Nicolas Dusart³, Guillaume Charrier³, Georgios Sarigiannidis⁶, Konstantinos Eleftheriadis²

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Climate change induces an increase in climate variability and a higher frequency and intensity of deleterious weather events at global and regional scale. The Mediterranean basin is among the most vulnerable areas in Europe. Rising temperatures in winter and spring can trigger early onset of plant growth, making the plants vulnerable to subsequent frosts. Vegetation is not necessarily damaged when leaf temperature drops below the freezing point of their cell contents. In the absence of a site nucleating formation of ice crystals, the cell sap remains in a supercooled liquid state. Epiphytic ice nucleation-active (INA) bacteria play an important role in crop frost damage because of their ability to catalyze ice formation on plant tissues at temperatures slightly below the melting point of water. In LIFE-FROSTDEFEND project, we investigate the relationship between PM mass concentrations, airborne and epiphytic bacteria populations in lemon orchards, taking into account plant phenological stage and local meteorological conditions, to assess tree vulnerability and frost risk. PM mass concentrations were monitored by means of low-cost systems equipped with PM, relative humidity and temperature sensors. These sensors were calibrated before installation, and their performance was evaluated. Air filter and leaf samples were analyzed over 2 years to determine bacterial populations. The role of meteorological parameters, as well as the long-range air mass transport (backward trajectory analysis) were also investigated. Airborne bacterial populations show a positive correlation with PM concentrations. Sahara dust events were associated (most times) with increased PM mass concentrations and bacteria populations. Epiphytic bacterial populations presented a complex fluctuation throughout the year, but a good correlation to average meteorological conditions and PM concentrations up to two months before

a measurement. An algorithm to forecast epiphytic INA bacterial populations as a frost damage risk has been created and incorporated in the FROSTDEFEND app.

This work is supported by LIFE FROSTDEFEND (LIFE20 CCA/GR/001747)

SE52 C03

DEVELOPING FROST RISK INDICATORS FOR APRICOT TREES

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The interaction between a tree's exposure to frost and its vulnerability can result in crop failure and economic losses for a grower. This represents the so called « risk of frost damage » that is more than the simple likelihood of a freezing event and involves complex phenomenon that depend on numerous biotic and abiotic factors. Characterizing the impact of these factors on the risk of frost damage is particularly important for fruit trees, in order to develop new frost protection measures. Quantification of the abundance of ice nucleation active bacteria and immersion tests of ice nucleation activity on dissected and undissected stems and reproductive organs of apricot trees are providing a better understanding of the conditions leading to ice formation in apricot tissues. Our results show that ice nucleation active bacteria are not involved in ice nucleation of plant material. These results contribute to distinguishing between the role of intrinsic factors (i.e. linked to the plant itself) and extrinsic factors (i.e. linked to ice nucleation active microorganisms) in the nucleation process.

SE52 C04

STRATEGIES AND FACTORS FACILITATING THE REDUCTION OF BACTERIAL ICE NUCLEI ON PLANTS FOR THE CONTROL OF FROST INJURY TO SENSITIVE PLANTS

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Endogenous ice nuclei capable of limiting the supercooling of plants at warm subfreezing temperatures ($> -5^{\circ}\text{C}$) are very low in abundance in young tissues of most crop plants and these plants thus can supercool and escape damaging ice formation. Frost control by preventing the establishment of ice nucleation active (Ice⁺) bacteria on these plants is therefore most feasible in crops subject to subfreezing temperatures shortly after their development. The numbers of bacterial ice nuclei on these plants can be reduced by the inoculation of various non-ice nucleation active bacteria on the plants early in their development in the spring, to prevent the establishment of Ice⁺ bacteria by a process of preemptive competitive exclusion. Such a process of frost protection is often more effective than the repetitive application of bactericides such as copper compounds needed to prevent the growth of Ice⁺ bacteria on the plants. Elimination of local sources of immigrant Ice⁺ bacteria, such as from cover crop species in perennial crops such as grape, can dramatically reduce the rate at which Ice⁺ bacterial populations establish on plants, thus achieving frost control. Irrespective of the methods by which Ice⁺ bacteria are reduced on plants, the

reduction of the incidence of frost injury is directly related to the extent to which ice can propagate from one plant part to another. Frost control achieved by reduction of bacteria ice nuclei is maximized when plant canopy density is low and leaves and flowers are not in direct contact, as well as when the duration of subfreezing temperatures are short – both reducing the likelihood that ice nucleated at a given site will propagate and seed ice formation in distal parts of the plant or canopy.

SE52 C05

EPIPHYTIC AND AERIAL ICE NUCLEATION ACTIVE BACTERIA IN LEMON TREE ORCHARDS

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In recent decades, European agricultural production has been significantly challenged by frost events, exacerbated by a confluence of meteorological, agricultural, and microbiological factors within agroecosystems. Notably, epiphytic bacteria of the genus *Pseudomonas* have been implicated in promoting ice formation through their ice nucleation activity (INA), thereby catalyzing ice nucleation in supercooled water. Various factors, including meteorological conditions, aerial migration patterns, and local microclimatic variations, influence the formation of epiphytic INA bacteria within the agroecosystem. This study investigates the seasonal dynamics and INA of epiphytic and airborne microbial communities in two lemon orchards in Aeghion, Greece. Seasonal fluctuations in the abundance of fluorescent ice-nucleating bacterial populations and the corresponding INA at aerial and epiphytic levels were assessed through a multidisciplinary approach integrating diurnal variations in particle pollution and their INA alongside classical microbiological and molecular biological techniques. Our findings reveal that the seasonal population dynamics of aerial and epiphytic bacteria parallel the quantitative fluctuations of aerosol particles, thereby laying the groundwork for developing computational models aimed at predicting the population dynamics of epiphytic INA bacteria and, consequently, the risk of frost damage in orchards. These findings suggest no discernible influence between the season and epiphytic bacteria formation, while not all epiphytic bacteria are INA active. Moreover, we have observed a trend where an increase in fluorescent ice-nucleating bacterial populations inversely correlates with ice nucleation activity at an estimated ratio of 100 bacteria per INA. Furthermore, experimental applications of multiple copper sprays in lemon orchards at higher altitudes demonstrated a notable reduction in INA activity, thereby indicating the potential efficacy of such



treatments in mitigating epiphytic INA bacterial populations and ice nucleation activity, thereby mitigating the risk of frost damage. These findings will elucidate the frost phenomenon affecting lemon trees and could pave the way for compelling future protective actions.

SE52 C06

UNDERSTANDING FROST VULNERABILITY IN CITRUS CROPS: EXPLORING THE INTERPLAY OF PLANT SUSCEPTIBILITY AND ICE-NUCLEATING BACTERIA

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Despite being mainly cultivated in subtropical and tropical regions, periodic freezing events have historically posed threats to citriculture in more temperate areas, as witnessed in historical events in Florida (1985) and Greece (2004). Global warming has extended the potential growing area including frost exposed area. *Citrus* sp are not frost-tolerant but are still able to survive mild freezing events by maintaining intracellular water in a supercooled state. This ability could be mitigated by the abundance of ice nucleation agents, among which Ice Nucleation Active (INA) bacteria are a critical factor. Indeed, epiphytic bacteria of the genus *Pseudomonas* could catalyze ice nucleation above and within plant tissues, increasing the ice nucleation temperature by several degrees. This study aimed to unravel the intricate relationship between plant response to temperature and INA bacteria, shedding light on the multifaceted factors influencing citrus frost damages. Notably, this study explored the susceptibility of evergreen citrus trees to intrinsic ice formation, with a focus on the acclimation to temperature, leaf morphological traits and the abundance of INA bacteria. We determined the relation between temperature and the ability to acclimate and deacclimate, predicting the sensitivity to freezing events. Although a critical threshold of -6°C was identified. Moreover, the presence of nucleating agents above the leaves could alter the pattern of freezing. This research provides crucial insights into the factors affecting Citrus frost vulnerability and contributes to our understanding of how changing climatic conditions may affect citriculture.

Funding: This research is funded by the European Union under the Life-Frostdefend project (LIFE20 CCA/GR/001747)



**14th IVS
SESSIONS**



CONFERENCE I

08.30-11.00 14th IVS 1

Recent Advances in Verticillium Biology through Genomics

Chair: Krishna Subbarao (Plant Pathology, University of California, Davis, USA)

SE01-14th IVS C01

MECHANISMS OF COTTON LEAF DEFOLIATION CAUSED BY VERTICILLIUM DAHLIAE DEFOLIATING STRAIN

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Verticillium wilt caused by *Verticillium dahliae* is a major disease that threatens cotton production worldwide. Based on the symptoms caused on cotton, *V. dahliae* strains can be divided into defoliating and nondefoliating pathotypes. Defoliating pathotype is able to completely defoliate cotton, leading to a significantly higher disease severity and greater yield loss than the nondefoliating pathotype. Elucidating the mechanisms underlying defoliation caused by *V. dahliae* is of vital importance. In our study, the auxin, jasmonic acid and cytokinin signaling pathways negatively regulated defoliation, while ethylene (ET) and salicylic acid (SA) signaling pathways played a positive role in defoliation. Furthermore, to identify secreted proteins that are involved in defoliation, the *V. dahliae* defoliating strain XJ592 and a nondefoliating strain XJ511 were sequenced, and 47 genes encoding secreted proteins were comprised of different sequences between the two pathotypes. Among them, *VdSAP* could induce cell death on *Nicotiana Benthamiana* leaves. Both the defoliation rate and disease index of cotton inoculated with *VdSAP* deletion strain were significantly lower than in plants inoculated with the wildtype strain XJ592, indicating the indispensable role of *VdSAP* in defoliation. *VdSAP* also interacts with cotton GhARP protein and inhibits its accumulation in vivo and in vitro. In this study, GhARP was proven to not only negatively regulate defoliation via *IDA* (Inflorescence Deficient in Abscission)-induced abscission signal pathway, but also inhibit the expression of genes encoding ET pathway related enzymes, thus playing a negative regulatory role in the ET pathway. Our study revealed a model that defoliating strain secretes *VdSAP* protein to restrain the accumulation of cotton GhARP protein, which in turn facilitates cotton defoliation by promoting ET and *IDA* signaling pathways.

SE01-14th IVS C02

MOLECULAR INSIGHTS INTO THE VERTICILLIUM-SMOKE TREE INTERACTION

Chen Tang

Beijing Key Laboratory for Forest Pest Control, College of Forestry, Beijing Forestry University, Beijing 100083, China

Verticillium dahliae is a notorious soilborne pathogen worldwide, causing vascular wilt diseases on a broad range of woody plant species such as olive, ash, and smoke trees

(*Cotinus coggygria*). Although the pathogen side of the *V. dahliae*-smoke tree interaction has been extensively examined at the molecular level, the tree responses have not been thoroughly investigated. In this study, a chromosome-scale genome assembly for *C. coggygria* was obtained, enabling the identification of 878 cell-surface and intracellular immune receptors. A dual transcriptomic analysis (RNA-seq) was carried out on root tissues of smoke tree plants during the early infection stages by *V. dahliae*. Statistical analysis of plant-side revealed that the pattern-triggered immunity and effector-triggered immunity defense pathways were downregulated in *C. coggygria*, and the major temporal changes in plant gene expression during *V. dahliae* infection. By comparing the transcriptome data across different hosts infected by *V. dahliae*, certain shared pathways encompassed in disease resistance and defense mechanisms were identified. An RNA-Seq dataset on the pathogen side demonstrated that the largest change in *V. dahliae* gene expression occurs at 2 dpi, with most transcription factor-encoding genes being repressed after this time. We characterized 42 effector-encoding genes highly induced during *V. dahliae* infection, and preliminary planning is underway to explore their function. This research will aid in advancing crucial knowledge on Verticillium-host interactions on early immune transcriptional reprogramming, and will uncover important components of plant immunity, providing a theoretical basis for disease resistance breeding of tree hosts.

SE01-14th IVS C03

SECRETOME: THE MIRACULOUS ARSENAL OF VERTICILLIUM DAHLIAE DURING HOST INFECTION

Xiao-Bin Ji¹, Steven J. Klosterman², Krishna V. Subbarao³, Jie-Yin Chen^{1,4} and Dan-Dan Zhang^{1,4}

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Fungal pathogens employ their secretomes to modulate plant defenses to facilitate colonization. *Verticillium dahliae* encodes more than 700 potential secreted proteins, and hundreds are delivered into the extracellular space with generalist functions. Currently, around 40 secreted proteins have been shown to contribute to pathogenesis via diverse modes of action. Among these, effectors manipulating host immunity are key determinants of successful infection by this pathogen. In filamentous phytopathogens, effectors manipulating host immunity via alterations to the subcellular



localization of their host targets are prevalent. For example, some effectors can promote a particular subcellular localization of a host target or prevent the normal host protein localization pattern during defense. Effectors disturbing plant immunity by directly mediating nuclear export of host resistance receptors are rare. Our research has shown that six disparate secreted proteins of *V. dahliae* that simultaneously include nuclear localization signals and nuclear export signals and are capable of spontaneous nucleo-cytoplasmic transport in the plant cell. These effectors mainly play roles in pathogenesis by interfering with their corresponding nuclear export receptors, thereby disrupting downstream signaling and gene expression. This is a novel finding, since the effector hijacks plant immunity by mediating host resistance receptor nuclear export by disrupting the host nucleocytoplasmic transport system. The transport pathway has likely been co-opted by plant pathogens during host-pathogen co-evolution to promote virulence.

SE01-14th IVS C04

COMPARATIVE GENOMICS DISTINGUISHES MICROSCLE- ROTIA-PRODUCING AND NON-MICROSCLEROTIA-PRO- DUCING VERTICILLIUM SPECIES

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The phytopathogenic fungus *Verticillium dahliae* produces resting structures, known as microsclerotia (MS), that enable it to survive for years in the soil. Although signaling components, a hydrophobin, and transcription factors required for MS production have previously been characterized in *V. dahliae*, there is little known of the differentiation between MS-producing species of *Verticillium* and those that do not produce MS. In this study, comparative genomics analyses were undertaken to identify sequences present in *V. dahliae* and absent in non-MS producing *Verticillium* species. Comparative whole-genome alignments in this study included three genomes of *V. dahliae*, one of *V. alfalfae*, and two of *V. nonalfalfae*. The resting structures of *V. alfalfae* and *V. nonalfalfae* are dark resting mycelia, rather than the discreet MS of *V. dahliae*. Sequences unique to *V. dahliae* were queried against the genomes of *V. klebahnii*, *V. nubilum*, and *V. tricorpus*. *V. klebahnii* produces MS, *V. nubilum* produces chlamydospores exclusively, and *V. tricorpus* produces MS, dark resting mycelia, and chlamydospores. In addition, cDNAs expressed during microsclerotia development in *V. dahliae* were aligned with the identified sequences. Some candidate genes identified were deleted and the knockout strains were assessed for MS production. Fundamental analyses to identify sequences required for MS production may yield new insights into *Verticillium* wilt control.

SE01-14th IVS 001

THE KSS1 OF VERTICILLIUM DAHLIAE REGULATES VIRULENCE, MICROSCLEROTIA FORMATION, AND NITROGEN METABOLISM

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Verticillium dahliae is a pathogenic fungus causing vascular wilt diseases in over 200 plant species globally, impacting both economically important crops and ornamental trees. The fungus's ability to form melanized microsclerotia (MS) allows it to persist in soil for extended periods, making it challenging to control once established. Previous research identified the mitogen-activated protein kinase VdSte11 (MAPKKK) as crucial for MS formation, penetration, and virulence in *V. dahliae*. This study aims to characterize two MAPK homologs, VdSte7 and VdKss1, in the fungus. The study involved identifying and characterizing VdSte7 and VdKss1 in *V. dahliae*. Deletion mutants of VdSte7 and VdKss1 were generated to assess their roles in MS formation and virulence. Phosphorylation assays were conducted to investigate interactions between VdSte11, VdSte7, and VdKss1. Proteomic and phosphoproteomic analyses were employed to explore changes in sterol biosynthesis and nitrogen metabolism associated with the deletion of VdKss1. Deletion of VdSte7 or VdKss1 led to significant impairments in melanized MS formation and virulence in *V. dahliae*. Phosphorylation assays indicated that VdSte11 and VdSte7 can phosphorylate VdKss1. Proteomic analysis revealed alterations in sterol biosynthesis upon VdKss1 deletion, while phosphoproteomic analysis implicated VdKss1 in regulating nitrogen metabolism. VdKss1 MAPK signaling regulates nitric oxide (NO) resistance and inorganic nitrogen metabolism. Additionally, VdRlm1 was identified as a potential downstream target of VdKss1, involved in ammonium nitrogen utilization. This study sheds light on the regulatory protein network in *V. dahliae* affecting MS formation and nitrogen metabolism. The findings enhance understanding of the fungal pathogenesis mechanisms and may contribute to the development of strategies for controlling *Verticillium* wilt diseases.

SE01-14th IVS 002

UNRAVELING THE ROLE OF MITOCHONDRIAL EF-TU IN THE PATHOGENESIS AND HEAT STRESS ADAPTATION OF THE VASCULAR PATHOGEN VERTICILLIUM DAHLIAE

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Elongation factor Tu (EF-Tu) is a well-studied and abundant protein in bacteria, playing a crucial role in protein biosynthesis during translation. While EF-Tu is commonly associated with prokaryotes, it is also found in eukaryotes localized within plastids. In eukaryotes, such as plants, EF-Tu exhibits a moonlighting effect, contributing to heat stress resistance. It is also known that an epitope of EF-Tu, elf18, triggers innate immunity of Brassicaceae plants, through recognition by receptor EFR. Despite extensive studies on bacterial EF-Tu, there is a noticeable gap in the literature regarding fungal mitochondrial EF-Tu, particularly in its interactions with host plants and its role in heat resistance. Our study



aimed to investigate the localization of mitochondrial EF-Tu in *Verticillium dahliae* cells, its impact on heat resistance, and its role in the pathogenicity of Arabidopsis. To determine the protein's cellular localization, a C-terminal GFP fusion of mitochondrial EF-Tu (mEF-Tu) was overexpressed in *V. dahliae* race 1 tomato strain 7OV and examined using confocal microscopy. Pathogenicity experiments were conducted with the transformed *V. dahliae* strains on targeted genotypes of Arabidopsis plants. Additionally, the vitality of fungal spores after heat-shock treatments was assessed. Our results confirmed the expected localization of mEF-Tu in mitochondria and exosomes. Intriguingly, pathogenicity experiments unveiled a susceptibility role of the Arabidopsis EFR receptor during interaction with *V. dahliae*. One possible hypothesis is that the pathogen manipulates plant immune reactions by disrupting hormone homeostasis, particularly through the overexpression of ethylene. Furthermore, the study revealed that overexpression of EF-Tu in the pathogen enhances its ability to overcome heat stress.



CONFERENCE I

11.30-14.00 14th IVS 2

Recent Advances in Verticillium Wilt Management

Chair: Krishna Subbarao (Plant Pathology, University of California, Davis, USA)

Franco Nigro (Soil, Plant and Food Sciences, University of Bari Aldo Moro, Italy)

SE02-14th IVS C01

INTEGRATED MANAGEMENT OF THE SMOKE TREE WILT

Yonglin Wang

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Smoke tree, *Cotinus coggygia*, is one of the most important plant species cultivated in landscape ecology in China and providing the picturesque red-leaf scenery in Beijing during autumn. Verticillium wilt, caused by *Verticillium dahliae*, is a serious problem in the smoke tree-growing regions, causing economic losses and losses in the highly valued scenery. Because of an extended host range and prolonged survival in the soil, Verticillium wilt control is a difficult task. Therefore, implementation of an integrated disease management strategy is required. Few studies have focused on how to combat Verticillium wilt in smoke trees. Herein, three application methods (root irrigation, trunk injection, and both together), as well as one biocontrol bacterium (*Bacillus subtilis*) and 4 fungicides (azoxystrobin, propiconazole, carbendazim, and prochloraz) were selected to examine different treatments for disease control. Analysis of the data from 2021 to 2023 revealed a decreasing trend in the disease index year to year during the application period. Further, root irrigation with propiconazole combined with trunk injection of carbendazim and prochloraz was the most effective treatment within the three years, and this combination was most effective in treating both mild and severe disease. In general, our work established an effective integrated disease management strategy to control smoke tree wilt and provides a technical reference for the control of Verticillium wilt in trees.

fumigation of whole spinach seeds. The treatment entailed the use of 3% NO₂ over a 3-day exposure period. The fumigation treatment significantly reduced both the external and internal *V. dahliae* pathogen, with reductions ranging from 55.6% to 92.9%. The treatment effectively eliminated *P. effusa* from spinach seed. Remarkably this approach did not affect seed germination and showed potential to enhance plant growth. The findings demonstrate that the NO₂ treatment is effective at reducing both *V. dahliae* and *P. effusa* in spinach seeds, is easy to apply, and provides a promising solution for managing seedborne pathogens.

SE02-14th IVS C03

DO WE DEFINE RACES ON THE BASIS OF SINGLE AVR GENES OR ON THEIR PHYLOGENETIC LINEAGE?

Krishna V. Subbarao

University of California, Davis, USA

Following the gene-for-gene model, the exploration of the function of genes governing pathogen avirulence and host resistance has had an unparalleled impact on breeding for resistance in crops against individual pathogens. *Verticillium dahliae* is a soilborne fungal pathogen that causes a vascular wilt on many economically important crops. Based on the resistance or susceptibility of differential cultivars in tomato, isolates of *V. dahliae* have been divided into three races. Although Verticillium wilt resistance is described in many crops, the designation of races has only been possible in tomato and lettuce, owing to the availability of differential cultivars in these two crops. Avirulence (Avr) genes within the genomes of the three races have been identified based on comparative genomics, and their functions characterized. While the availability of the specific Avr genes exemplifying the three races has provided an unprecedented opportunity to identify races in the pathogen populations from hosts in which differential cultivars are unavailable, it also muddles the relationship between races identified via host differentials and Avr genes in a broad host range pathogen such as *V. dahliae*. The races identified via host differential cultivars or via the corresponding Avr genes were congruent in tomato and lettuce until only two races existed. However, this relationship failed when race 3 was defined in tomato as the Avr gene defining race 2 was absent in strains considered race 2 in lettuce. Thus, the previously considered race 2 strain from lettuce remains unassigned to a race currently. This raises the fundamental question on whether we define races based on host differentials or individual Avr genes. How to resolve this discrepancy remains unclear at this time.

SE02-14th IVS C02

NITROGEN DIOXIDE FUMIGATION REDUCES THE VIABILITY OF VERTICILLIUM DAHLIAE AND PERONOSPORA EFFUSA IN SPINACH SEEDS

Steven J. Klosterman¹, Samuel S. Liu¹, Krishna V. Subbarao², Yong-Biao Liu¹

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The fungal plant pathogen *Verticillium dahliae* and the oomycete pathogen *Peronospora effusa* are transported worldwide via global spinach seed trade. Seedborne *V. dahliae* causes Verticillium wilt of mature spinach and so is not a concern on spinach crop production. However, it affects the production of a wide range of crop plant species grown in rotation with spinach, including lettuce. *Peronospora effusa* only infects spinach, causing highly destructive downy mildew disease. This research focused on limiting the introduction of *V. dahliae* and *P. effusa* by nitrogen dioxide (NO₂) gas



SE02-14th IVS C04

RESPONSE TO VERTICILLIUM DAHLIAE INFECTION IN A GENETICALLY RELATED SET OF OLIVE CULTIVARS: PRELIMINARY RESULTS

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Verticillium wilt (VW), caused by *Verticillium dahliae*, is the most significant pathogen in many olive-growing regions all around the world. The most effective and economically efficient control measure for Verticillium wilt in olive is the use of resistant cultivars, forming the foundation of integrated disease management strategies. Despite this, resistance is not widely employed, and the selection of olive cultivars for new orchards is primarily influenced by pedoclimatic conditions, market demands or other phytosanitary considerations. Unfortunately, many cultivars meeting these criteria are susceptible to VW. Due to the outbreak of the olive quick decline syndrome, caused by the quarantine bacterium *Xylella fastidiosa*, only two olive varieties (Leccino and FS17) are allowed for the new plantations in Apulia (Southern Italy). However, the susceptibility of the cv Leccino to Verticillium wilt is well documented in literature, while not much data is available to date on the behaviour of FS17, which is the result of an open pollination cross with Frantoio as a mother plant. This study characterized the responses of several olive genotypes to *Verticillium dahliae* infection under controlled conditions, laying the foundation for future genotype-phenotype association studies. The tested olive varieties included two groups of genetically related cultivars, each exhibiting varying degrees of resistance to Verticillium wilt. The first group included the susceptible Leccino, and the related cvs Pendolino, Leccio del Corno, Cima di Mola, and Mignola; the second group included the resistant Frantoio and the related cvs Ogliarola Barese, Taggiasca, Laurina, and FS17 or Favolosa. Six-months-old self-rooted cuttings were utilized, and plantlets were inoculated by immersing the roots in a conidial suspension (2×10^6 ml) of defoliating and non-defoliating strains of the pathogen for 2 minutes. Disease development was assessed based on the incidence and severity of foliar symptoms, and the area under the disease progress curve (AUDPC) was calculated by using the Percent Severity Index (PSI). The severity of vascular discoloration on the stem section was assessed using an empirical scale (ranging from 0 to 4), after 5 months. The results confirmed the well-known resistance of the Frantoio cultivar and the susceptibility of the Leccino cultivar. However, they also suggested that no clear correlations exist between genetically related cultivars. Notably, among the most relevant findings, FS17 exhibited significantly higher susceptibility compared to the genetically related but resistant Frantoio cultivar.

SE02-14th IVS C05

ESSENTIAL OILS AS A NEW APPROACH TO MANAGE VERTICILLIUM WILT OF TOMATO, EGGPLANT, AND OLIVE UNDER CONTROLLED CONDITIONS

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Managing Verticillium wilt is challenging because of the broad range of hosts on which the disease occurs, and the production of long-lasting resting structures (microsclerotia) by the pathogen on infected hosts that are released into the soil. Controlling this disease necessitates an integrated approach including the consideration of biocontrol products. This study evaluated the impact of essential oils (thyme, oregano, sage, and rosemary) on the severity of Verticillium wilt in tomato, eggplant, and olive. Greenhouse trials were carried out by applying essential oils as foliar treatment at 2% concentration, on tomato, eggplant, and olive plants previously inoculated with a conidial suspension of *Verticillium dahliae* isolates, including both defoliating and non-defoliating pathotypes collected in Apulia (Southern Italy). Plants were inoculated by immersing the roots in a conidial suspension (10^5 CFU/ml) of *V. dahliae* for 10 minutes. Inoculated but untreated plants served as positive controls, whereas non-inoculated plants served as healthy controls. Disease development was assessed based on the incidence and severity of foliar symptoms, and the area under the disease progress curve (AUDPC) was calculated for each treatment and host by using the Percent Severity Index (PSI). The severity of vascular discoloration on the stem section was assessed using an empirical scale (ranging from 0 to 4), five- and three-months post-inoculation for olive plants and the other two crops, respectively. Finally, severity data were converted to PSI. The results of the study demonstrated that thyme and oregano essential oils were effective in reducing disease severity in olives, eggplant, and tomato. Thyme essential oil reduced the disease severity of *V. dahliae* by 50, 46.5, and 28% in tomato, olive, and eggplant, respectively, whereas oregano essential oil reduced the disease severity of *V. dahliae* by 46.7, 32.7, and 20% in tomato, olives, and eggplant, respectively. Further research is necessary to standardize essential oils formulations, application dosages, and rates, which could be used as natural fungicides for the management of *V. dahliae*.

SE02-14th IVS C06

ROOT EXUDATES AND SOIL BACTERIA ARE ANTAGONISTIC REGULATORS OF DORMANCY AND GERMINATION IN *VERTICILLIUM LONGISPORUM* MICROSCLEROTIA

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Verticillium longisporum, the causal agent of Verticillium stem striping in oilseed rape (*Brassica napus* L.) can survive and persist in the soil for many years by its dormant resting structures, microsclerotia. Since microsclerotia formation is a crucial survival mechanism of this pathogen, and there is currently no effective method to control this disease, studying the factors regulating microsclerotia dormancy and germination is of great significance. In experiments assessing the impact of soil microbes on microsclerotia dormancy, microsclerotia were exposed on autoclaved and non-autoclaved soil surfaces at varied soil moisture levels. Results revealed that microsclerotia germination was induced by increased moisture levels but almost completely inhibited in unsterile natural soil, whereas autoclaved soil allowed their germination. To obtain a deeper understanding, we isolated bacteria from the soil and conducted a series of bioassays to assess their effects on microsclerotia. Remarkably, bioassays using two-compartment petri dishes confirmed that the germination inhibiting effect on microsclerotia was attributed to bacterial volatile compounds. Further analysis of bacterial volatiles and bioassays with individual volatile compounds revealed that volatile fatty acids are likely to be the crucial factors determining dormancy of microsclerotia. To explore germination-inducing factors, microsclerotia were first treated with soil bacteria, followed by exposure to plant root exudates and their polar/non-polar fractions. Results indicated that both host and non-host plant root exudates counteracted bacterial suppression, with polar fractions exhibiting stronger stimulation. Subsequent analysis and bioassays pinpointed glutamic acid within the polar fraction to be the most potent stimulant. Moreover, root exudates were found to rescue microsclerotia from bacterial suppression by reducing production and emission of volatile fatty acids by bacteria. Our research provides the basis for novel strategies of control of Verticillium stem striping by managing the soil inoculum.

this study was to isolate bacteria from healthy olive groves, as we hypothesized that there is a factor that can protect them from the soilborne pathogen *Verticillium dahliae*. A collection of 128 isolations of endophytic rhizobacteria from olive roots were obtained and tested in vitro, regarding their ability to inhibit the fungal growth at dual cultures in petri dishes with PDA substrate under standard conditions. 15 bacterial isolates reduced *V. dahliae* development by 50-58% and other 18 achieved inhibition rates of 60-68%. Subsequently, in planta experiments were conducted on eggplants, to determine if any of the most effective bacterial isolates were able to reduce the Verticillium wilt severity in greenhouse conditions. We observed that 5 of them were able to reduce significantly the virulence and symptom development of *V. dahliae* in eggplants. The biocontrol agent BK21 yielded an overall advantage to the plants considering the delay (10 days) and the severity (46.24% milder) of the symptom development and the general physiological characteristics about stem and root development. Judging by the above results, great possibilities can potentially occur in biological management of Verticillium wilt using beneficial rhizobacteria.

SE02-14th IVS 001

BIOLOGICAL CONTROL OF *VERTICILLIUM DAHLIAE* USING ENDOPHYTIC RHIZOBACTERIA FROM OLIVE ROOTS

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Verticillium wilt is one of the most destructive diseases of olive trees and plenty of other plant species. Due to the vascular nature of this disease, it is not feasible to be managed with conventional registered chemical treatments. Taking this into account, we turned to biological control methods which can potentially manage this pathogen both directly and/or indirectly by inducing the plant defence system. The goal of



CONFERENCE I

18.00-20.30 14th IVS 3

Recent Advances in Verticillium Biology - Plant Interaction

Chair: Epaminondas Paplomatas (Agricultural University of Athens, Greece)

Aliki Tzima (Agricultural University of Athens, Greece)

SE03-14th IVS 001

NLP PROTEIN FAMILY MEMBER OVEREXPRESSION INCREASES VIRULENCE IN VERTICILLIUM DAHLIAE

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Verticillium dahliae is a widespread xylem-invading fungus and the causal agent of vascular wilt in several plant species worldwide. For successful entry into the host and subsequent disease establishment, the pathogen is known to utilize a wide range of virulence factors. The current study furthers our knowledge regarding the function of VdNEP, an NLP protein (Nep1-like protein), as one such factor in plant hosts. Initially, eggplant leaves treated with purified VdNEP showed necrosis. As a next step, overexpression of VdNEP by incorporation of extra gene copies was performed. This led to increased virulence in eggplant, cotton and tomato plants. Since the increase in VdNEP expression did not correlate with the number of inserts, the implication of the genomic context of the insertion sites was considered. Fascinatingly, a transformant derived from a defoliating strain with high VdNEP transcript levels, successfully managed to cause disease symptoms in tomato, something that did not occur with the corresponding wild-type isolate. More specifically, the amount of fungal DNA in greenhouse-grown plants infected with this VdNEP-overexpressing transformant, was significantly higher (22 times), than that in the wild-type infected tomatoes. As an additional step, a VdNEP-EGFP fusion was constructed in order to enable VdNEP localization in fungal cells and during infection. Our findings further support the critical role of VdNEP in *V. dahliae* infection.

This work was supported by the General Secretariat of Research and Technology of Greece under the "SCIENTIFIC and TECHNOLOGICAL COOPERATION between RTD Organizations in GREECE and in the U.S.A." (project 05-NON-EU 46) and the Hellenic Foundation for Research and Innovation (HFRI), under the HFRI PhD Fellowship grant (#1953). S.K. acknowledges the USDA National Institute of Food & Agriculture and Federal Appropriations (project PEN4839).

SE03-14th IVS 002

THE ROLE OF 1-AMINOCYCLOPROPANE-1-CARBOXYLIC ACID (ACC) BIOSYNTHESIS AND DEGRADATION IN THE VERTICILLIUM DAHLIAE-PEPPER INTERACTION

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Verticillium dahliae is a notorious vascular pathogen, causing Verticillium wilt in over 200 host species and leading to extensive crop yield losses. This hemibiotrophic fungus invades the xylem, a nutrient-poor environment in which survival is not straightforward. The aim of our research is to understand how the pathogen manipulates the plant to survive and thrive in the xylem. One of our hypotheses is that the ethylene biosynthesis pathway plays a role in this process. *Verticillium dahliae* can produce ethylene by three different pathways, including the 1-aminocyclopropane-1-carboxylic acid (ACC) pathway which is also used by plants. ACC, a precursor of ethylene, can be degraded by the fungal enzyme ACC deaminase (ACD) to α -ketobutyrate and ammonia. The pathogen can use these compounds as carbon and nitrogen source, respectively. Gene expression of ACD was found to be upregulated during an in vitro experiment in nutrient limiting conditions. To study the mechanism in planta, plant infection experiments were set up using the *V. dahliae* VD1-pepper (*Capsicum annuum* L.) pathosystem. In the presymptomatic stage, the fungal ACC pathway was activated in the roots, while key genes in the plant ACC pathway were expressed in the stem and ACC levels significantly increased in the hypocotyl. Fungal ACD expression was upregulated in the roots in the presymptomatic phase and in the stem in the symptomatic phase. To understand the molecular mechanism behind activation of plant ACC biosynthesis, gene expression of ten fungal ethylene-inducing enzymes was studied in vitro and in planta. Four of these genes, including a gene not present in several other strains of *V. dahliae*, were found to be expressed during infection either in roots or stem. Our data suggest that *V. dahliae* manipulates ACC production in the plant and converts ACC to α -ketobutyrate and ammonia under nutrient deficient conditions.



SE03-14th IVS 003

ENHANCING PLANT DEFENSE MECHANISMS: IMMUNIZATION EFFECTS OF AUTOCLAVED VERTICILLIUM DAHLIAE SPORES

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The soil-borne fungus *Verticillium dahliae* poses a significant threat to over 200 species of dicotyledonous plants, causing a severe vascular disease. In our study, we investigated the potential protective and immunization effects of autoclaved *V. dahliae* spores against this pathogen using *Arabidopsis* and eggplants as model organisms. Our findings demonstrated a notable enhancement in protection against *V. dahliae* when autoclaved spores were applied to eggplants and *Arabidopsis*. Compared to untreated controls, plants treated with autoclaved *V. dahliae* spores exhibited reduced disease severity and diminished pathogen colonization. Furthermore, our molecular analysis unveiled a significant upregulation of defense-related genes, *PR1* and *PDF1.2*, in *Arabidopsis* plants treated with autoclaved *V. dahliae* spores, highlighting the plant's activation of its defense mechanisms. Moreover, pathogenicity experiments conducted with the *Arabidopsis* mutant *cerk1*, deficient in chitin perception, revealed a notable loss of protection against *V. dahliae* when treated with autoclaved spores of the pathogen. This underscores the crucial role of the chitin receptor *CERK1* in the immunization of *Arabidopsis* against *V. dahliae* using autoclaved spores. In summary, our study sheds light on the immunization effects of inactive *V. dahliae* spores in enhancing plant defense mechanisms, while also elucidating key insights into the underlying molecular mechanisms involved in plant-pathogen interactions.

of *Vrp* in fungal propagules and plant tissue. Mutant strains obtained, were evaluated for growth and conidia production in a variety of media. Furthermore, pathogenic capability of the mutants was tested on the susceptible plant host eggplant and results correlated with quantification of fungal biomass at the crown level. The research findings shed light into the role and function of an F-box protein in *V. dahliae* pathogenic capability and virulence.

The research work was supported by the Hellenic Foundation for Research and Innovation (HFRI) and the General Secretariat for Research and Technology (GSRT), under the HFRI PhD Fellowship grant (GA. no. 1953).

SE03-14th IVS 005

THE VDSNF1 GENE REGULATES VIRULENCE AND METABOLOME OF THE WILT FUNGUS VERTICILLIUM DAHLIAE AFFECTING ITS PERCEPTION BY THE HOST PLANT

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The soil-borne fungus *Verticillium dahliae* causes economically important losses in numerous crops. No therapeutic measures are available, therefore it is important to evaluate potential targets for alternative management of the disease. In the present study, the role of the *VdSNF1* gene in virulence, physiology and the metabolomic content of *V. dahliae* was investigated. Pathogenicity assays on *Arabidopsis* plants showed that inactivation of the *VdSNF1* gene in three mutants drastically reduced symptoms. At 35 dpi, the wild-type and complemented strains showed increased biomass compared to $\Delta VdSNF1$ strains, which remained at low levels. Expression of the *PR1* gene, associated with the salicylic acid pathway, was higher at 10 dpi in plants infected with the 70 Δ SF4 strain that showed lowest biomass at 35 dpi, compared to the untreated control. In plants infected with all other strains, the *PR1* gene was underexpressed. Similarly, the gene *PDF1.2 β* , which is associated with the jasmonic acid pathway, was higher expressed at 10 dpi in plants infected by the mutant strain 70 Δ SF22. On the contrary, in eggplants, a 10 times induction of the *PR1* gene was observed at 5 dpi (days post inoculation) upon infection with the wild-type strain, while *PR1* was underexpressed in plants infected by mutant strains with the *vgb1* (G protein beta subunit) or the *VdSNF1* gene inactivated. Furthermore, transcription levels of a trehalose synthase (*VDAG_03989*) were significantly higher in spores of mutant strain 70 Δ SF20 at 24 h after exposure to simulated xylem medium (SXM) medium, compared to the wild type and the complemented strain derived from it. Metabolomic analysis showed reduced trehalose, glucitol and glucose production in the strain 70 Δ SF4 grown on solid SXM medium and increased synthesis of phosphoric acid, L-proline, L-isoleukine, L-Norleucine, and glycine. Finally, reduced mycelial growth at 33°C observed for mutant strains did not correlate with *VdSNF1* disruption.

SE03-14th IVS 004

EXAMINING THE FUNCTION OF AN F-BOX PROTEIN IN VERTICILLIUM DAHLIAE PATHOGENIC BEHAVIOR

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Verticillium dahliae is a soilborne ascomycete that causes wilt disease in a wide range of plant species, leading to severe agricultural losses. Unraveling the function of genes involved in the life cycle and pathogenicity of this phytopathogen, can lead to the identification of possible targets for disease control. In the present study, we investigated the role of an F-box protein coding gene from *V. dahliae*, henceforth mentioned as *Vrp*. A disruption construct for the gene was generated in appropriate plasmid vectors, where the *Vrp* sequence was interrupted by an antibiotic resistance cassette, while at the same time an expression construct under the control of the native promoter, was generated for future complementation studies. However, disruption attempts in *V. dahliae* isolates were unsuccessful. As an alternate step, gene overexpression was undertaken. More specifically, a *Vrp-EGFP* fusion under the control of a strong fungal promoter was introduced in different *V. dahliae* strains, in order to visualize localization

SE03-14th IVS 006

PLANT XYLEM METABOLITE AND STARCH DYNAMICS IN THE *VERTICILLIUM DAHLIAE* – PEPPER INTERACTION

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Despite xylem's nutrient scarcity, *Verticillium dahliae* thrives and flourishes in this environment, highlighting its impressive adaptability to nutrient-poor conditions. In this study, the *V. dahliae* VD1/pepper pathosystem was used as a model and we adopted a unique interdisciplinary approach combining biochemistry, ecophysiology, and microscopy to unveil novel insights into fungal behavior within the xylem. To study changes in plant xylem metabolites during *Verticillium* infection, gas chromatography-mass spectrometry (GC-MS) was carried out on xylem sap obtained from both healthy and inoculated plants. BIOLOG phenotypic arrays were used to investigate the capability of *V. dahliae* to use the identified xylem sap metabolites as carbon and nitrogen sources. Starch dynamics were investigated using microscopic analyses, which involved vibration microtome sectioning and lugol staining. GC-MS analysis of xylem sap from both healthy and infected pepper plants revealed nutrient enrichment, particularly in amino acids and sugars, at 14 days post inoculation (dpi). This time point is linked to fungal biomass transition from roots to shoot and stunted growth development. Starting at 21 dpi, a notable depletion of organic acids and sugars in both roots and shoots was observed, coupled with a strong increase in fungal biomass. By 28 dpi, healthy control plants showed a pronounced rise in sugars along with starch accumulation in ray parenchyma cells, while no starch accumulation was observed within infected plants. This observation, coupled with results from BIOLOG phenotype assays, suggests that *Verticillium* may use these carbon sources thereby disrupting sugar dynamics within the plant. As a result, insufficient resources can be allocated for plant growth, possibly explaining the manifestation of stunted growth. In conclusion, our findings suggest that *V. dahliae* manipulates the metabolic pathways in pepper to access nutrients, enabling its survival in the xylem. Further investigations are needed to delve into the underlying mechanisms of this metabolic manipulation.

SE03-14th IVS 007

SURVIVAL, PERSISTENCE AND INFECTION EFFICIENCY OF *VERTICILLIUM DAHLIAE* PASSED THROUGH THE DIGESTIVE SYSTEM OF SHEEP

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Verticillium wilt caused by *Verticillium dahliae*, affects a wide range of economically important plants. It survives in the soil as microsclerotia for more than 15 years. Once introduced into an area, the pathogens can be disseminated by several means. The present study was carried out to determine the survival, persistence and infection efficiency of *V. dahliae* passed through the digestive tract of sheep. Eggplant, turnip, tomato and pepper plants were artificially inoculated with 32 *V. dahliae* isolates. Thirty three days post inoculation the disease incidence and severity for eggplant, turnip, tomato and pepper plants were 99.6, 96.2, 62.9 and 18.0%, and 80.1, 49.8, 19.8 and 7.8%, respectively. The infected plant material was used to feed four, one-year old sheep. PCR assays revealed the presence of *V. dahliae* DNA in fecal samples received from animals' rectum on days 1, 2, 3, 4 and 5, whereas the pathogen DNA was not detected on 0, 6 and 7 days after feeding. Pathogenicity tests were conducted by transplanting eggplant plants into soil substrate amended with 20% decomposed manure, collected from the four animals fed with the infested forage. Fifty two days after transplanting, manure treated plants exhibited *Verticillium* wilt symptoms whereas two months later, disease incidence, disease severity and percentage of positive *V. dahliae* isolations from stem tissues were 58.3%, 30.7% and 48.3% respectively. Symptoms or positive isolations were not observed in control plants (transplanted in 100% soil substrate). This is the first report of the active role of *V. dahliae* passed through the digestive system of sheep, as effective inoculum for host plants, in relation to the span persistence and transmission via the sheep carrier. This knowledge is important for planning and implementing effective management practices against *V. dahliae*.

CONFERENCE I

08.30-11.00 14th IVS 4

Biological Control of Verticillium Wilt Pathogens

Chair: Ioannis Stringlis and Sotiris Tjamos (Agricultural University of Athens, Greece)

SE04-14th IVS 001

UNVEILING THE EFFICACY: BACILLUS VELEZENSIS K165 VOLATILE ORGANIC COMPOUNDS IN VERTICILLIUM WILT BIOCONTROL

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Verticillium dahliae is one of the most devastating soilborne pathogens globally, posing significant challenges due to the absence of effective chemical control methods and the scarcity of resistant plant varieties. Consequently, there is an urgent need for innovative disease management strategies. In this context leveraging the biocontrol capabilities of beneficial microbes such as *Bacillus velezensis* K165 emerges as a promising avenue. Here, we investigated the efficacy of volatile organic compounds (VOCs) produced by K165 against *V. dahliae* in *Arabidopsis thaliana*. Our findings revealed that *Arabidopsis* plants exposed to K165 VOCs exhibited reduced symptoms of *Verticillium* wilt compared to control plants. Further analysis of RNAseq data and pathogenicity experiments, utilizing selected *Arabidopsis* mutants, highlighted a positive interaction between K165 VOCs and the chitin perception mechanism in plants, as well as lignin accumulation. Gas chromatography-mass spectrometry analysis of the K165 VOCs profile identified acetoin as the major chemical compound emitted by K165. While acetoin did not directly inhibit the growth of *V. dahliae* *in vitro*, *Arabidopsis* plants exposed to acetoin displayed decreased severity of *Verticillium* wilt compared to controls. These findings suggest that K165 VOCs activate plant defense mechanisms, contributing to the biocontrol efficacy of K165 against *V. dahliae*. In summary, our study demonstrates the potential of utilizing K165 VOCs against *Verticillium* wilt. Understanding the mechanisms underlying the interaction between K165 VOCs and the plant defense system provides valuable insights for developing sustainable strategies to combat *V. dahliae* and enhance crop health.

The work was supported by the Hellenic Foundation for Research and Innovation (HFRI) under the 3rd Call for HFRI PhD Fellowships (Fellowship Number: 5352).

SE04-14th IVS 002

IMPACT OF FUSARIUM OXYSPORUM FO12 ON FE ACQUISITION AND SYSTEMIC RESISTANCE GENE EXPRESSION AGAINST VERTICILLIUM DAHLIAE

Sevillano-Caño J.¹, Córdoba-Galván C.¹, Sánchez-Rodríguez A.¹, García del Rosal M.¹, Agustí-Brisach C.¹

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The induced systemic resistance (ISR) represents a pivotal plant-defense mechanism activated by beneficial rhizospheric microorganisms which can also induce responses to nutrient stresses, such as iron deficiency (Fe). The activation of both responses (ISR and Fe deficiency responses) relies upon common regulators such as ethylene (ET) and nitric oxide (NO). This partial overlapping between ISR and Fe deficiency regulatory pathways allows that in Fe-deficient conditions ISR could be triggered by rhizospheric microorganisms. Thus, this work aimed (i) to evaluate the ability of the non-pathogenic *Fusarium oxysporum* FO12 to trigger Fe deficiency responses in the roots, and (ii) to induce plant systemic resistance mechanisms in the leaves. To this end, 30-day-old *Arabidopsis thaliana* Col-O plants were inoculated with FO12 at a concentration of 10^6 conidia ml^{-1} in a hydroponic setup, both with and without Fe supplementation. The plant reduction capacity of Fe^{3+} and the relative gene expression of Fe acquisition, ET, NO, and salicylic acid (SA) related genes in the roots and leaves were determined. In parallel, identical *Arabidopsis thaliana* Col-O plants were cultivated in sterile peat and inoculated by irrigation with FO12 and with *Verticillium dahliae* strain V180 (non-defoliating pathotype) at 10^6 conidia g^{-1} of soil to assess the ability of FO12 to control this vascular pathogen. Disease severity was assessed periodically, and the Relative Area Under Disease Progression Curve (RAUDPC) was determined. Our findings indicate that FO12 significantly upregulated the expression of Fe, ISR, and SAR-related genes under both Fe sufficiency and Fe deficiency conditions. Furthermore, FO12 effectively reduced disease severity, as evidenced by lower RAUDPC values compared to control plants (only water). These results underscore the potential of FO12 to elicit iron-deficiency responses and plant systemic resistance mechanisms, positioning it as a promising candidate for use as both an iron biofertilizer and a plant-resistance inducer.

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SE04-14th IVS 003

BIOLOGICAL CONTROL OF HIGHLY INVASIVE AILANTHUS ALTISSIMA USING VERTICILLIUM NONALFALFAE ISOLATE VERT56

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The invasion of Tree-of-Heaven (*Ailanthus altissima*), which is native to Southeast Asia, has been documented on all continents except Antarctica and causes biodiversity loss and degradation of ecosystem function. Tree-of-Heaven usually spreads from urban heat islands into rural areas and is expected to expand its range due to climate change. In search for effective control agents, *Verticillium nonalfalfae* has shown promise as a biocontrol agent due to its distinct adaption to *Ailanthus*. The isolate Vert56, originating from Styria, Austria, induced severe wilting symptoms on inoculated *Ailanthus*-trees within a short period of time, without development of root sprouts or sucker shoots. Another advantage of this method is the systemic spread of the fungus within clonal root systems leading to dieback of neighboring *Ailanthus* trees. However, the use of *V. nonalfalfae* as a control agent must also consider potentially occurring non-target effects, as the genus *Verticillium* also includes well-known plant pathogens, particularly in agriculture, that pose significant threats to infected plants. Therefore, comprehensive risk assessment studies were carried out to determine the feasibility and safety of this approach: until now, about 70 species have been tested for susceptibility to isolate Vert56. So far, all tested ligneous plants proved to be tolerant or resistant to the pathogen, whereas only three herbaceous plants, i.e. *Petunia x hybrida*, *Cucumis sativus* and *Spinacia oleracea*, which are known as susceptible species, displayed symptoms of wilting after artificial inoculation. Based on the current results, an emergency authorization with limited indications regarding application period and area has been granted in Austria since 2017. Further investigations of non-target effects, the pathogen's persistence in the soil and in plant debris and on potential pathways of transmission are currently ongoing with the goal to achieve an "active substance authorization" for *V. nonalfalfae* isolate Vert56 within the European Union.

SE04-14th IVS 004

THE TALE OF DISCOVERING A BIOCONTROL AGENT AGAINST VERTICILLIUM WILT

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In our endeavour to explore the potential of soil application of microbiological growth media, such as nutrient broth and potato dextrose, in safeguarding eggplants against *Verticillium dahliae* by modulating the soil microbiome, we isolated and characterized the strain *Pseudomonas putida* Z13. Through *in vitro* experiments, we discovered that Z13 exhibits inhibitory effects on the growth of *V. dahliae*, as well as other pathogens like *Botrytis cinerea*. Notably, Z13 demonstrates the production of siderophores and chitinase, which are pivotal biocontrol traits. Moreover, genomic analysis of Z13 revealed the presence of genes responsible for inducing systemic resistance, including those encoding pyoverdine and

pyrroloquinoline quinone. In tandem with genome mining and *in vitro* tests, untargeted metabolomic analysis revealed that Z13 synthesizes a diverse array of antibiotic compounds, antioxidants, and plant defense signalling compounds. Subsequent *in planta* trials demonstrated the efficacy of Z13 in protecting both eggplants and tomato plants against *V. dahliae*. Utilizing a GFP-transformed Z13 strain, we observed efficient colonization of the rhizosphere, corroborating with the swarming and swimming activities of Z13. Furthermore, RNAseq analysis unveiled the ability of Z13 to stimulate the defense responses in tomato plants against *V. dahliae*. Overall, these findings suggest that the application of Z13, holds promise as a sustainable and effective strategy for managing *V. dahliae* and other plant pathogens in agricultural systems.

SE04-14th IVS 005

ASSESSMENT OF ROCKET EXTRACT AND RHIZOSPHERIC MICROORGANISMS FOR MANAGING VERTICILLIUM WILT IN EGGPLANTS: INSIGHTS INTO SYSTEMIC RESISTANCE AND BIOSYNTHETIC PROFILING

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This study aimed to assess the effectiveness of rocket (*Eruca sativa*) extract in alleviating *Verticillium* wilt in eggplants, investigate the potential of rhizospheric microorganisms for controlling the disease, and evaluate selected strains for their ability to induce systemic resistance while analyzing their genomic and biosynthetic profiles. Application of rocket extract significantly mitigated *Verticillium* wilt symptoms in eggplants compared to controls. Microorganisms isolated from rocket treated soil, including *Paraburkholderia oxyphila* EP1, *Pseudomonas citronellolis* EP2, *Paraburkholderia eburnea* EP3, and *Paraburkholderia oxyphila* EP4 and EP5, demonstrated efficacy against *Verticillium dahliae*, resulting in a reduction of disease severity and incidence in planta. Notably, strains EP3 and EP4 induced systemic resistance in eggplants against *V. dahliae*. Genomic analysis revealed shared biosynthetic gene clusters, such as ranthipeptide and non-ribosomal peptide synthetase-metallophore types, among the isolated strains. Metabolomic profiling of EP2 revealed the production of metabolites associated with amino acid metabolism, putative antibiotics, and phytohormones. Overall, the application of rocket extract significantly reduced *Verticillium* wilt symptoms in eggplants, while the isolated microorganisms exhibited efficacy against *V. dahliae*, inducing systemic resistance and sharing biosynthetic gene clusters. Metabolomic profiling highlighted potential disease-suppressing metabolites. This research contributes to environmentally friendly disease management in agriculture by demonstrating the potential of plant extracts and rhizospheric microorganisms such as *Paraburkholderia* sp. and *Pseudomonas citronellolis*.

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P001

COLLETOTRICHUM EPIDEMIOLOGY IN OLIVE ORCHARDS FROM THE REGION OF PREVEZA, GREECE AND EFFECT OF FUNGICIDE APPLICATION ON PATHOGEN SURVIVAL

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Anthrax disease has become pervasive in olive orchards globally, causing enormous decreases in yield. To evaluate effective control measures it is important to assess survival of the pathogen, in plant tissues. In the present study, an epidemiological survey was conducted during the period 2023 -2022, in two olive orchards (PS and PP) situated in the region of Preveza in Greece, with and without plant protection applied, respectively. The survey included detection of *Colletotrichum* spp. by isolation in different olive tree tissue during different vegetative stages of the crop and after specific control measures were applied. In orchard PS, despite regular plant protection applications, anthracnose infestation was noted in about 15-10 out of 250 trees in a point (BAD-PS) with favorable conditions for the disease. In orchard PP, anthracnose symptoms were observed in all 23 trees, along with high incidence of *Bactrocera olea* infestation. During 2023 a plant protection scheme against anthracnose was undertaken, including spraying mixtures of strobilurins and triazoles at blossom, triazoles at immature fruit and cooper fungicides early in Spring (March) or in Autumn after hail. Pyrethroids were applied against *Bactrocera olea*. Isolation of *Colletotrichum* spp. from flowers (May) in field PS indicated reduced isolation frequency in trees (E5-1) with Bordeaux mixture application in March, compared to trees with good aeration (A-GOOD_PS). Similar results were observed for isolation of *Colletotrichum* spp. from leave petioles. Interestingly reduced frequency was retained also in immature fruits sampled in June and end of August 2023. In field PP, initial plant protection applied was not able to reduce isolation frequency of *Colletotrichum* spp. from flowers compared to untreated control. Quantification of the pathogen biomass in olive flowers, fruit, leaf petioles and stems is underway. Correlation of results with local climatic conditions will lead to the development of an integrated protocol for disease management.

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P002

THE LANDSCAPE AS AN ESSENTIAL FRAMEWORK FOR A MORE EFFECTIVE IPM: TWO EXAMPLES IN FIELD AND PROTECTED CROPS

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Since Stern and colleagues (University California) wrote a seminal article 60 years ago for establishing the concept of integrated control, we have successively understood the complexity of the biotic relationships in agroecosystems

and the influence which the abiotic conditions have on them. Methods. Analyses of biotic and abiotic relationships have increasingly considered the multiple embedded ecological and managerial scales at which processes take place from the field to the landscape. The study of mechanisms involved in such processes include a bottom-up approach, from the molecular to the agroecological levels. This presentation underlines the importance of the composition and configuration of the landscape where agroecosystems are inserted. *Results.* To illustrate such a statement, two examples dealing with IPM in field crops and in protected crops are provided. The first example summarizes the results had in the study carried out to detect the most relevant landscape features influencing the abundance of phytophagous and predatory insects on maize and the incidence of insect-borne viruses. In a second example, we describe how the landscape surrounding greenhouses in the Mediterranean provides resources that allow us to take advantage of the early season entry of native predatory mirids in greenhouses. However, the influence of landscape on the pest and natural enemy population dynamics in greenhouses is difficult to be determined due to the variability of arthropod exchanges between inside and outside greenhouses according to several features of greenhouse structure, ventilation regime and window screening. The conclusions from the two studies can help to understand the complexity of biotic interactions in agroecosystems, how to manage these interactions and prevent us from using simple rules in IPM.

P003

THE EFFECT OF CHITOSAN ON THE INFECTION AND DISEASE OF ZUCCHINI PLANTS CAUSED BY CUCUMBER MOSAIC VIRUS

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Chitosan is a naturally-occurring polysaccharide with a potential to reduce the impact of plant pathogens. The aim of this study was to record the short- and long-term effect(s) of chitosan against the infection (laboratory and natural) of zucchini by cucumber mosaic virus (CMV), under field conditions. A medium molecular weight chitosan (TCI America) was applied (1 % solution in 1 % acetic acid) to zucchini plants of a local Greek variety (Kompokolokytho) at the first true leaf stage. Plants treated with only acetic acid or non-treated ones were used as controls. One day later, half the plants were mechanically inoculated with a Greek isolate

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of CMV. Ten days after treatment (end of June 2023), plants were tested by ELISA to confirm infection and subsequently infected and healthy plants (180 in total) were used to establish a field experiment at Ypato (Viotia prefecture) with three replications of 10 plants per treatment. For the next two months, symptoms, plant growth (plant size, flower, and fruit number) and yield (harvested fruits), as well as CMV spread in healthy plants were recorded on a weekly basis. The applied concentration of chitosan did not affect the percentage of plants that were infected by CMV either by mechanical inoculation in the laboratory or by natural infection in the field. The disease impact as recorded by the size of the plants (diameter) and the number of flowers, was not significantly affected; only slight differences were recorded between the naturally infected chitosan-treated plants and the non-treated ones. The analysis of the other parameters is in process. The applied chitosan scheme was not efficient to reinforce the selected zucchini variety against CMV infection. Increased concentration perhaps in combination with different functional additive(s) may increase the efficacy of the selected chitosan product.

P004

SOIL SOLARIZATION EFFICIENTLY REDUCES FUNGAL SOILBORNE PATHOGENS' POPULATION, PROMOTES LETTUCE PLANT GROWTH AND AFFECTS THE SOIL BACTERIAL COMMUNITY

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The intensive nature of lettuce cultivation consisting in successive crop cycles within the same year and the lack of adequate crop rotation practices promote the development of high pathogen populations, affecting crop production and making necessary the implementation of other effective control measures which will conserve at the same time soil health. In this study, the impact of various soil disinfestation methods such as solarization, chemical disinfestation and application of a biofungicide were evaluated in a commercial field which has been repeatedly used for lettuce cultivation. The populations of the soilborne pathogens *Rhizoctonia solani*, *Pythium ultimum*, *Fusarium oxysporum* and *Fusarium equiseti* were measured via RT-qPCR before and after the implementation of the specific disinfestation methods. Although all the tested methods significantly reduced the population of the four soilborne pathogens, soil solarization was the most effective. In addition, solarization reduced the number of lettuce plants affected by the pathogens *R. solani* and *F. equiseti*, influencing significantly at the same time the agronomic parameters of lettuce plants, in terms of fresh plant weight. Amplicon sequence analysis of 16S rRNA encoding genes used to study the soil bacterial community structure showed that Firmicutes, Proteobacteria, and Actinobacteria were the predominant bacterial phyla in soil samples. In general, solarization had positive effects on Firmicutes and negative effects on Proteobacteria and Actinobacteria. Soil fumigation with dazomet increased the

relative abundance of Firmicutes and Proteobacteria, and reduced the corresponding values of Actinobacteria, while the biofungicide had no significant effects on the three predominant bacterial phyla. The results of this study are expected to contribute towards the implementation of the most effective control method against the most common soilborne pathogens in intensively cultivated fields, such as those cultivated with leafy vegetables.

P005

DEEP LEARNING-BASED DIAGNOSIS OF LEAF WITH PEST DAMAGE DURING VEGETABLE SEEDLING PRODUCTION

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The introduction of deep learning-based diagnostic techniques is effective for the rapid pest detection to help high quality vegetable seedling production. Here, we developed the diagnostic algorithm of leaves with pest damage lesions from vegetable seedlings. First, the images of target seedlings (watermelon and cucumber) with pest damage by *Tetranychus urticae* and *Thrips palmi* were obtained by using automated image collection device. To make training dataset, each leaf was marked with polygon by using 'Labelme' program. Codes were created to indicate the host plant, damaged part, and target pest. The number of training dataset was obtained as 2,056 and 3,251 in watermelon and cucumber leaves, respectively. The leaf classification was developed with instant segmentation algorithm, 'YOLOv8', and its performance was evaluated with mean average precision (mAP50). The mAP50 values were 80 % and 79 % in watermelon and cucumber, respectively. The diagnosis of the leaf's abnormality by the pests were conducted with image classification algorithm, 'ResNet50'. The accuracy values were 75 % and 69 % in watermelon and cucumber, respectively. Future research on more data acquisition and elaborating the classification algorithms might contribute to develop pest diagnosis system in vegetable seedling production.

P006

EVALUATION OF LINALOOL AND EUGENOL AGAINST TUTA ABSOLUTA AND OTHER PESTS

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Our study aimed to explore the efficacy of linalool and eugenol, main components of essential oils (EOs), against *Tuta absoluta* (Lepidoptera: Gelechiidae), *Aphis gossypii* (Hemiptera: Aphididae) and *Tetranychus urticae* (Acari: Tetranychidae) as well as their side-effects on the insect predators *Macrolophus pygmaeus* and *Nesidiocoris tenuis* (Hemiptera: Miridae).

T. absoluta eggs, *A. gossypii* nymphs and *T. urticae* adults were sprayed before released in a dish with a treated leaf of tomato, cucumber or bean, respectively. The same procedure was followed for *M. pygmaeus* and *N. tenuis* nymphs. Mortality

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rates were recorded 24h after treatment.

Our study showed that among the three pests, linalool caused a high mortality rate (70 %) against *T. urticae* adults when used at a concentration of 0.1 %. However, mortality was lower in case of *T. absoluta* and *A. gossypii* (48 % and 28 %, respectively). Eugenol caused a high mortality rate against *T. urticae* adults (74 %). It is noteworthy to mention that eugenol caused high mortality against *T. absoluta* eggs (62 %) and 46 % against *A. gossypii* nymphs. The application of linalool or eugenol caused no mortality on *M. pygmaeus* and *N. tenuis* nymphs.

Linalool and eugenol showed a high potential in the control of *T. absoluta* and *T. urticae* without harmful effects on their natural enemies. These results are promising and should be further investigated on tomato plants and under field conditions.

P007

EFFECT OF MICROORGANISMS AND RESISTANCE INDUCERS AGAINST RACE 1 OF FUSARIUM OXYSPORUM F. SP. LACTUCAE IN A CLOSED SOILLESS SYSTEM

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Different strains of experimental non-pathogenic-*Fusarium oxysporum* and *Fusarium solani*, commercial biocontrol agents (*Streptomyces griseoviridis*, *Bacillus subtilis* and *Beauveria bassiana*), as well as products based on calcium oxide and potassium phosphite were tested in a closed soilless system against race 1 of *Fusarium oxysporum* f. sp. lactucae, the causal agent of Fusarium wilt of lettuce. The treatments were applied to lettuce seedlings, grown for two days in a peat medium before being artificially inoculated with the pathogen, and were repeated 5 times at -7day-intervals. The experimental non-pathogenic *F. oxysporum* MSA35 and FC3 strains provided a higher disease severity reduction (83-54 % efficacy) than the commercial *B. subtilis* (31-29 % efficacy), *S. griseoviridis* (22 to 52 % efficacy) and *B. bassiana* (39 to 63 % efficacy) formulations. The highest fresh weight was observed in the case of treatments with non-pathogenic *F. oxysporum* MSA35 and FC3 strains, thereby generally reflecting the observed disease reduction. Calcium oxide and potassium phosphite generally resulted in disease protection, but a great variability in efficacy was observed. The obtained results showed good possibilities for the non-pathogenic *Fusarium oxysporum* strains against the pathogen in soilless systems.

P008

SEQUENTIAL APPLICATION OF DIMPROPYRIDAZ (AXALION®) AND BEAUVERIA BASSIANA PPRI 5339 (VELIFER®) FOR EFFECTIVE CONTROL OF BEMISIA TABACI IN TOMATO CROPS

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The silverleaf whitefly *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) is a major agricultural insect pest that causes substantial economic losses by affecting a wide range of crops globally. The control of *Bemisia tabaci* poses challenges due to its rapid reproduction, its ability to develop resistance to insecticides, and its capacity to transmit plant viruses. The decline in the registration of new insecticide active ingredients over time in the European Union highlights the urgent need for innovation toward integrated pest management (IPM) methods. One such method involves combining conventional insecticides with biological control agents in spray programs. BASF has discovered and developed the active ingredient dimpropridaz (Axalion®), a pyridazine pyrazolecarboxamide insecticide with a novel mode of action (IRAC MoA group 36) for the control of piercing and sucking insect pests, including whiteflies (Aleyrodidae). Additionally, BASF has developed the entomopathogenic fungus *Beauveria bassiana* PPRI 5339 (Velifer®), a naturally occurring (soil born) parasite of various arthropod species including whiteflies (Aleyrodidae). This study investigated the efficacy of a sequential application of these two novel insecticides on a fresh tomato crop variety (*Solanum lycopersicum*) grown under greenhouse conditions as an alternative strategy to a conventional spray program used in the southeast of Spain (Almeria) to control *Bemisia tabaci* Q and B biotypes. The results demonstrate that applying Axalion® followed by Velifer® in a -7day interval spraying sequence is an effective strategy to prevent the post-knockdown recovery of the pest, resulting in a similar level of control as the conventional spray program. This strategy has the potential to support farmers in implementing IPM programs for controlling silverleaf whitefly by providing flexibility in managing residue levels while protecting against the development of resistances.

P009

ESSENTIAL OILS ON THE CONTROL OF SWISS CHARD AND SPINACH LEAF MINING FLIES

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The organic production of Swiss chard and spinach is hindered by the lack of effective and sustainable control means able to reduce the impacts caused by leaf mining flies, such as the widely known *Pegomya betae* Curtis (Anthomyiidae). In this study, we tested the efficacy of essential oils (EOs) in controlling leaf mining flies on Swiss chard (*Beta vulgaris* L.) and spinach (*Spinacia oleracea* L.) under field conditions.

The test was organized in a randomized complete block design, with three replicates. EOs used in the tests were obtained from thyme (*Thymus vulgaris*), rosemary (*Rosmarinus officinalis*), and "Grosso" lavender (*Lavandula x intermedia* "Grosso"). Water was used as control. Each EO was used at 0.05 % concentration. Foliar sprays from stages BBCH 13-19 to the harvest (BBCH 46-49) were applied weekly. The severity of infestation (number of infested leaves over the total amount

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of leaves) was monitored weekly.

Preliminary results showed that the severity of infestation, compared to the control, was significantly lower for rosemary EO on Swiss chard and for lavender and rosemary EOs treatments on spinach.

This is the first open field trial evaluating the effectiveness of EOs for the control of Swiss chard and spinach leaf mining flies. The use of EOs might be considered an alternative to chemical pesticides because of their low toxicity on non-target organisms and the low environmental persistence.

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P010

MAKING A PREDATOR BETTER SUITED TO AGROSYSTEMS: PESTICIDE RESISTANCE OF *NESIDIOCORIS TENUIS* REUTER (HETEROPTERA: MIRIDAE)

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Integrated pest management often combines the application of biological control with conventional methods such as chemical pesticides. Predators for biological control of pests, commercially available and with higher pesticide resistance can be better suited to agrosystem practices. The aim of this study is to investigate the susceptibility of wild populations of the predator *Nesidiocoris tenuis* Reuter (Heteroptera: Miridae) to pesticides used against whiteflies and the tomato borer, common prey of *N. tenuis* and also increase their resistance via artificial selection.

The contact toxicity of three pesticide active ingredients, abamectin, spinosad, spiromesifen and flupyradifuron, was investigated on adults and immatures (nymphs N5/N6) of four wild populations and one commercial. Furthermore, the mirids were artificially selected in order to increase their pesticide resistance to abamectin and spiromesifen.

We found that there are wild populations of *N. tenuis* more resistant than the commercial population to both abamectin and flupyradifuron, during the adult stage and the nymphal stage, as well. This finding suggests that natural populations of this biocontrol agent have standing genetic variation for pesticide resistance, therefore they can be artificially selected to make populations more homogenous for the resistant phenotype and achieve a higher resistance ratio.

Resistance to pesticides may have appeared in wild populations of this predator due to the long-term use of these active ingredients in agriculture. Resistant biocontrol agents can be combined with chemical control in IPM strategies.

The work was funded by the flagship project "Innovative plant protection for primary agricultural production-InnoPP" and the Horizon EU project "NextGenBioPest"

P011

AMICROBIOME STUDY ON HEALTHY AND PHYTOPHTHORA CRYPTOGEA-INOCULATED LETTUCE

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In recent years, root rot caused by the soil-borne oomycete *Phytophthora cryptogea* has emerged in hydroponic lettuce cultivation in Flanders. The disease is an increasing problem for lettuce growers as up to 85 % of the hydroponic lettuce growers are facing economic losses up to €50 000 per ha per year. *Phytophthora cryptogea* produces flagellated, asexual zoospores which can easily spread the disease in the recirculation water. After infection, lettuce roots slime and show rotting symptoms, resulting in disappearance of the roots and eventually death of the lettuce crop. Although the disease has also been observed in lettuce cultivation in several other countries, research on this pathosystem is very limited. Here, we investigated the rhizosphere of healthy lettuce plants and lettuce plants inoculated with *P. cryptogea* at different time points, for different lettuce varieties and for cultivars with a different tolerance level against *P. cryptogea*. We found that the rhizosphere of the lettuce plants was dominated by Pseudomonadota, Planctomycetota, Cyanobacteriota, Bacteroidota and Actinomycetota, irrespective of time, presence of the pathogen, lettuce variety and tolerance level. However, perMANOVA analysis revealed that each of the four parameters (time, pathogen, variety, tolerant/susceptible) has a significant effect on the microbial community, and especially time and the presence of the pathogen were found as the two main drivers influencing microbial communities. Generally, we found a higher alpha-diversity (zOTU richness) for inoculated plants compared to control plants. Especially, the richness decreases over time for the control plants, while it remains the same or increases over time for the inoculated plants.

These results confirm an earlier performed study on naturally infested lettuce plants, where we found significant differences in microbial communities between non-symptomatic plants and plants showing symptoms of *P. cryptogea*, and a higher richness for these symptomatic plants.

P012

NEW METHODS FOR EVALUATING E-PROBE DETECTION PERFORMANCE IN MICROBIOME DATA: A CASE STUDY OF *NITROBACTER HAMBURGENSIS* IN WINTER WHEAT SOIL

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Winter wheat is one of the most important cash crops in Oklahoma, with incomes ranging from 400\$ million to 1\$ billion yearly. Previous studies have reported that soilborne microbiomes associated with winter wheat enhance plant growth and pathogen suppression. Greenhouse essays towards microbiome identification suggest *Nitrobacter hamburgensis* acts as an enhancer for winter wheat production, and data collected from field trials has reinforced this finding. Microbe Finder: MiFi is a fast and reliable organism detection tool in

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high-throughput sequencing (HTS) data using e-probes for specific identification of organisms. E-probe performance for the identification of *Nitrobacter hamburgensis* was evaluated through the Receiver Operating Characteristic (ROC) Curve and Youden-Index estimation. Simulated HTS samples were generated with varying read abundances mimicking 20 serial dilutions with 100 replicates each to obtain the true and false positive rates for each dilution. Detection was carried out using five sets of e-probes with different lengths and thresholds. Youden Indexes (J) were identified for each dilution, e-probe size, and detection threshold, and the optimal Index was selected for the minimum number of reads detected. ROC curves paired with the Youden Index display that detection thresholds, reads present in the sample, and e-probe lengths have an influence on detection performance. Shorter e-probes perform better in low abundance samples, while detection with long e-probes is more robust in high abundance scenarios. If more sensitive detection is required, then e-probes with the highest J index and lowest target read number should be selected. This integrative analysis allows an approach to e-probe assessment for organism identification considering sensitivity and specificity in a single index value.

P013

MICROBIAL INDUCTION OF PLANT RESILIENCE TO DROUGHT STRESS (MICRORES)

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In the past decade, yields of major food crops worldwide have decreased due to drought. Over the past years, it has become evident that microorganisms associated with plants can enhance drought tolerance, allowing sustainable crop growth under abiotic stress conditions. In this study a total of 146 bacterial isolates belonging to the main 4 phyla, Proteobacteria (110), Bacteroidota (18), Actinobacteria (11) and Firmicutes (5) isolated from the island of Terschelling (The Netherlands), were tested for their potential to alleviate drought stress in *Arabidopsis thaliana*. Among the bacteria exhibiting drought tolerance enhancement, *Flavobacterium* sp. 98 showed the most promising activity. To decipher the chemical and genomic basis of this phenotype, we employed comparative genomics, targeted mutagenesis and metabolomics. The genomic analysis of the *Flavobacterium* spp. in the collection using our in-house comparative genomics tool (bacLIFE), revealed a correlation between drought stress alleviation and the presence of the thiamine thiazole synthase gene. Interestingly, foliar applications of thiamine has been widely described to alleviate a broad-spectrum of stresses, including drought. A site directed mutagenesis to knock-out the thiamine thiazole synthase gene has been carried out.

Currently, in vivo and in vitro experiments are ongoing to elucidate the role of this gene in the alleviation of drought stress.

P014

DIFFERENTIAL RESPONSES OF TOMATO MONEYMAKER AND WILD RELATIVES TO THE SOILBORNE PATHOGEN *VERTICILLIUM DAHLIAE*

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The devastating soilborne phytopathogenic fungus *Verticillium dahliae* (Vd) causes vascular wilt disease to many economically important crops, but it is hard to control in a sustainable manner. Harnessing rhizosphere microbiomes holds promise for enhancing crop resilience to Vd. Additionally, exploring the potential of crop wild relatives remains an extra resource to sustainably combat this pathogen. Here we study wild tomato relatives, known to be resistant to Vd, and compare their growth and disease development to that of a cultivated tomato cultivar upon infection by Vd. Initially, soil from the experimental field of Agricultural University of Athens was collected and analyzed for its physicochemical properties and its microbial load. Our analyses showed that it is a clay loam texture soil, rich in plant nutrients and with considerably higher microbial diversity than the potting soil commonly used in greenhouse experiments. Next, in our experiments, field soil was used in mix with sterile potting soil. Cultivated tomato *S. lycopersicum* cv. Moneymaker and two wild tomato relatives (*S. pimpinellifolium* and *S. lycopersicum* x *S. cheesmanii*) were used and infected by Vd. race 1 isolate 7OV. Growth parameters of tomato plants were measured and disease development of Moneymaker and the wild relatives to Vd were evaluated. Moneymaker plants grew taller in sterile potting soil than in the mix with field soil, but showed higher disease severity. That indicates the effect of soil type and microbiome on growth and disease outputs, respectively. The phenotype of wild relatives, grown in sterile potting soil, showed that both responded different compared to Moneymaker. They also responded different compared to Moneymaker in terms of growth and disease development. These preliminary results form the basis to further study plant traits and the microbiome of wild tomato relatives that contribute to their enhanced resistance to Vd.

P015

EVALUATING GENERAL SUPPRESSIVENESS OF BIOCHAR WITH ORGANIC AMENDMENTS IN CONJUNCTION WITH SOIL MICROBES AGAINST BACTERIAL WILT OF TOMATO

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Manipulation of soil microbial composition through organic amendments represents an important strategy for enhancing soil suppressiveness. Previous research has demonstrated the potential to induce general suppressiveness of biochar

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through organic amendments in conjunction with soil microbes. The objective of this study was to thoroughly evaluate the general suppressiveness of biochar against tomato bacterial wilt caused by the *Ralstonia solanacearum* species complex (RSSC).

Rice husk biochar was subjected to organic amendment using multiple parallel mineralization methods to immobilize microbial populations on the substrate. Tomato cultivation in biochar was followed by an inoculation test with 5×10^7 cfu/g RSSC. In addition, 16S rRNA metagenomics was performed using an Illumina MiSeq platform.

Biochar with immobilized microbes (BIM) exhibited significant disease suppression against bacterial wilt of tomato, in contrast to those treated with chemical fertilizer or autoclaved as a control. Evaluation of RSSC survival in BIM using RSSC selective media showed a decrease in RSSC density in BIM, while an increase was observed in biochar treated with chemical fertilizer. This underscores that the disease suppressive properties of BIM are primarily due to microbial activity rather than inherent biochar properties. Metagenomic analysis of the BIM identified Rhizobiales, Micrococcales, Burkholderiales, and Xanthomonadales as predominant, suggesting that suppression of RSSC in BIM was facilitated by established microbial communities within the biochar.

This study successfully introduced microorganisms into biochar to create a substrate conducive to plant growth. Notably, disease suppression was observed in BIM, which was largely attributed to microbial activity. This manipulation of biochar with microbial population holds great promise for its application in agriculture and provides a potential avenue for the development of high-performance biochar tailored for agricultural purposes.

P016

REDUCTION OF WATERCORE OCCURRENCE IN APPLE BY APPLICATION OF CALCIUM AND BORON COMPOUNDS

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The apple watercore is a physiological disorder that leads to serious economic losses by reducing storability and quality. The main objective of this study was to develop a chemical spray formulation to mitigate this disorder. Experiments were conducted at an apple farm in South Korea over three years (2019, 2020, and 2021). The spray chemical formulations were prepared using pure calcium, boron, and calcium plus boron ($\text{CaO} + \text{B}_2\text{O}_3$). Foliar spray treatments were applied five times from May and fruits ripening period, with intervals of either 10 or 15 days (periodic treatment) and twice during month of May- June and July- August (monthly treatment). The effect of spray program on reducing apple watercore was investigated over the three-year experimental period. Additionally, the weight, firmness, sugar content, acidity, and color (Hunter L, a, and b values) of the fruits were estimated. Based on the fruiting location, the incidence of watercore symptoms was found to be higher in fruits harvested from the upper parts of the tree compared to those harvested from the lower parts. In comparison with the control (non-treatment),

the watercore symptom was reduced by 66 %, 2010 89% in 2019, and 69 % in 2020. There was a significant effect of monthly spraying treatments on reducing apple watercore compared to control (no-treatment), while there was no difference in the effect of periodic treatments. Furthermore, the study found no statistically significant difference in weight, hardness, sugar content, acidity and color (Hunter L, a, and b values) between treated and untreated apple fruits.

P017

STRATEGIES FOR SELECTING POTENTIALLY EFFECTIVE BIOFUMIGANT SPECIES FOR OPTIMAL BIOFUMIGATION OUTCOMES

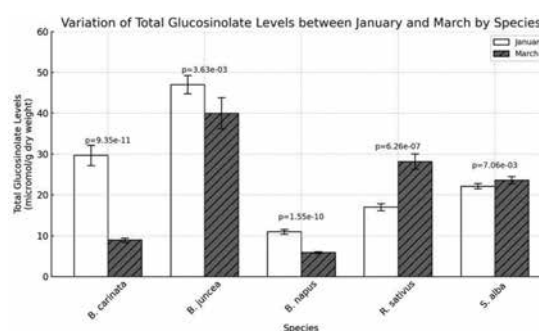
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Biofumigation is a key strategy for controlling soil-borne diseases in horticulture. This practice includes growing biofumigant crops and incorporating their chopped tissues into the soil. This study focuses on selecting biofumigant species suited for outdoor cultivation in continental climates, particularly examining five fast-growing Brassicaceae species: *Brassica napus*, *B. juncea*, *B. carinata*, *Raphanus sativus*, and *Sinapis alba*. These species are recognized for their biofumigant potential and isothiocyanate production.

The field trial, conducted on 24 plots using a randomized block design (with fallow as control), assessed the agronomic behavior of these species. Soil preparation involved initial physical-chemical analysis and fertilization. Growth was tracked using near (SPAD, portable reflectometer) and remote sensors (drones with multispectral cameras), focusing on vegetative development indicators and combined chlorophyll-vegetative indices (TCARI/OSAVI). Leaf or root samples were collected for glucosinolate analysis, following modified ISO Norm (1992) protocols. These samples underwent freezing, lyophilization, and grinding for glucosinolate extraction. The glucosinolates were purified using DEAE Sephadex ion-exchange resin and enzymatically desulfated. High-Performance Liquid Chromatography (HPLC) analyzed the desulfoglucosinolates with specific solvent gradients and temperature controls. Quantification utilized response factors from ISO Norm (1992) and other sources for accuracy. This analysis aimed to correlate glucosinolate levels and biomass with remote sensing data (TCARI/OSAVI), chlorophyll content (SPAD), and various growth indices.

The study revealed significant inter-species variability in glucosinolate composition and concentration.



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These variations were evident between different analysis dates, underscoring the importance of understanding species-specific glucosinolate profiles. The research examines crucial indicators for evaluating the agronomic viability of potential biofumigant species, aiming to identify the most effective species for intercropping or rotation in soil-borne disease management. This comprehensive methodology, leveraging advanced monitoring techniques, aims to optimize biofumigation outcomes in horticulture. The research was funded by the Plan Estatal de Investigación Científica, Técnica y de Innovación 2023–2021 (PID125545-2021OR-C22).

P018

SPECTRAL ANALYSIS FOR CEREAL LEAF BEETLE DETECTION IN WINTER WHEAT: TOWARDS PRECISION PEST MANAGEMENT

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The cereal leaf beetle (CLB) poses a significant threat to winter wheat production, necessitating advanced detection methods for effective pest control. This study utilized a full-range spectroradiometer to collect spectral signatures from CLB-affected flag leaves, spanning 2150 wavelengths across visible (VIS), near-infrared (NIR), and short-wave infrared (SWIR) spectra. Damage was categorized into four classes: no damage, slight damage by adult CLB, moderate damage by CLB larvae, and severe damage by CLB larvae. Analysis of the spectral signatures revealed a recognizable pattern: higher damage level corresponded to increased reflectance in the VIS region of the spectra, while the opposite trend was observed in the NIR and SWIR. Vegetation indices were calculated from the collected spectra as spectral indicators for stress parameters in the crops. Their correlations with the damage levels were examined and interpreted using correlation heat maps. In particular, indices based on VIS (mainly green and red) and NIR spectra showed the highest correlations with CLB damage classes. Key indices included R_{green}, NDVI750, GNDVI, SIPI and NDVI, all of which are commonly used on remote sensing platforms such as satellites and multispectral drones. In addition, we applied dimensionality reduction techniques to spectral data, including Principal Component Analysis (PCA), Uniform Manifold Approximation and Projection (UMAP) and t-distributed Stochastic Neighbor Embedding (t-SNE). These techniques revealed the potential for further damage classification and enabled more precise pest management strategies. By harnessing these findings, we propose a site-specific approach to pest management for mitigation of environmental impact of pesticide overuse. This approach enables targeted interventions by focusing pesticide application on hotspots with infested plants, thus optimizing resource use and reducing agricultural losses. This study highlights the significance of spectral analysis in developing precise pest management strategies. Further research could explore the integration of remote sensing technologies into real-time monitoring systems for proactive pest control.

P019

BARBERRY DECREASE EMERGENCE OF NEW RACES AND POPULATION GENETIC DIVERSITY OF THE WHEAT STRIPE RUST PATHOGEN AFTER FUNGICIDE TREATMENT

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Wheat stripe rust, caused by the heteroecious rust fungus *Puccinia striiformis* f. sp. *tritici*, is a destructive disease worldwide and has a serious impact on wheat production. Recently, it demonstrated that barberry (alternate host) play an important role in generating new races and providing aeciospore inoculum to wheat causing stripe rust infection. However, the effect of susceptible barberry treated by fungicides on reducing the appearance of new races and genetic diversity of the stripe rust population has been unknown. Therefore, this study aimed at assessing the influence of the production of new races and population genetic diversity after fungicide treatment by comparing populations from wheat adjacent to barberry with/without fungicide spraying at early infection of pycnial stage in race compositions on differentials, and genetic diversity level using the 20K chip of the rust. As a result, the new race composition of the treatment (17 races) was simpler than that of the control (46 races). The former had a lower ($\alpha=16.415$) than the latter ($\alpha=18.261$) in virulence diversity based on near-isogenic lines. Population genetic diversity of the treatment was significantly lower than that of the control. Heterozygosity level of the treatment ($F_{hom}=0.3435$, Tajima's $D=0.17$) was significantly lower than that of the control ($F_{hom}=0.4140$, Tajima's $D=0.96$) ($P<0.05$). The nucleotide diversity of the treatment ($\pi=0.00015$) was significantly lower than that of the control ($\pi=0.00020$) ($P<0.01$). In conclusion, in areas where sexual reproduction occurs, timely fungicide application at pycnial stage of the stripe rust on barberry can significantly limit the population composition, even delay the appearance of new races, and population genetic diversity of the rust in fields, supporting the extension of growing wheat resistant varieties and achieving sustainable control of wheat stripe rust.

P020

USE OF SALICYLIC ACID-LOADED CHITOSAN NANOPARTICLES FOR THE MANAGEMENT OF CUCURBIT POWDERY MILDEW

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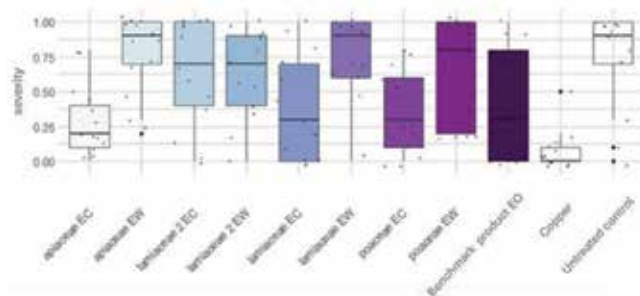
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Powdery Mildew (PM) is one of the most important foliar diseases in cucurbits, with substantial worldwide economic impact on yield quality and quantity. Nanotechnology serves as a promising novel tool in the agricultural sector for plant disease management. At this juncture, a combination of

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nanotechnology with natural substances such as chitosan may improve their efficacy along with their unique modes of action. The present study aims to gain insights into the priming effect of chitosan nanoparticles loaded with salicylic acid (NCS-SA), and the defense responses induced by NCS-SA in zucchini (*Cucurbita pepo* L.) against PM. The efficacy of NCS-SA against the pathogen was evaluated in a susceptible commercial zucchini cultivar in greenhouse trials. Overall efficacy of NCS-SA on leaves was calculated using the Area Under the Disease Progress Curve (AUDPC) values obtained from the disease severity data. RNA sequencing approach (RNA-Seq) was chosen to elucidate the induced defense mechanisms of zucchini plants. Our results showed that zucchini plants treated with NCS-SA showed reduced PM severity under high inoculum pressure. Furthermore, the transcriptomic analysis, at three time points during the priming phase, depicted the differential expression of the inositol phosphate metabolism, the phosphatidylinositol signaling pathway, circadian rhythm, DNA replication, ribosome, and proteasome pathways. Thirty hours post inoculation, the pathways of plant hormone signal transduction, starch and sucrose metabolism, and the MAPK signaling pathway were differentially expressed. Our results indicate that the foliar application of NCS-SA induced zucchini defense mechanisms against PM results in significant reduction of disease severity, therefore could be used in novel targeted disease management strategies.

families since their efficacy range reached 100 % and their variability is smaller than essential oil reference product.



On the other hand, not surprisingly, copper treatment confirms the highest protection against the oomycete. It is interesting to note that 2 oils belonging to the same botanical family (Lamiaceae 1, Lamiaceae 2) show opposite control efficacy, this could allow speculation on the weak co-relation between the origin of the oil and its biological effect.

P021

EFFICACY SCREENING OF DIFFERENT FORMULATES BASED ON ESSENTIAL OILS AGAINST GRAPEVINE DOWNY MILDEW

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Traditional fungicides were part of the boost to grapevine production worldwide, providing control to fungal pathogens. Nevertheless, over the years public concern about environmental and human health raised constantly. Thus, recently, an increased number of studies regarding more eco-friendly active ingredients such as plant essential oils (EOs) showed up. This work aims to show the rationale of a screening process to evaluate the action of four different essential oils belonging to three botanical families *Lamiaceae*, *Apiaceae*, *Poaceae* tested with two different formulations: Emulsifiable Concentrate (EC) and Emulsion in water (EW), against the causal agent of grapevine downy mildew. Trials were carried out in greenhouse conditions on 2-year-old grafted vines. For antifungal assay different potted grapevines were sprayed with copper and EO benchmark product, based on sweet orange EO as reference, the 8 formulates based on EOs and bi-distilled water as control. Artificial inoculations with a sporangia suspension of *P. viticola* have been performed with 10 drops of 10 µl on 5 leaves per each thesis, 3 days after treatment. Severity was evaluated at symptom appearance. The experiment was repeated three times. Results (figure) showed a clear different trend between the two formulates ($p < 0.001$) and a considerable variability among essential oil products both in formulates and benchmark product. *Lamiaceae* and *Poaceae* formulated with EC are the most promising botanical

P022

HARNESSING NATIVE BIOAGENTS FOR SUSTAINABLE MANAGEMENT OF FALL ARMYWORM

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Exploitation of biological agents in the management of the invasive fall armyworm is considered the most sustainable strategy. This is particularly important for sub-Saharan Africa, where smallholder farmers form majority of staple food producers. A field study was thus conducted to assess the effectiveness of simultaneous deployment of a parasitoid *Telenomus remus* (Hymenoptera: Pterigidae), and native entomopathogenic fungi *Bauveria bassiana* and nematode *Metarhizium anisopliae* in managing fall armyworm *Spodoptera frugiperda* (Lepidoptera: Noctuidae).

T. remus was released on 4 weeks old maize followed by application of *B. bassiana* and *M. anisopliae* after 24 hours. Treatments were sole *T. remus*, *T. remus* + *B. bassiana*, *T. remus* + *M. anisopliae* and *T. remus* + *B. bassiana* + *M. anisopliae*. Egg mass parasitism, larval mortality, incidence of *S. frugiperda* infestation, leaf and cob damage, and yield were assessed. FAW egg mass parasitism were similar across sole *T. remus*, *T. remus* + *B. bassiana*, *T. remus* + *M. anisopliae* and *T. remus* + *B. bassiana* + *M. anisopliae* treatments. Larval mortality of *S. frugiperda* 72 hours after treatment with the entomopathogens was similar among parasitoid and entomopathogen combinations, and these were higher than sole parasitoid. Maize leaf damage and new infestation by *S. frugiperda* significantly reduced and remained low after deploying the native bioagents. Maize cob damage at harvest and yield were not different among sole parasitoid and parasitoid + entomopathogen combinations. The parasitoid *T. remus* can be complemented with either entomopathogenic nematode *M. anisopliae* or entomopathogenic fungi *B. bassiana* as part of integrated management of *S. frugiperda* in maize.

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P023

MIXED INSECT PEST POPULATIONS OF DIASPIDIDAE SPECIES UNDER CONTROL OF OLIGONUCLEOTIDE INSECTICIDES

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Diaspididae are one of the most serious small herbivorous insects with piercing-sucking mouth parts and are major economic pests as they attack and destroy perennial ornamentals and food crops. Chemical control is the primary management approach for armored scale infestation. However, chemical insecticides do not possess selectivity in action and not always effective enough for the control of armored scale insects. Our previous work with aphids, psyllids, mealybugs showed that green oligonucleotide insecticides (olinscides) based on antisense DNA fragments of pest rRNA genes are highly effective against armored and soft scale insects. Moreover, olinscides possess affordability, selectivity in action, fast biodegradability, and a low carbon footprint. Insect pest populations undergo microevolution and olinscides should take into account the problem of insecticide resistance. By using olinscides 11 nucleotides long, sufficient selectivity can be achieved. In the case of more complex biocenoses, for example forests, the length of the olinscides can be increased up to 18 nucleotides, thereby providing the necessary selectivity in action. Ribosomal RNA makes it possible, on the one hand, to use itself as an advantageous target because it comprises majority of cell RNA, whereas, on the other hand, it is sufficiently variable among organisms to create well-tailored insecticides. If insecticide resistance occurs, different strategies can be applied. Generally, new olinscides can be created displacing target site to the left or to the right from the olinscides-resistance site of the 28S rRNA. Research results obtained within the framework of a state assignment V.I. Vernadsky Crimean Federal University for 2024 and the planning period of 2024–2026 No. FZEG-2024-0001.

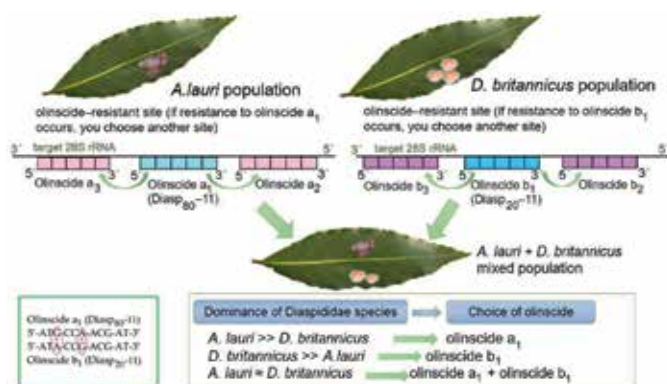


Figure. Algorithm of insect pest control with olinscides in mixed insect pest populations of Diaspididae species

P024

PREVENTIVE POTENTIAL OF COLORED-FLESHED POTATO EXTRACTS AGAINST THE PATHOGEN RHIZOCTONIA SOLANI IN POTATO CROPS

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The global demand for food will increase by 50 % by 2050, posing the challenge of producing food more effectively with limited resources. Potatoes, highly demanded worldwide, face the threat of the *Rhizoctonia solani* pathogen, leading to increased use of agrochemicals, which pose risks to the environment and human health. Therefore, developing new formulations based on natural products to prevent this disease is a promising alternative.

In this context, previous studies by our research group have reported antifungal activity in colored-fleshed potato extracts (FCP) against *R. solani* in potato crops. However, a more detailed study of its preventive effect is required.

Therefore, our research objective is to determine the type of application and effective dosage of FCP extract in *Solanum tuberosum* plants inoculated with *R. solani*.

For this purpose, a greenhouse trial was conducted where FCP was applied in different forms and dosages, in randomized design with three repetitions per treatment and positive and negative controls for the pathogen. Additionally, photosynthetic parameters, enzymatic activity and antioxidant capacity, phenolic compounds, organic acids, and crop yield were determined.

In detail, in terms of photosynthetic performance, the type of application played a significant role where the spray application improved significantly parameters by 2.8 % compared to commercial fungicides. Additionally, enzymatic response and antioxidant capacity showed that lower doses of the extract provided better protection. Under FCP treatment, there was a decrease in the release of organic acids, suggesting an improvement in nutrient uptake by the plant, and an increase significantly in crop yield by 55 % compared to commercial fungicides.

In conclusion, the type of application significantly impacts the plant protection system, and in the case of FCP, it is improved at lower doses, proving to be highly effective. Thanks to these findings, we will delve into the optimization of the formulation that leads to an effective final product.

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P025

GENERATION OF A HIGHLY EFFECTIVE BIOFUNGICIDE, BASED ON GRAPEVINE BY-PRODUCT RESIDUES, FOR THE CONTROL OF RHIZOCTONIA SOLANI IN POTATO CROPS

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The control and prevention of diseases caused by pathogens in crops are constant concerns in agriculture, also in Chile, where the fungus *Rhizoctonia solani* has reduced potato production by up to 43 % without the use of preventive agrochemicals. Although commercial solutions exist, their adverse effects on human health and the environment are negative. Therefore, the search for new sources of effective antifungals presents itself as a promising alternative.

In this context, biological activity has been observed in grapevine residue extracts, due to their high concentration of stilbenes, which show *in vitro* antifungal effects against *R. solani*. Consequently, our objective is to determine the *in vivo* effectiveness and storage conditions of a prototype preventive biofungicide based on grapevine waste in *Solanum tuberosum* plants inoculated with *Rhizoctonia solani*.

To this end, a stability test was conducted under ambient temperature conditions, 4°C, -20°C, and 28°C, with and without light exposure. The total concentration of phenolic compounds was measured weekly using the Folin-Ciocalteu method, and antioxidant capacity was assessed using the chemical antioxidant methods for each treatment.

Initially, stability results at week 6 showed that the concentration of total phenols was more stable with an increase of 2.7 % under temperature of 28°C and darkness (TE), compared to the treatment at ambient temperature and light (TA), which increase by 20.15 %. Regarding color intensity, TE was the most stable condition, with only a 29 % of decrease. On the other hand, the antioxidant capacity of the extract was more stable in TE according to the TEAC and CUPRAC methods, where it only increase of 0.8 % and 10.09 %, respectively, compared to TA, which showed the highest variation.

In conclusion, temperature appears to be a variable of greater significance regarding the stability of the extract, where increasing the temperature results in the extract showing greater stability over time.

P026

PHYTOPHTHORA CINNAMOMI INHIBITION BY PHENYLACETIC ACID: MECHANISM OF ACTION AND ITS ISOLATION FROM TRICHOMONASCUS VANLEENENIUS CULTURES

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Oomycetes represent a group of fungi-like microorganisms that include some important pathogens affecting forest ecosystems. One of them is *Phytophthora cinnamomi*, which is known to be the main factor causing dieback, a plant disease responsible of the destruction of the *Quercus*, *Fagus*, *Pinus*, *Eucalyptus* and other valuable species. The aim of the present work was to characterize naturally-produced inhibitory molecules extracted from the culture media of other microorganisms such as the yeast *Trichomonascus vanleenenius*, first described in this work to produce phenylacetic acid (PAA).

PAA was obtained using organic solvent extraction, purified by high performance liquid chromatography (HPLC) fractionation and identified by mass spectrometry and nuclear magnetic resonance (NMR) analyses. Once identified, the anti-oomycete potential of PAA was tested by an *in-vitro* growth inhibition bioassay against *P. cinnamomi*. In addition, its plausible mechanism of action against *P. cinnamomi* was determined by confocal microscopy cellular viability and cell wall integrity bioassays.

PAA was successfully isolated from *T. vanleenenius* culture media and it showed mycelial growth inhibition bioactivity against *P. cinnamomi*. The *in-vitro* bioassays that were carried out in order to determine the mechanism of action of PAA at the cellular level showed that PAA increases cell mortality by disrupting both cellular wall and membrane integrities.

Taking all this information into account, we conclude that PAA is a natural compound produced by *T. vanleenenius*, among other organisms, which has biocide (anti-oomycete) bioactivity against *P. cinnamomi* mainly based on osmotic balance disruption. These facts allow PAA to be considered as a potential active ingredient of a future treatment to combat dieback worldwide expansion.

P027

ALTERNATIVE PLANT PROTECTION STRATEGY FOR STRAWBERRY DISEASE CONTROL

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Growing outbreaks of strawberry pathogens require new insights into its management. Strawberry diseases caused by *Botrytis cinerea* reduce yield by up to 50 %. The constant use of pesticides on strawberries leads to resistance. EU Green Deal encourages the reduction of the use of chemical pesticides. We aimed to evaluate plant-based substances as alternative plant protection products for strawberry pathogens control. The primary experiments were conducted at LAMMC Institute of Horticulture Laboratory of Plant

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Protection in 2023. Strawberries (cv. Elsanta) grown in plastic pots (3 litres, 19x15) containing peat substrate with NPK and microelements Mn, Cu, Mo, B, Zn and Fe. Plants were watered when needed, maintaining a similar substrate humidity. The experiment was arranged in randomised blocks, with 4 replicates (32 plants) per treatment. The first application was at 10 % flowering (BBCH 65–61), and spraying was repeated later every 10–7 days (total 4 times). Treatments: 1) CON - control, 2) CHEM - chemical fungicide, 3) BACT - bacterial suspension, 4) THY - *Thymus vulgaris* essential oil. The results revealed the treatment with BACT gave the highest yield (11.67 kg/32 plants). However, the highest fruit weight was in the CHEM (7.65 g/1 fruit). Fruit size highest was in BACT (26.96 mm) and CHEM (26.82 mm). The THY treatment had the highest soluble solid and ascorbic acid content. The firmness of the BACT and CHEM were similar. Based on primary results, we can conclude that BACT and THY can potentially serve as alternative plant protection products. However, further investigations are needed. Acknowledgement. This project has received funding from the Research Council of Lithuania (LMTLT), agreement No [S-NORDFORSK6-23-].

P028

FIELD TRIALS TO TEST EFFICACY AND SIDE EFFECTS OF NOVEL BIOINSECTICIDES ON BEMISIA TABACI

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Global temperature rise has favoured the spread of emerging crop pests and diseases. Among these is the whitefly *Bemisia tabaci*, a vector for economically important virus such as begomovirus. Vector control is crucial to reduce viral infections, but the overuse of chemical pesticides has resulted in the emergence of pest resistance and environmental harm. Therefore, alternative methods of integrated pest management and control, such as biopesticides and biological control agents, are being investigated.

The VIRTIGATION project aims to develop rapid and long-lasting solutions for emerging viral diseases caused by begomoviruses (whitefly-transmitted) and tobamoviruses (mechanically transmitted) on cucurbits and tomatoes in Northern Europe and the Mediterranean Basin. As part of this project, Fundación TECNOVA (TEC, Almería, Spain) has collaborated on implementing biorational whitefly control. This trial tested the efficacy of three new bio-formulates developed by KU Leuven (Belgium) against *B. tabaci*. This test was conducted in a randomised block design in a greenhouse tomato crop. To evaluate the efficacy of the products, adults and nymphs of *B. tabaci* were counted. In addition, assessments were realized on potential side effects to auxiliary insects: parasitoid activity of *Eretmocerus eremicus*, occurrence of predator *Nesidiocoris tenuis* and pollinator activity of *Bombus terrestris*. The performance of these natural products was compared to two reference market products, a chemical insecticide and a bio-insecticide. TEC results showed that the new bio-formulates were more

effective against whitefly populations than the commercial products, as well as less toxic towards the predator *N. tenuis*, which is commonly used as a biocontrol agent. Additionally, lower numbers of *N. tenuis* due to the chemical insecticide use failed to control whitefly population levels, highlighting the importance of product compatibility with biocontrol agents for successful integrated pest management.

P029

PROFILING RICE FARMS FOR THE OCCURRENCE OF MAJOR DISEASES AT DIFFERENT GROWTH STAGES IN THE ASHANTI REGION OF GHANA

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Diseases of rice reduce grain yield and quality significantly in major producing areas of Ghana. The limited availability of information on their incidence and severity led to the execution of this study to monitor and document major diseases affecting rice at different growth stages of the plant on farmers' rice fields in Ashanti Region of Ghana.

Rice disease surveillances were conducted in 2022 and 2023 in nine rice-growing communities. A total of 108 farms were assessed for disease incidence and severity using the Standard Evaluations System of Rice (SES). These surveys were done at four growth stages of rice viz. nursery, vegetative, reproductive and ripening stages.

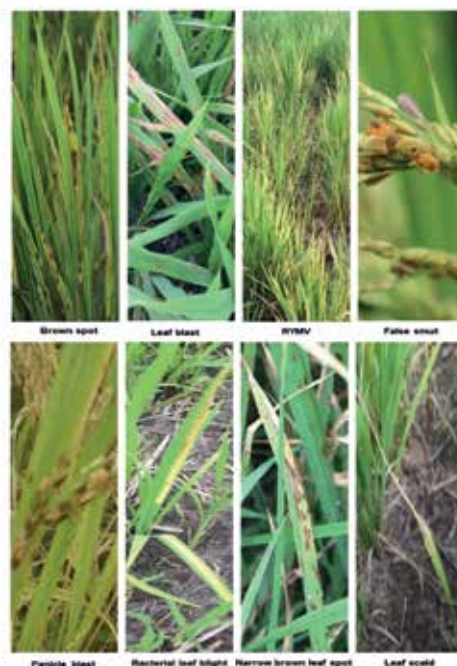


Fig. 1. Major diseases observed on rice fields in the Ashanti Region of Ghana.

Ten rice diseases were observed on rice fields in the Ashanti Region. Five diseases were observed at all growth stages of the rice plant and generally progressed with time. Brown

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spot and leaf blast were the most predominant diseases within the region with mean incidences of 75.4 % and 68.7 % respectively at the ripening stage. Again, both similarly exhibited higher severity from nursery to ripening stage. There was no significant difference in disease occurrence among the communities for all the observed diseases with the exception of False smut, with Noboam recording the lowest incidence. Co-infection of pathogens was observed in several instances during the surveillance at all growth stages of rice. Brown spot and leaf blast continue to be the most important diseases of rice as they were observed from nursery to ripening stage of the plant. This study is useful in designing an integrated disease management for major rice diseases observed in the region.

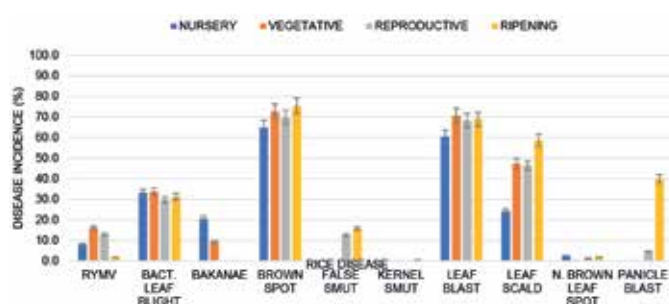


Fig. 2. Mean disease incidence of diseases at the various growth stages of rice.

P030

CORRELATION BETWEEN TETRALINIPROLE RESISTANCE AND RYANODINE RECEPTOR MUTATIONS IN LABORATORY-SELECTED STRAIN AND FIELD POPULATIONS OF PLUTELLA XYLOSTELLA IN TAIWAN

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Insecticide resistance in *Plutella xylostella* (Linnaeus) (Lepidoptera: Plutellidae) is one of the major constraints on the production of cruciferous crops globally. For effective management of insecticide resistance, it is necessary to develop a molecular detection tool for predicting insecticide resistance levels based on the mutation frequency of target sites. In this study, a susceptible strain of *P. xylostella* (SH_{ggt}) was exposed to tetraliniprole selection under laboratory conditions to obtain the TET_{Sel} strain. Mutation frequencies of *P. xylostella* ryanodine receptor (*PxRyR*) gene and other parameters of cross resistance in selected strain and tetraliniprole resistance in field were determined. The results have shown that continuous selection over 30 generations resulted in the development of resistance ratios (RRs) of ~6,971.0fold for the TET_{Sel} strain. Three out of seven field populations have developed more than thousand-fold resistance to tetraliniprole. Among the three investigated target-site mutations in *PxRyR*, only I4790K was detected in laboratory-selected strain. However, I4790K and G4946E were detected in field populations. A positive correlation between tetraliniprole RRs and K allele frequencies was observed in both the TET_{Sel} strain and field populations of *P. xylostella*. These results imply a possible link between

anthranilic diamides resistance development and I4790K mutation frequencies of *PxRyR*, which can be used to develop effective strategies for diamide resistance management in *P. xylostella*.

P031

INSECT PEST POPULATION DYNAMICS IN RICE: A CASE IN ASHANTI REGION OF GHANA

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Ghana is striving to be self-sufficient in rice and its rapidly expanding. Rice intensification, like most monocultures, has increased reports of pest outbreaks in some areas. This study aimed at profiling rice insect pest dynamics under current production practices.

Insect pest populations over rice growth cycle were monitored over 2 years in 108 rice fields in 9 key rice growing communities in Ashanti Region. Insect pests were sampled using sweep net, and direct observation along transect from nursery to maturity stage. Deadhearted rice plants were counted as indirect incidence of stem borers and samples with live larvae incubated in the laboratory until moth emergence. A total of 29 insect pest species were identified, of which 13 were considered major pests. The major insect pests were the stem borers *Chilo zacconius*, *Scirpophaga sp.*, *Maliarpha separata*, *Nymphula sp.*, *Diopsis thoracica*; leaf beetles *Chnootriba similis*, *Xanthadalia sp.*, *Lema sp.*, *Chaetocnema sp.*, *Diadisa viridicyanea* and sucking bugs *Leptocorisa sp.*, *Riptortus sp.*, *Aspavia armigera*. The leafhoppers *Nephotettix modulates*, *Cofana spectra*, *Cofana unimaculata* were prevalent but were not considered major pest, because no leafhopper burn associated with them was recorded. As vectors of yellow mottle virus, they could be important in areas where the disease is prevalent. All the major pest populations, except *Riptortus sp.*, peaked at the vegetative stage. Rice leaf beetles occurred in all surveyed areas but species distribution differed among locations, and *Nymphula sp.* seems to be endemic in some locations. The mean incidence of deadheart was 8.2 % while leaf damage by caseworm and leaf beetles was 11.42 % at 2.3 severity. Major insect pests and population build up at different growth stages of rice in the study area is understood and can form basis for the development of IPM package for rice production.

P032

EVIDENCE FOR THE PARTICIPATION OF CHEMOSENSORY PROTEINS IN RESPONSE TO INSECTICIDE CHALLENGE IN CONOPOMORPHA SINENSIS

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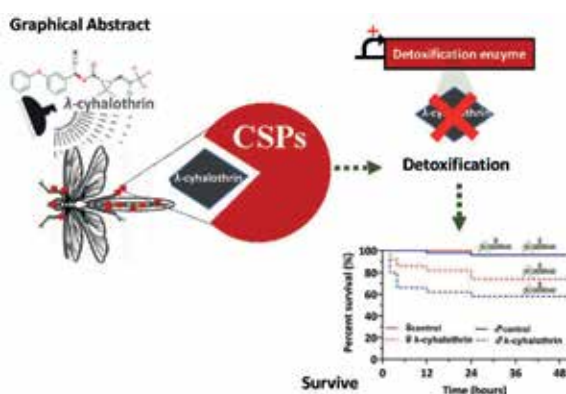
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Chemosensory proteins (CSPs) are a type of efficient transporters that can bind various hydrophobic compounds. Previous research has shown that the expression levels

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of some insect CSPs were significantly increased after insecticide treatment. However, the role of CSPs in response to insecticide challenge is unclear. *Conopomorpha sinensis* is the most destructive borer pest of litchi and longan in the Asian-Pacific region.

Here, we studied the expression patterns and potential functions of twelve CSP genes (*CsCSPs*) from *C. sinensis* in response to λ -cyhalothrin exposure. The spatiotemporal distribution of *CsCSPs* suggested that they were predominantly expressed in the female abdomen, female legs, and male legs. The expression levels of *CsCSPs* were affected in a time-dependent manner after λ -cyhalothrin treatment in both sexes of *C. sinensis* adults. Compared to the control group, the expression levels of *CsCSP1*, *CsCSP2*, *CsCSP9*, and *CsCSP12* in females were significantly increased by two to four times, while only one *CsCSP*, three *CsCSPs* and two *CsCSPs* were significantly upregulated in males at three time-points posttreatment, respectively. Homology modeling and molecular docking analyses showed that the binding energy value of *CsCSP1*–12 to λ -cyhalothrin was negative, and the binding energy between *CsCSP9* and λ -cyhalothrin was the lowest (-11.35 kJ/mol). Combined with expression alterations of *CsCSP1*–12, the results indicate that four *CsCSPs* were involved in binding and ferrying of λ -cyhalothrin in *C. sinensis*.



Caption: The sex-biased variance of CSPs might be closely related to sex-specific insecticide metabolism and survival rates of *C. sinensis* adults in response to λ -cyhalothrin challenge.

In conclusion, we have identified genes belonging to the CSPs family in *C. sinensis*. The sex-biased variance of expression levels of *CsCSP1*, *CsCSP2*, *CsCSP9*, and *CsCSP12* was related to sex-specific insecticide metabolism and different survival rates of female and male *C. sinensis* adults.

P033

RISKS FOR MICRONUTRIENT LOSSES DUE TO VECTOR-BORNE PLANT VIRUSES OF NUTRITIOUS CROPS IN SUB-SAHARAN AFRICA

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Vector-borne plant viruses decimate food crops worldwide: they present a complex system of interactions, involving

the host plant, the viruses and the insects that transmit the viruses. The global impact becomes apparent when we consider that 75 % of the world's food comes from only 12 plant and 5 animal species, and that 20-40 % crop yields are lost annually to pests and diseases. These effects are felt more acutely in vulnerable regions such as Sub-Saharan African (SSA). This climate and health-focused project aims to: 1. Assess how plant viruses are impacting the availability of foods that support a healthy African traditional diet, and 2. Identify SSA countries at greatest risk of micronutrient restriction due to these viruses.

Using computational data analysis, this project is linking publicly-available crop production, food security and micronutrient composition datasets with vector-borne plant virus epidemiology information and our recently-determined African traditional diet composition.

This project is identifying regions of SSA where the most traditionally-important food items and groups are most at risk from vector-borne plant viruses alongside the concurrent impact on key micronutrient supplies. It is also identifying SSA regions at risk of specific micronutrient deficiencies, in the context of contributions to food supply quantities. Next steps include integrating climate change impact data to project possible future risks to regions vulnerable to virus-induced losses of traditionally-, and nutritionally-important crop. Results are highlighting significant gaps in knowledge of vector-borne plant virus impacts on food supplies.

The impacts of vector-borne plant viruses are understudied: international surveillance efforts should be increased to ensure risks to nutritionally-diverse food supplies are understood so that appropriate management strategies can be implemented.

P034

RESISTANCE MECHANISMS TO GLYPHOSATE AND GLUFOSINATE IN ELEusine INDICA

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Eleusine indica (goosegrass) is one of the most economically important weed species in tropical and sub-tropical regions globally. Glyphosate and glufosinate, two of the most important herbicides, have been used for *E. indica* control. Consequently, resistance to these two herbicides have evolved in many goosegrass populations around the world, including those in China. We investigated the target-site resistance mechanisms to glyphosate and glufosinate. For glyphosate resistance, there are multiple mechanisms: target-site EPSPS double mutation (TIPS), EPSPS overexpression (inducible), EPSPS copy number variation (CNV). TIPS mutation in combination with EPSPS overexpression and CNV usually gives higher level of glyphosate resistance without evident resistance costs. EPSPS promoter sequence analysis revealed two insertions (3 and 12 bp) in the resistance (R) samples in the 5'UTR. Promoter activity analysis showed that the 12bp insertion

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resulted in significant enhancement of the R promoter activity, whereas the 3bp insertion had little effect. So, it is likely this 12 bp insertion in the R promoter causes inducible EPSPS overexpression. To understand genomic nature of EPSPS CNV, we obtained high-quality reference genomes for R and susceptible (S) plants, fine assembled ESPPS gene duplication and revealed a unique rearrangement of EPSPS gene involving chromosome sub-telomeres, hot spots for structural variation and thus adaptive trait evolution. For glufosinate resistance, we identified a novel point mutation resulting in Ser59Gly substitution in glutamine synthetase (GS1) in R plants. Rice calli and seedlings transformed with the mutant GS1 gene were resistant to glufosinate. Yeast-expressed GS1 was more tolerant to glufosinate than the wild-type variant. Structural modelling of the mutant GS1 revealed that Ser59 is not directly involved in glufosinate binding but is in contact with some important binding residues (e.g. Glu297) and especially with Asp56. Importantly, the same Ser59Gly mutation was also found in geographically isolated glufosinate R populations from Malaysia and China.

recurrent outbreaks pending efficient control measures to curb its severe impact. The epidemiology of the disease is still unclear as host plants serving as virus reservoirs and the role of whiteflies in early infections are yet to be elucidated.

Aknowledgements. This study is co-financed by the European Union-Next Generation EU, Greece 2.0 National Recovery and Resilience plan, National Flagship Initiative "Agriculture and Food Industry" as part of the project "Innovations in Plant Protection for sustainable and environmentally friendly pest control, InnoPP - TAEDR0535675-".

P036

GENETIC DIVERSITY OF 'CANDIDATUS PHYTOPLASMA SOLANI' AFFECTING GRAPEVINE IN GREECE

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'*Candidatus* Phytoplasma solani', a prokaryotic phytopathogen known to occur in Greece, has a wide range of host plant species, including grapevine, for which this pathogen is of major economic importance. Since grapevine cultivation contributes significantly to the agricultural economy of Greece, a systematic monitoring of the presence of 'Ca. Phytoplasma solani' in vineyards in the Regional Units of Greece has been initiated by the Laboratory of Bacteriology of BPI (LB-BPI) with the aim at determining the geographical distribution of this pathogen in the country and characterizing its genetic diversity. For this monitoring, archived DNA samples of grapevine plants, which have been extracted in the LB-BPI and stored at - 20°C, were screened for the presence of 'Ca. Phytoplasma solani' using molecular diagnostic methods (real-time/conventional PCR, 16S rDNA sequencing). These samples originated from: A) asymptomatic grapevine plants, sent to the LB-BPI by Phytosanitary Inspectors since June 2022 for testing the presence of *Xylella fastidiosa* and Grapevine flavescence dorée phytoplasma, in the frame of the Official National Surveys and Phytosanitary Checks in order to issue plant health certificates or plant passports. B) symptomatic grapevine plants, sent by growers, cooperatives, agronomists, etc to the LB-BPI for disease diagnosis. On the 'Ca. Phytoplasma solani' strains identified, multiple genotyping was performed using a series of genes and phylogenetic analysis was implemented on the gene sequences obtained. The screening of the DNA extracts has led to the identification of 'Ca. Phytoplasma solani' in eight Regional Units, including main wine-producing regions in Greece. Genotyping of the identified strains is ongoing to obtain information on the diversity of pathogen's haplotypes in the country. The obtained results are expected to shed light into the epidemiological traits of this important for grapevine phytopathogen in Greece and contribute to the development of an efficient program of measures for its management.

P035

TOMATO LEAF CURL NEW DELHI VIRUS AS AN EMERGING PATHOGEN OF GREEK CUCURBITS

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The whitefly-transmitted tomato leaf curl New Delhi virus (ToLCNDV, genus *Begomovirus*) is an emerging pathogen of Cucurbitaceae crops in the Mediterranean basin and was first reported in Greece, in 2018. This ongoing study is to record ToLCNDV spread and understand the drivers of its epidemics in the Greek cucurbits.

To determine virus incidence in crops and the range of natural hosts, surveys were performed in major cucurbit production areas targeting cultivated and weed species. From 2022 onwards, more than 1700 samples belonging to 64 plant species in 26 families were collected and tested for ToLCNDV by ELISA; cucurbits were also tested for additional viruses. To examine the genetic diversity of ToLCNDV, selected ELISA positive samples were analysed by rolling circle amplification followed by genome sequencing.

ToLCNDV occurs in zucchini and/or cucumber crops in Lasithi (Crete), Ilia, Argolida (Peloponnese) and Viotia prefectures in rates of up to 90 %. Severe diseases are observed in ToLCNDV-infected open-field zucchinis especially when in mixed infections with cucurbit aphid-born virus or cucumber mosaic virus. More than 55 weed species have been tested however, infection of only squirting cucumber (*Echallium elaterium*) was confirmed. All ToLCNDV isolates from cucumber collected in Irapetra (Crete), zucchini in Ilia and squirting cucumber in Argolida shared closest genetic relationship with the ToLCNDV-ES strain.

In Greek cucurbits, ToLCNDV is a main component of



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P037

OCCURRENCE OF CUCUMBER MOSAIC VIRUS AND WATERMELON MOSAIC VIRUS IN GREEK CUCURBITS AND THEIR INTERACTIONS WHEN TRANSMITTED BY APHIS GOSSYPYII

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The aphid-transmitted cucumber mosaic virus (CMV, *Cucumovirus*) and watermelon mosaic virus (WMV, *Potyvirus*) affect open field cucurbits resulting in severe crop losses, worldwide. The aims of this study were to record their incidence in Greek crops and study their interaction when transmitted by *Aphis gossypii* Glover.

In surveys, performed in 2022 and 2023, in open field crops in Greece, more than 500 cucurbit plants with virus-like symptoms were sampled and tested by ELISA for the presence of CMV, WMV, and other aphid-transmitted viruses. A colony of *A. gossypii*, grown on zucchini plants was used to test the transmission of CMV and WMV in single, sequential and mixed infections using zucchini plants of cv Orfeus F1, in 15 replications each. Ten wingless aphids, after a starving period of 1 h, were given an acquisition period of 3 min in single (with one or each one of CMV and WMV) or mixed infected plants. Aphids were then transferred individually to healthy plants, for 24 h that were tested 3 wks later for virus presence.

CMV and WMV were detected in almost all sampling areas occasionally in rates of up to 100 % in individual crops. Mixed infections were commonly encountered in rates of up to 42 %. In aphid-transmission studies, CMV and WMV were transmitted with a similar efficiency when acquired from plants with single infections (mean transmission rate of 88 %). Significantly lower rates (~80 %) were recorded for both viruses when acquired either from mixed-infected plants or in any combination of sequential acquisition from single infections.

WMV and CMV are prevalent viruses in the Greek open field cucurbits. Their mixed infections occur also frequently, but it seems that they do not favor their spread by *A. gossypii*. However, further research is required to understand all the complex plant-virus-vector interactions.

P038

PATHOGENIC SEED HEALTH SCREENING AND FUSARIUM MYCOTOXINS IN WHEAT SEED LOTS PRODUCED IN URUGUAY

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Identification and quantification of seed pathogens and mycotoxin safety are useful for the development of integrated management strategies. The aims of the study were a) to

determine the level of infection of wheat seed lots in the northern coastal area of Uruguay; b) to study the relationship between the level of infection of wheat seed lots and the weather conditions during their production; c) to determine the level of mycotoxins produced by the *Fusarium* genus in each lot; d) to correlate the prevalence of *Fusarium* in the seed lots with the concentration of mycotoxins detected in each lot. To achieve these objectives, 23 lots from the main wheat production area in Uruguay (2020), were selected. For each lot, 400 seeds were used for the determination of the level of infection by blotter test. The pathogens detected in the test were isolated on culture media and the main strains detected were identified by molecular techniques. Additionally, the levels of *Fusarium* mycotoxins were determined by HPLC-MS/MS. The data obtained were analyzed in relation to the climatic data of each farm. Preliminary results showed the presence of pathogens such as *Bipolaris sorokiniana*, *Fusarium graminearum*, *Drechslera repentis*, *Zymoseptoria tritici*, *Ustilago tritici*, and the contaminants *Alternaria* spp. and *Epicoccum* spp. The presence/absence of mycotoxins will be connected to *Fusarium* detection in each lot and discussed based on their MRLs. The findings will offer valuable insights into the fungal load and safety of seed lots produced under different weather conditions, for optimal seed use in the wheat sector.

P039

ENTEROBACTER CLOACAE COMPLEX A NOVEL PLANT PATHOGEN GROUP AFFECTING SUGARCANE LEAVES

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Enterobacter cloacae complex is a group of opportunistic pathogens found in sewage, soil, hospital environments, and the intestinal tract of humans and animals. The *Enterobacter cloacae* complex has recently been identified as a causative agent of reddish streaks in sugarcane in Mexico. However, the response of the varieties planted at the national level is unknown; therefore, this research aimed to evaluate the response of the four varieties with which the 830,000 ha are planted nationally. Twelve isolations previously identified by the multilocus phylogenetic approach as *Enterobacter cloacae* complex were evaluated to determine their responses on ITV1424-92-, Mex290-69-, CP 2086-72, and Motz-Mex 401-01 commercial varieties. They were inoculated by the injection method with 10^7 CFU mL⁻¹ and maintained in greenhouse conditions for 45 days. Early symptoms appeared 15 days after inoculation in the Motz-Mex 401-01, in which small reddish streaks were clearly visible on the adaxial surface of the sugarcane leaves, when they were inoculated with TAB16, 2CV, HC1, and SM1 strains. The Mex290-69- variety was found to have the lowest incidence percentage, and the ITV1424-72- and CP2086-72- varieties were found to have the highest. The results demonstrate that the *Enterobacter cloacae* complex is the cause of reddish streaks in sugarcane, and this emerging disease is already found in the seven producing regions of Mexico. Identification of



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the *Enterobacter cloacae* complex will enable producers to implement plant protection measures to reduce the spread of the inoculum and the selection of resistant genotypes to reduce producers' losses from this bacteria group.

P040

RELATIONSHIP OF CLIMATE WITH LATE BLIGHT (PHYTOPHTHORA INFESTANS (MONT.) DE BARY) ON POTATO (SOLANUM TUBEROSUM L.) CROP

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Potato (*Solanum tuberosum*) is the fourth most important crop worldwide. The crop is affected by the climate variability, mainly in relation to diseases severity and pathogens survival, which is the case of late blight caused by *Phytophthora infestans*. The objective of this study was to evaluate the effect of the climatic elements on late blight severity. The study was carried out in the municipality of Mosquera, Cundinamarca, Colombia, from December 2015 to May 2016. The potato varieties Diacol Capiro (DC), ICA-Unica (IU) and Pastusa Suprema (PS) were included in this study. Data of late blight severity together with data of precipitation (Pr), relative humidity (RH), temperature (T), wind speed (WS), solar brightness (SB) were recorded, and with these data the agroclimatic indices water index (IH), temperature degree day (DD) and evapotranspiration reference (ETo) were calculated. There was a significant positive correlation ($p < 0.05$) of late blight severity in the three varieties with IH ($0-158 \pm 58$), precipitation ($0-63 \text{ mm} \pm 23 \text{ mm}$) and minimum temperature ($2-10 \text{ }^\circ\text{C} \pm 2 \text{ }^\circ\text{C}$), and significant negative correlation ($p < 0.05$) of *P. infestans* severity with SB ($2-7.7 \text{ h day}^{-1} \pm 1.7 \text{ h day}^{-1}$). The DD ($0-4 \text{ }^\circ\text{Cd} \pm 1 \text{ }^\circ\text{Cd}$) presented a significant positive correlation with DC and PS, and positive correlation with the mean temperature ($11-15.3 \text{ }^\circ\text{C} \pm 1.41 \text{ }^\circ\text{C}$) with DC. The ETo ($0.4-3.3 \text{ mm day}^{-1} \pm 0.87 \text{ mm day}^{-1}$) and maximum temperature ($19.9-23 \text{ }^\circ\text{C} \pm 0.75 \text{ }^\circ\text{C}$) were significantly and negatively correlated with DC and PS, and between VV ($5.27-7.95 \text{ km h}^{-1} \pm 0.79 \text{ km h}^{-1}$) with DC. It was concluded that climatic variables influenced late blight severity and *P. infestans* survival.

P041

ENHANCING GERMPLASMS ACCESS: CASE STUDY TO IDENTIFY GENETIC VARIANTS FOR CASSAVA BROWN STREAK DISEASE RESISTANCE IN AN INTERNATIONAL GENE BANK COLLECTION

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Genebanks, through ex-situ conservation, play a pivotal role by preserving crop diversity, providing a crucial resource for breeding resilient and disease-resistant crops. In particular, cassava (*Manihot esculenta*), a key carbohydrate source for over 500 million people in Africa and both regions, faces challenges due to Cassava Brown Streak Disease (CBSD).

The International Center of Tropical Agriculture (CIAT) holds the world's largest cassava collection, encompassing 5,963 accessions from 28 countries, primarily from South America. The goal of this study was to perform a genome-wide association analysis to identify genetic variants associated with CBSD. This analysis was conducted on a panel of 234 accessions of South American germplasm conserved at CIAT, previously phenotyped by an independent research group (Sheat et al., 2019). Samples were genotyped using DArTseq, resulting in a total of 121,405 single-nucleotide polymorphism (SPN) markers. After applying a series of filters for sample and marker selection, we assessed two statistical models using the Gapit v3.0 tool. We compared the obtained results across models and annotated interesting markers. Throughout the study, we identified 28 SNP markers distributed across various chromosomes, associated with disease severity or presence/absence of virus infection, based on Sheat et al. results. After annotating markers, one of the 28 was previously reported; the others were located within or near genes previously identified with functions related to pathogen recognition and immune response activation. A set of new cassava accessions, potentially resistance to the virus, were identified within the genebank. The results of this study represent a valuable contribution for improving cassava against CBSD and highlight the importance of genetic resources, to provide breeders with the raw materials needed to develop new crop varieties resistant to pests and diseases. This helps enhance agricultural productivity, promote food security, and contributes to global efforts in mitigating the impact of plant-related threats on crops and ecosystems.

P042

IS EUROPEAN WHEAT PRODUCTION READY FOR THE RE-EMERGENCE OF STEM RUST?

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Wheat stem rust, caused by *Puccinia graminis f.sp. tritici*, continues to spread northwards, threatening European wheat production. This has already been evidenced by local outbreaks and epidemics from Sicily to Sweden, raising the question of whether Europe is prepared for future stem rust attacks. To this end, the European stem rust populations have been monitored by the RustWatch network, funded by the European research initiative "HORIZON 2020". This, along with national projects, investigated the susceptibility of wheat varieties to stem rust and the efficacy of selected fungicides. In order to monitor European stem rust populations, infected samples were collected from various areas, both from wheat

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fields and the alternate host barberry. They were analysed using international differential sets and molecular markers which showed that although only a few genetic groups dominate the European stem rust population, the genetic diversity of stem rust isolates in the neighbourhood of barberry was very high.

In Germany, the stem rust resistance of registered wheat varieties is evaluated as part of VCU testing. Taken the cropping area, 28 % of the tested varieties proved to be resistant, 33 % moderately susceptible and 38 % highly susceptible to stem rust. In a national research project called GetreideProtekt, we tested a panel of 280 European wheat varieties in multi-year and multi-site field trials. Using association mapping, only three genes, *Sr31*, *Sr24* and *Sr38*, were localised individually or in combinations thereof. For all three genes, some virulent isolates have already been found in Europe. Strategies for optimised fungicide use were also developed. Several fungicides proved to be effective, especially when applied close to the time of infection.

In conclusion, more high-throughput methods for phenotyping and genotyping of stem rust isolates, as well as new sources of resistance, are needed to protect the European wheat production from future stem rust attacks.

P043

DYNAMIC TRANSCRIPTIONAL REPROGRAMMING ACTIVATED BY WHEAT TANDEM KINASE 1 LEADS TO ENHANCE RESISTANCE TO YELLOW RUST DISEASE

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Plant diseases pose significant threats to global food security. Stripe rust is one of the most wheat-devastating diseases caused by the fungus *Puccinia striiformis* f. sp. *tritici* (*Pst*). Developing disease-resistant crops offers a cost-effective solution for sustainable wheat production. *Yr15* is a broad-spectrum resistance gene derived from wild emmer wheat (*Triticum dicoccoides*) gene that confers resistance against >2000 *Pst* isolates from 47 countries around the globe. *Yr15* encodes a tandem kinase-pseudokinase protein architecture designated wheat tandem kinase 1 (WTK1), which is a new player in plant immunity. WTK1 induces form of immune cell death, called the hypersensitive response (HR), that occurs after fungal haustoria development. To investigate the transcriptional changes activated by WTK1 in response to infection with *Pst*, we conducted transcriptional time-course analyses of a susceptible durum wheat genotype, Kronos, compared with its resistant near-isogenic transgenic line, Kronos+WTK1, after inoculation with *Pst*. To simultaneously identify differentially expressed genes between genotypes over time, we used the R package Moanin. By spline clustering, we identified 20 differentially expressed gene clusters that exhibit various regulation patterns. Thirteen clusters were upregulated in the resistant line and were enriched in genes involved in pathogen recognition, signal

transduction, phytohormone production, and HR cell death responses. Pairwise comparisons and weighted gene co-expression network analyses pinpointed 11 modules specific to time points post-infection. We identified a genotype-specific module containing upregulated genes in the resistant Kronos+WTK1 genotype. This module displayed an enrichment of genes involved in basal defense pathways, such as phytoalexin production, callose deposition, and antifungal activity, as well as ROS and HR-cell death immune responses. Thus, the comprehensive transcriptome analysis of wheat NILs revealed intricate genetic networks triggered by WTK1 in response to *Pst* infection. These interactions result in robust defense signaling pathways, ultimately leading to disease resistance.

P044

EVALUATION OF SWEET CHERRY (PRUNUS AVIUM) RESISTANCE TO PSEUDOMONAS SYRINGAE

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Sweet cherry production in Latvia is located at the Nordic margin of its traditional cultivation; therefore, a breeding program has been focused mainly on winter hardiness and fruit quality. Due to the increase in production, local cultivar assortment, climate change and associated pathogen expansion risks, disease resistance nowadays is also considered. *Pseudomonas syringae* is a pathogen of global importance on various fruit crops, causing canker, dieback, and necrosis on leaves, blossoms and fruits. It is a highly diverse species complex divided into pathovars, phylogroups, and genomospecies. The damages caused can vary greatly, depending on the pathogen and the host susceptibility. In Latvia, a diverse population of *P. syringae* has been found on fruit crops, including *Prunus*. This study aimed to evaluate the resistance of sweet cherry genotypes from the local breeding program (No.-1654/23/11-10.9.1e) to diverse *P. syringae* strains. Immature, detached fruits of five locally bred genotypes and two control cultivars (resistant and susceptible) were inoculated with 21 pathogen strains originating in Latvia from different woody hosts. The final disease assessment was done one week after inoculation. Each genotype was susceptible to one ('Regina') to seven ('Bigarreau Burlat') pathogen strains. Genotypes from local breeding programs were susceptible to three to four strains. One *P. syringae* pv. *syringae* strain from sour cherry was the most aggressive and virulent on all tested genotypes, while three others from the same host did not cause noteworthy necrosis. Our data demonstrated the significant variation of the host resistance and virulence of the pathogen, confirming the significant role of the host's resistance in disease control. The study will be continued on more locally bred genotypes, including on woody tissues, since the bacterial canker is a major limiting factor in the cherry industry due to the lack of effective control measures.

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EVALUATION OF RESISTANCE OF OLIVE CULTIVARS TO COLLETOTRICHUM ACUTATUM, THE LEADING CAUSE OF OLIVE ANTHRACNOSE DISEASE IN GREEK ORCHARDS

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Olive anthracnose disease, caused by fungi belonging to *Colletotrichum* spp., poses a significant threat to olive cultivation worldwide, causing severe yield losses and deterioration of olive oil quality. Among *Colletotrichum* species, strains belonging to *C. acutatum* species complex are considered dominant in Greece. Despite efforts to control it, effective management remains elusive, necessitating the exploration of alternative strategies. This study aimed to evaluate the susceptibility/resistance of six Greek olive varieties to olive anthracnose disease. As part of the above objective, detached olive drupes at a comparable ripening stage from three Greek olive oil varieties (cv. Koroneiki, cv. Koutsourelia, cv. Mastoeidis) and three table olive varieties (cv. Gaidourelia, cv. Kalamon, cv. Konservolia) were artificially inoculated by a *C. acutatum* strain following two different inoculation methods: a) wound inoculation with fungal conidia suspension, and b) spraying of fungal conidia suspension without wound presence. Disease progression was monitored, and the susceptibility of each variety was assessed based on symptom development. As expected, fruits subjected to mechanical injury manifested accelerated symptomatology progression compared to those inoculated by the pathogen via spraying without associated wounding. Results showed that the Mastoeidis cultivar exhibited the highest resistance to anthracnose in both inoculation methods. Among the varieties with non-wounded drupes, the Kalamon and Gaidourelia cultivars exhibited the lowest disease progression rates, while the Koroneiki cultivar demonstrated the highest susceptibility to the disease. Regarding varieties subjected to pathogen inoculation without wounding, besides the Mastoeidis cultivar, the Koutsourelia cultivar displayed notable resistance profiles against the disease compared to the other cultivars under evaluation. These findings provide valuable insights into the resistance of Greek olive cultivars to olive anthracnose disease, which can be exploited to develop an integrated management system to control the disease in Greek orchards efficiently.

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P046

MOLECULAR MECHANISM OF RECEPTOR-LIKE KINASES IN RESPONSE TO WHEAT HIGH-TEMPERATURE RESISTANCE AGAINST PUCCINIA STRIIFORMIS F. SP. TRITICI

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Plants sense various pathogens and activate immunity responses through receptor-like kinases (RLKs), which are involved in massive signal transduction pathways upon perception of pathogens. Xiaoyan6 (XY6) is a wheat cultivar possessing non-race-specific and durable high-temperature all-stage (HTAS) resistance against *Puccinia striiformis* f. sp. *tritici* (Pst) since 1980s. The transcriptome analysis of wheat XY6 and Pst throughout the transition from compatibility to incompatibility under temperature regulation showed that numerous RLKs had significantly higher expression levels, indicating RLKs may play a key role. Three candidate RLKs including TaCRK10, TaXa21, and TaLecRK-IV.1 were identified to elucidate the molecular mechanism of high-temperature resistance of XY6 to Pst.

Upon Pst inoculation and high-temperature treatment, cell surface-localized cysteine-rich receptor-like kinases (CRKs) TaCRK10 interacts with and phosphorylates the histone variant Tah2A.1 in the membrane, then phosphorylated Tah2A.1 protein is transferred into the nucleus to activate the HTAS resistance of XY6 to Pst. In addition, TaCRK10 might directly induced plant defense responses through modulating the salicylic acid (SA) signaling pathway and reactive oxygen species (ROS) production. The leucine-rich repeat (LRR) RLK TaXa21 involved in HTAS resistance of XY6 through interacting with WRKY proteins. The extracellular signal is received by the LRR domain of TaXa21 with kinase domain phosphorylation and transmits to transcription factor TaWRKY62 through TaWRKY76, leading to the ET-mediated defense responses. The L-type lectin RLK TaLecRK-IV.1 positively regulated high-temperature resistance of XY6 to Pst and was not significantly associated with ROS production. In-depth exploration of the molecular mechanism of HTAS resistance of XY6 has a guiding significance for wheat breeding under the background of greenhouse effect.



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A REALLY INTERESTING NEW GENE (RING) IN WILD EMMER WHEAT ASSOCIATES WITH THE NITROGEN-CARBON BALANCE THAT AFFECT APHID FEEDING

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Wheat is a staple crop that suffers from severe economic yield losses worldwide by insect herbivore feeding. Cereal aphids are one of the major wheat pests, solitary feed from the plant phloem sap. They consume water and nutrients from the plant, thus depending on the host metabolism. To increase yield, it is imperative to identify new genes and alleles that confer resistance to herbivore attacks. The goal of our research was to identify markers associated with aphid fitness by exploiting the tetraploid wild emmer wheat (WEW; *Triticum turgidum* ssp. *dicoccoides*) germplasm, one of the progenitors of domesticated wheat, possesses untapped resources from its numerous desirable traits, including aphid resistance. Thus, we measured the chlorophyll level of 203 WEW accessions and conducted genome-wide association studies (GWAS). We identified a SNP that is associated with the alternative allele that possessed high chlorophyll and high aphid progeny relative to the wildtype allele. A metabolic profile indicated that several nitrogen-containing compounds were higher in the alternative allele accessions, while non-nitrogen compounds were lower in these accessions relative to the wildtype allele. This SNP is located 1,102 bp upstream to the terminator of a RING-type Zinc finger. To study the function of this gene, we ectopically expressed in tobacco leaves the RING gene from the sequenced WEW accession 'Zavitan,' which is associated with the wildtype allele. The RING overexpressed leaves had shown a reduction in aphid progeny and nitrogen-containing metabolites relative to the empty vector. We suggest that the RING is involved in determining the carbon-nitrogen balance of the wheat, which affects the aphid performance.

P048

UNIVERSITY OF CORDOBA-BALAM AGRICULTURE OLIVE BREEDING PROGRAM: DEVELOPING NEW CULTIVARS RESISTANT TO XYLELLA FASTIDIOSA

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Since the beginning of the *Xylella fastidiosa* epidemic in southern Italy, different levels of resistance were observed among different olive cultivars. Several studies have corroborated this variability and cultivars such as 'Leccino' and 'Fs-17' have shown high levels of resistance, while others such as 'Ogliariola Salentina' and 'Cellina di Nardo' have shown the opposite behavior. This variability has been the basis of the first olive breeding program to develop new cultivars

resistant to *X. fastidiosa*. The project started in 2017 and is being developed within the framework of collaboration between the University of Córdoba (UCO) and the company BALAM Agriculture.

To date, 44 preselections have been selected for their good agronomic characteristics, propagated and planted in different experimental fields in Spain and Italy. Currently, resistance to *Xylella fastidiosa* is being evaluated at field level in the area where the disease epidemic is developing, southern Italy.

P049

GENOME EDITING OF A DOMINANT RESISTANCE GENE FOR BROAD-SPECTRUM RESISTANCE TO BACTERIAL DISEASES IN RICE WITHOUT GROWTH PENALTY

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The bacterial pathogens *Xanthomonas oryzae* pv. *oryzae* (Xoo) and *Xanthomonas oryzae* pv. *oryzicola* (Xoc) have evolved a large family of transcriptional activation-like effectors (TALEs), which induce transcription of susceptibility (S) or/and resistance (R) genes in rice through direct binding to EBEs (effector binding elements) in the promoters, resulting in disease development or defense response. To endow the elite rice cultivar Nangeng 46 with broad-spectrum resistance against both Xoo and Xoc, we have engineered the defective promoter of the endogenous *xa23* gene through accurate insertion of a designed EBE_{Xoc/Xoo} array, which comprised 10 EBEs responding to distinct Xoo and Xoc TALEs, using CRISPR/Cas-9 mediated genome editing method. We discovered that each EBE retained its functionality and specificity in TALE-induced Xa23 expression and hypersensitive response (HR). The generated N46(Xa23R) line exhibited robust disease resistance to 50 Xoo and 30 Xoc strains from China, Philippines, and Japan. Therefore, promoter editing of *Xa23* alleles offers a promising way to rapidly upgrade commercial rice cultivars for broad-spectrum disease resistance.

P050

FORMULATION OF A STRAIN OF TRICHODERMA ASPERELLUM WITH CHITOSAN FOR PLANT GROWTH PROMOTION AND BIOCONTROL IN WHEAT AND RICE

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Trichoderma spp. are well-recognized for multiple positive interactions with plants, including growth promotion and defense against pathogens. This study focused on the selection of local strains for the development of a dry formulation. We evaluated fourteen strains. Plant growth promotion was evaluated in greenhouse trials using INIA Tacuarí rice cultivar and Génesis 4.33 wheat cultivar. For this, seeds were inoculated with a spore suspension of 1×10^6 conidia/mL (10 pots x two independent repetitions). Wheat and rice plants were harvested at 50 and 60 days after sowing and dried at 85° C until constant weight to obtain the

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dry weight of the aerial part. Biological control potential and synergism with chitosan was evaluated *in vitro* by the dual culture method on PDA plates, with and without chitosan supplementation, against rice pathogens *Nakataea oryzae* and *Rhizoctonia oryzae-sativae* and two isolates of wheat pathogen *Fusarium graminearum* (2 plates x 3 repetitions). For production and formulation of a dry preparation of selected *T. asperellum* ILB397 spores, production was carried out in rice-filled plastic bags, which were harvested with water after 12 days of growth. The spore suspension was supplemented with chitosan at 1.5 % as a protective polymer, and the suspension was spray-dried in a Bucher spray dryer at 70° C inlet temperature and 45° C outlet temperature. The collected powder had 3.2×10^9 sp/g (75 % viability). The formulated spore powder was vacuum packed and stored at 4° C and 20° C. Wheat seeds were inoculated with the rehydrated formulation at different concentrations. The intermediate concentration of 1 g/80ml (2.7×10^7 spores/ml) showed an initial priming effect on seed germination (36 % germinated seeds) versus water inoculated seeds (28 %) or unformulated fresh spores (32 %) at 7 days since sowing in greenhouse assays. Higher concentrations led to detrimental effects on seed germination. Adjustments of formulation and doses are underway.

P051

DEVELOPMENT OF STANDARDISED METHODS TO SCREEN THE EFFICACY OF NOVEL BIOLOGICAL CONTROL FUNGICIDES AGAINST PATHOGENS IN VIVO AND IN VITRO

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Crop protection is fundamental to food security and growers nowadays are under increased pressure to find effective ways of control. Plant disease control of fungal pathogens relies heavily on the use of chemical fungicides, and varietal resistance. However, repeated chemical fungicide use, can lead to issues with fungicide resistance development. Public concern on the use of fungicides and its impact on environment and health has also been on the rise. Taken together, there is an increasing interest in the use of nonchemical fungicides. One type of nonchemical fungicide is biological control which includes multiple product classes, such as microorganisms and natural plant extracts. Advantages of biological control include their multiple modes of action, which reduces the development of pathogen resistance to fungicide. Unfortunately, screening of biological control fungicides for their efficacy is time consuming and requires the need of multiple experimental methodologies. Here we present standardised *in vitro* and *in vivo* tests for screening biological control fungicides against multiple fungal pathogens. These standardised tests will help to speed up the development and availability of biological control fungicides. Eventually the standardised tests will also lead the way to fungicide resistance monitoring for biological control.

P052

EVALUATION OF THE IMPACT OF TRIAALFA ESSENCE APPLICATION ON BACILLUS AMYLOLIQUEFACIENS MBI600 (SERIFEL) PLANT COLONIZATION AND PLANT DEFENSE RESPONSES

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Bacillus amyloliquefaciens subsp. *plantarum* has been extensively used as a BCA agent against fungal and bacterial diseases and as a biofertilizer. Soil amendments, particularly organic amendments (OA), are shown to modify soil characteristics such as concentration of nutrients (e.g., P, K, Fe), pH, NO₃ content, organic material, and structure, resulting in the modification of the microbiota community and the enhanced disease control. The combination of those two tools as a strategy for integrated disease management gained the interest of many research groups that explored the use of OA-BCA combinations and their effects on plant growth, and/or on the soil microbial community structure, as well as their effectiveness against pathogens. This project aimed to provide a solid contribution to the impact of the OA, TriaAlfa Essence (trade mark Hechenbichler GmbH) on MBI600's Serifel (trade mark BASF SE) colonization and interactions with plant hosts and its ability to trigger plant defense responses. Efficient colonization of phyllosphere and fruit by MBI600 following combined Serifel/TriaAlfa application was confirmed on four economically important plant species by two different methods; the number of colony-forming units (cfu) and the newly developed qPCR method. The biocontrol efficacy of Serifel combined with TriaAlfa against *Erysiphe necator*, the causative agent of powdery mildew of *Vitis* species, was evaluated in field experiments. Results showed differences in the BCA's colonization among plant species and the two niches while the combinational treatment decreased disease frequency and severity at the early stages of disease development of the *E. necator*. In parallel, a mild effect on the tomato defense mechanisms was observed by TriaAlfa Essence and the combinational treatment with Serifel via the overexpression of *pr1b1* gene compared to control at 1- and 3-days post application. Our findings suggest that TriaAlfa and Serifel synergistically contribute to plant health maintenance and disease management through direct and indirect modes of action.

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SURFACTIN AND FENGYCIN CONTRIBUTE DIFFERENT FUNCTIONS TO THE BIOLOGICAL ACTIVITY OF *BACILLUS SUBTILIS* NCD-2 AGAINST COTTON VERTICILLIUM WILT

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Bacillus subtilis strain NCD-2 is a promising biocontrol agent for plant soil-borne diseases. A previous study indicated that strain NCD-2 could produce surfactin and fengycin simultaneously. In this work, the exact roles of surfactin and fengycin in strain NCD-2 against cotton verticillium wilt were determined.

In this study, the exact roles of surfactin and fengycin in strain NCD-2 against cotton verticillium wilt were determined by knockout of the surfactin synthetase gene *srfAA* and the fengycin synthetase gene *fenC*.

Compared to the wild-type strain NCD-2, the fengycin-deficient mutant Δ FenC decreased the antifungal ability toward *Verticillium dahliae*, but the surfactin-deficient mutant Δ SrfAA mainly decreased the swarming and biofilm formation abilities of strain NCD-2. Comparatively, the fengycin and surfactin double mutant Δ FenC/SrfAA decreased the antifungal activity and swarming and biofilm formation abilities to more significant levels. The colonization abilities of the cotton rhizosphere were compared between the wild-type (WT) strain NCD-2 and its derivative mutants using plate-counting methods. Compared to the wild-type strain NCD-2, Δ SrfAA and Δ FenC/SrfAA exhibited the worst colonization ability. RT-qPCR results confirmed that knockout of the *srfAA* and *fenC* genes decreased the expression of biofilm formation-related genes. The biocontrol abilities of the strains against cotton verticillium wilt were evaluated in a greenhouse. All mutants decreased the biocontrol abilities against cotton verticillium wilt, while Δ FenC/SrfAA showed the worst biocontrol ability, followed by Δ FenC.

Taken together, fengycin was the main antifungal active compound, and surfactin affected root colonization. Both the antifungal ability and root colonization of strain NCD-2 played important roles in suppressing cotton verticillium wilt.

In the present study, three bacterial strains have been tested as biocontrol agents against the fungal pathogen *Rhizoctonia solani*, a soilborne necrotrophic pathogen. Two of these strains (BO4A33 *Bacillus* sp. and BO4A42 *Psychrobacillus* sp.) were isolated from maize, while *Lactiplantobacillus plantarum* comes from olive. A *Trichoderma*-based commercial biocontrol product was used as control.

Rhizoctonia solani was inoculated into the soil two weeks before sowing. Treatments with *Trichoderma* and with bacterial strains were carried out by adding 10 mL of cell suspension (10^8 CFU/mL) to 100g of soil, at two points in time: one week before and on the same day of sowing.

The crop responses to treatments were automatically evaluated by using a multiple rgb cameras setup installed in the greenhouse. The germination fraction detected with a custom image-analysis algorithm was compared to the manual count of the seedlings.

After harvesting, several root-related parameters and the dry mass of 10 roots per replication were measured.

S61 and BO4A33 outperforming and equaling, respectively, the germination percentage observed in the control treatment with *Trichoderma*.

The automated sensing system enabled to quantify seed germination in the different treatments with high correlation ($R^2 = 0,9437$) with manual measurements.

Root-related parameters shown to be related to roots dry mass. Finally, the root system of plants treated with bacteria was more developed than when infected with rhizoctonia (Fig. 1).

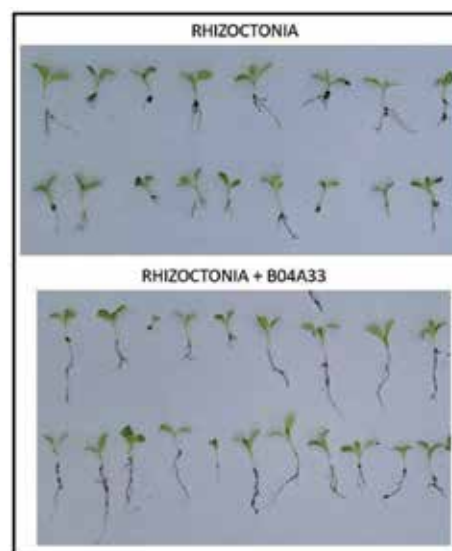


Fig. 1.

Bacteria used in this experiment have a biocontrol effect against rhizoctonia and increase lettuce germination and growth.

The use of cameras allowed to successfully acquire data regarding the germination and health conditions. These technologies could be helpful for automatization of lab evaluations and in-field assessments.

P054

NEW BIOCONTROL AGENTS AND AUTOMATED TOOLS TO EVALUATE IN VIVO EFFECT AGAINST *RHIZOCTONIA SOLANI*

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In recent years, interest has grown in the use of biocontrol agents as a viable alternative or support to synthetic chemicals for the management of phytopathogens and the enhancement of plant growth.

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EVALUATING THE EFFICACY OF POTENTIAL BIOCONTROL AGENTS FROM DISEASE-SUPPRESSIVE SOIL IN REDUCING GANODERMA BASAL STEM ROT INCIDENCE

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Ganoderma basal stem rot (BSR), caused by *Ganoderma boninense*, is the most lethal fungal disease affecting oil palms. Oil palms planted in disease-suppressive Blenheim soil (DS-BhS) (Typic Quartzipsamment) exhibited lower BSR incidence compared to those planted in disease-conducive Bernam soil (DC-BeS) (Typic Endoaquept). The soil microbiota has been observed to play a crucial role in disease suppression in DS-BhS. Hence, DS-BhS was selected as the medium for isolating biocontrol agents (BCAs). Amongst 21 microbial isolates (fungi, bacteria, and actinomycetes) isolated from DS-BhS, three microbes were shortlisted as potential biocontrol agents towards *G. boninense* based on a slide culture assay. Of these, *Trichoderma yunnanense* D6, *T. simmonsii* D19, and *Streptomyces abikoensis* M24 (isolated from DS-BhS) were discovered to have strong biocontrol activities against *Ganoderma boninense*. *Trichoderma yunnanense* and *S. abikoensis* induced cytoplasmic vacuolation and shrinkage, deformation of the mycelia, and lysis of mycelia of *G. boninense*. Additionally, *T. simmonsii* formed haustorial-like pegs, penetrating and establishing inter-cellular growth within mycelia of *G. boninense*. When applied as a consortium in nursery trials, a reduction of 4 to 55 % in BSR disease severity was observed. Furthermore, none of the three isolates were pathogenic to oil palm or induced growth setbacks. These findings revealed that the consortium of two *Trichoderma* and one *Streptomyces* derived from DS-BhS can be applied for managing BSR disease sustainably.

as a source of strains with potential use in the production of biopesticides.

A new fungal strain (isolate BPF3) characterized as belonging to *Epicoccum nigrum* (Pleosporales, Ascomycota), a well-known cosmopolite endophyte sourced from the flowers of *Bethencourtia palmensis* gave an organic extract with potent antifeedant activity against insect pests including *Myzus persicae*, *Rhopalosiphum padi*, *Spodoptera littoralis*, and certain fungal phytopathogens such as *Botrytis cinerea*. A bioactive compound responsible of most of the activity of the extract has been purified and identified as (E)-2-hydroxy-2,4-dimethyl-5-(prop-1-en-1-yl)furan-3(2H)-one.

This work will present the optimization of the production of this bioactive compound by the strain BPF3 (time-course fermentation and medium modification) and the study of the antifungal activity of its optimized extract against *B. cinerea*, *Fusarium oxysporum* and *Alternaria alternata*.

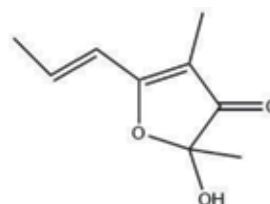


Fig. 1. (E)-2-hydroxy-2,4-dimethyl-5-(prop-1-en-1-yl)furan-3(2H)-one.

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BIOCONTROL POTENTIAL OF TRICHODERMA SPECIES AGAINST FOMITOPSIS MELIAE CAUSING CITRUS BROWN WOOD ROT DISEASE IN THE SOUTHWESTERN UNITED STATES

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Citrus brown wood rot, caused by *Fomitopsis meliae*, is a major cause of tree mortality in lemon orchards in the southwestern United States. *F. meliae* sporulates on abandoned wood debris on the orchard floor and spreads through windborne basidiospores and infects tree branches and the main stem through wounds. There are no products registered in the USA for the management of this disease. The objective of this study was to assess the biocontrol potential of *Trichoderma* species recovered from rhizosphere soil samples in Yuma lemon orchards against *F. meliae*. A total of 48 isolates of *Trichoderma* were evaluated under laboratory conditions against *F. meliae* using dual culture assay. The results indicated that 24 isolates could inhibit the mycelial growth of *F. meliae* significantly. Among them, four isolates (i.e. 1T1, 1T24, 1T40, 1T42) showed the highest antagonistic activity with 81.5 - 87.9 % inhibition. Cell-free metabolite extract and volatile metabolites of these 4 effective isolates also inhibited the growth of *F. meliae* in the culture medium. Under greenhouse conditions, these 4 isolates were effective in protecting wounds of tree branches from infection by *F.*

P058

BIOACTIVE COMPOUND FROM EPICOCCUM NIGRUM STRAIN BPF3, AN ENDOPHYTE ISOLATED FROM THE FLOWERS OF BETHENCOURTIA PALMENSIS

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Islands are recognized as hubs for numerous endemic plants, uniquely confined to specific regions, serving as natural reservoirs for undiscovered microorganisms with potential medical and agricultural uses. Endophytic fungal microbiome from *Bethencourtia palmensis* has been shown in the past

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meliae. In summary, effective protection of branch wounds from infection by *F. meliae* may be achieved with wound treatment using the *Trichoderma* isolates.

P058

IN VITRO AND IN VIVO EVALUATION OF ENDOPHYTIC TRICHODERMA AS POTENTIAL BIOCONTROL AGAINST OIL PALM FUNGAL PATHOGENS

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Oil palm diseases, specifically Basal Stem Rot and Leaf Spot, have been associated with soil-borne pathogen *Ganoderma boninense* and foliar pathogen *Pestalotiopsis* sp., respectively. *G. boninense* results in palm fatality, whereas *Pestalotiopsis* sp. can lead to yield reduction. *Trichoderma* spp. has emerged as promising biocontrol agents due to their competitive nature. In this study, we effectively isolated and identified 104 isolates of *Trichoderma* spp. from soil, leaves, and oil palm roots. These were narrowed down to 10 isolates based on antagonistic characteristics, benchmarked against commercially available strains registered for oil palm. A dual culture test was used to examine the isolates, and the results showed that all 10 isolates inhibited the growth of *G. boninense*, *P. microspora*, *P. clavispora*, and *P. virgulata* with efficacy ranging from 72-87 % and 63-87 %, respectively. Two highly antagonistic strains, *Trichoderma harzianum* (LRT F35) and *Trichoderma asperellum* (VOP F13), were introduced into oil palm seedlings via seed soaking, foliar spraying, and soil drenching. Both isolates colonised seedlings endophytically via a single application round. The VOP F13 strain increased growth by 10 %. The fungal abundance in *Trichoderma*-treated and control plants was measured using the Wheat Germ Agglutinin (WAC) assay, which revealed the presence of 24 % chitin (in RFU) in treated plants. *T. harzianum* and *T. asperellum* were successfully differentiated using high-resolution melting-qPCR analysis. Scanning Electron Microscopy (SEM) study revealed sporulating hyphae in *Trichoderma*-treated leaves. During the three-month trial, no disease symptoms were observed on the leaves. Our *Trichoderma* isolates have proven successful inhibition against oil palm pathogens and colonisation in oil palm. This study underscores the importance of crop-specific biocontrol *Trichoderma* strains that establishes itself quickly with a single application, reducing costs and promoting environmentally friendly practices. Such strategies are pivotal in advancing the green revolution and fostering the transition away from chemical control methods.

P059

BACILLUS SPP. ISOLATED FROM SOLARIZED FIELDS PROMOTE LETTUCE PLANTS' GROWTH AND INDUCE SYSTEMIC RESISTANCE AGAINST THE SOILBORNE PATHOGEN RHIZOCTONIA SOLANI

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Lettuce is cultivated intensively throughout the year, in successive cycles in the same field usually without rotation and thus being susceptible to foliar and soilborne pathogens. The development of high pathogen populations makes necessary the implementation of various control methods. Certain bacteria, known as plant growth-promoting rhizobacteria (PGPR), have already been reported to be beneficial for plants, promoting plant growth and suppressing disease development. In this study, several bacterial strains, belonging to the genus *Bacillus* were isolated from the rhizosphere of lettuce plants cultivated in a field in which solarization was implemented. These strains have been investigated for their effect on the growth of lettuce plants and their biocontrol efficacy against the soilborne pathogen *Rhizoctonia solani*. The application of these bacteria on lettuce plants revealed that the tested strains resulted in significant increase in leaf and root weight, although in variable levels. Apart from the impact of these bacterial strains on plant growth characteristics, the expression levels of auxin-related genes of lettuce plants have been investigated. In addition, single applications of these strains on inoculated lettuce plants with *R. solani* resulted in sufficient control efficacy in comparison with the chemical treatment. Expression analysis of defense-related genes was carried out at different time points on lettuce plants treated with the four most effective bacterial strains and artificially inoculated with the pathogen. In general, a higher induction rate was observed for all the tested genes in plants treated with the bacteria, implying induction of systemic resistance. The isolation of indigenous bacteria from solarized soils might lead to a selection of beneficial plant growth-promoting rhizobacteria, which enhance plant growth and suppress disease development of a given cropping system.

P060

INVESTIGATING ANTIVIRAL ACTIVITIES OF IODINE-CONJUGATED ORGANIC AGENTS AGAINST MAJOR VIRUSES INFECTING PEPPER

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Plant viruses cause economic losses by reducing the quantity and quality of major crops. This issue is a growing concern due to the expansion of global trade and climate change. In addition, the emergence of new pathogen strains increases the difficulty of controlling viral diseases. Effective management strategies are therefore needed. The control strategy for

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viral diseases relies primarily on non-chemical and cultural practices, as no commercial viricides are currently available. Some compounds have been identified as effective against certain viruses, but their use in the field is limited due to issues such as concentration, toxicity, and efficacy. Therefore, it is imperative to discover novel antiviral agents that address the existing challenges associated with the identified antiviral candidate compounds. In this study, we evaluated iodine-conjugated organic compounds mixed with sialic acid, whey, and blood meal for virus disease management against seven viruses that cause significant yield losses and economic damage to plants. The candidate compounds reduced virus accumulation and symptom development. Treatment with candidate compounds, A4 and A5, reduced viral RNA accumulation to about half that of those in the control group and showed reduced symptoms along with healthier growth. In addition, we performed transcriptome analysis of treatment with two viruses, which suggested that the mechanism of viral RNA replication inhibition might relate to plant defense systems based on phytohormone pathways. This study demonstrated that treatments with naturally derived materials, such as iodine, nitrogen, and sialic acid-conjugated organic substances, may directly or indirectly impact the host plant's resistance to various virus infections. Moreover, our findings suggest that these natural candidate materials could be utilized for managing virus diseases in the field.

P061

SUSTAINABLE CONTROL OF WHITE ROT ON RED ONION "CIPOLLA ROSSA DI TROPEA"

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In Calabria (Southern Italy) the white rot severely affects the red onion "Cipolla Rossa di Tropea" a certified product playing an important role in local economy. The causal agent, *Sclerotium cepivorum*, produces overwintering sclerotia which can remain alive in soil for many years. Field trials were conducted to evaluate the efficacy of solarization and bio-solarization in controlling the disease. Soils were covered with transparent plastic sheets for 53 days starting from August 3, 2023, and litter of mushroom cultivation was used for the bio-solarization. The analysis of soil samples revealed a significant impact of treatments on the vitality of sclerotia and on the soil microbiome. Treated and untreated parcels were utilized to seed a commercial onion cultivar (November 30, 2023) and the incidence and gravity of the disease was evaluated at the harvesting time (end of February 2024). Solarization and bio-solarization proved very effective reducing the incidence of infected plants by 62 and 73 % in solarized and bio-solarized parcel, respectively (Fig. 1). Furthermore, the average weight of plants was 45.7, 43.9, and 20.8 g in solarized, bio-solarized, and untreated parcels, respectively.

In parallel to field trials several bacteria showing a strong inhibitory activity against *S. cepivorum* were isolated from sclerotia of naturally infected onions. In dual culture essays

the diameter of the fungal colonies was reduced up to 50 % (Fig. 2). Interestingly, the inhibitory activity of bacteria was influenced by the strains of the pathogen, being generally stronger against the strain from which they were isolated. These preliminary results suggest the coevolution of pathogens and antagonists and support the use of sclerotia as a source of effective biocontrol agents. The combination of solarization and/or bio-solarization with antagonistic bacteria isolated from sclerotia may be strategic to develop effective and sustainable control methods against the white rot of onion.



Fig. 1. Representative red onion "Cipolla Rossa di Tropea" from untreated (left), solarized (middle) and bio-solarized (right) parcels.



Fig. 2. Inhibition halos produced by bacteria isolated from sclerotia of *S. cepivorum*.

P062

IN VITRO EFFICACY OF SOME PLANT EXTRACTS ON THE INHIBITION OF PECTOBACTERIUM CAROTOVORUM, THE CAUSATIVE AGENT OF BACTERIAL SOFT ROT

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Pectobacterium carotovorum, is one of the most important and widespread bacterial pathogens of a variety of plant products in storage. The aim of the present study was to evaluate the efficacy of some plant extracts on *Pectobacterium carotovorum* in the laboratory conditions. For this purpose, flower, leaf and seed hydroethanolic extracts of cloves (*Syzygium aromaticum*), savory (*Satureja hortensis*) and fennel (*Foeniculum vulgare*) respectively were prepared. Overnight culture of *Pectobacterium carotovorum* (PTCC 1675) in nutrient agar was cultured into nutrient broth and standardized with 0.5 McFarlands. The minimum inhibitory

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concentrations of the plant extracts were determined by a serial two-fold dilution method in 96- well plates. The plates were incubated at 25°C for 48 h with the lid on. The wells were then examined for evidence of growth and MIC values were determined as the lowest antimicrobial concentration that inhibited visible growth of the test microorganism. Experiment was repeated two times. MIC value for the fennel and savory extracts was 1/32 while the value for clove extract was 1/16; hence, according to our results, fennel and savory showed stronger antimicrobial activity against *Pectobacterium carotovorum* as comparison with clove. These results in overall are promising to use some plant extracts as potential botanical tools to control microorganisms in plant and their products.

P063

HARNESSING THE AVOCADO MICROBIOME AS A SOURCE OF BACTERIA FOR THE BIOCONTROL OF AVOCADO WHITE ROOT ROT

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Plants associate with a complex array of microorganisms in both the rhizosphere (soil surrounding the root and influenced by its exudates) and endosphere compartments (within the root). These microorganisms in close contact with the plant can significantly impact plant health and development.

In this study, we aim to investigate the microbial biodiversity associated with avocado trees affected by the white root rot caused by the soilborne fungal pathogen *Rosellinia necatrix*, as well as to identify key microorganisms involved in suppressing this disease. To do so, we conducted a comprehensive survey utilizing 16S rRNA sequencing, to identify those microorganisms associated with healthy avocado trees. Differential abundance analysis was used to investigate the bacterial taxa associated with healthy and diseased avocado trees. The analysis revealed the enrichment of bacteria, primarily from the genus *Variovorax*, in healthy trees. Despite the extensive research on *Variovorax* in biotransformation, biosynthesis, and bioremediation within ecological contexts, its potential in the biological control of fungal diseases remains relatively unexplored.

This study highlights that the avocado root microbiome is a rich source of bacteria with strong antagonism against fungal pathogens of high economic relevance. These bacteria could be used as effective biological control agents in agriculture, offering a more sustainable approach to combating fungal diseases.

P064

EFFICACY OF THREE ESSENTIAL OILS AGAINST CAUSAL AGENTS OF GREY MOULD, BOTRYOSPHAERIA AND EUTYPA DIEBACK

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Growing concerns regarding overuse of agrochemicals and their detrimental impact on the environment have increased the demand for new "green" tools for plant protection. In the past few years, essential oils (EOs) have gained particular attention as compounds with antimicrobial and insecticide properties. This work aims to investigate the antifungal effect of three EOs (cinnamon, clove and basil) against *Botrytis cinerea*, *Diplodia seriata* and *Eutypa lata* (causal agents of grey mould, Botryosphaeria and Eutypa dieback, respectively, in grapevine) through *in vitro* screening tests. Mycelial plugs (5 mm Ø) were placed on potato dextrose agar (PDA) amended with three rates for each oil (0.025, 0.05 and 0.1 % v/v). Mycelial growth was measured when the control colony (inoculated in water amended PDA) reached the edge of Petri dishes. *Botrytis cinerea* mycelium was reduced by 0.05 % of cinnamon oil (-88 %, in comparison to control), as well as 0.1 % of both clove and basil EOs (-79 and -29 %, respectively). In contrast, complete growth inhibition of *D. seriata* was achieved at 0.05 % of cinnamon and clove EOs amended PDA. On the other hand, full control of *D. seriata* was not observed at any of the basil EO concentrations tested. However, a limited growth reduction (-2 %) at 0.1 % indicated that higher rates could potentially be tested for greater control. For *E. lata*, different rates were required for each oil: a completely inhibited mycelial growth was observed in 0.025 % cinnamon EO, and 0.05 % clove EO amended PDA, implying that lower concentrations could be effective as well; while 0.1 % basil EO amended media had a partial antifungal effect (-16 %). Based on these results, all EOs showed potential for control of mycelial growth of all pathogens. Further trials for basil and cinnamon oils are needed to determine the optimal concentrations along with *in vivo* applications.

P065

THE BIOLOGICAL FORMULATION SERIFEL® (BACILLUS AMYLOLIQUEFACIENS STRAIN MBI600) CONTROLS COLLETOTRICHUM ACUTATUM IN FLOWERS AND INDUCES DEFENSE RESPONSES IN OLIVE TREES

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Anthraxnose is one of the most devastating fungal diseases affecting olive trees, leading to severe losses and deterioration of olive oil quality. Biological control agents offer environmentally friendly disease control, minimizing harm to non-target organisms and promoting sustainable

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practices. The present study aimed to investigate the action of the biological agent Serifel® (BASF SE) against *Colletotrichum acutatum* on olive flowers and its potential to induce defense responses in young olive trees. To visualize the infection process on olive flowers, the greek *C. acutatum* reference strain was successfully transformed with the GFP (Green Fluorescent Protein) gene, exhibiting high fluorescence in spores and hyphae. Transformation and fluorescence intensity was stable after 5 serial transfers on medium without antibiotic. Moreover, the gfp transformant exhibited the same infectivity as the wild type on detached olive fruits. In the flower pathogenicity assay, significantly reduced infection by the pathogen was observed by confocal microscopy in flowers that had been sprayed with the biological agent in advance. To assess defense responses induced by Serifel®, young trees, under stable conditions of temperature, humidity and irrigation were sprayed with the biological agent. Two days later, they were inoculated with *C. acutatum* spore suspension. It was determined that Serifel induced the expression of olive genes *BGlu* and *Mpoel* compared to mock plants both in the pathogen's presence and absence. Concluding the present study provides strong evidence for an important role of the biological formulation Serifel® to control *C. acutatum* in olive flowers and induce defense responses in olive trees.

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P066

ROOTING FOR BACILLUS SPP.: CONTROL OF PHYTOPHTHORA ROOT ROT IN HYDROPONIC LETTUCE CULTIVATION

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In Belgium and neighboring countries, the introduction of the soil-borne oomycete *Phytophthora cryptogea* into hydroponic lettuce cultivation has resulted in significant economic losses. The pathogen infects plant roots by releasing zoospores into the recirculation water, causing slimy root decay and diminishing lettuce crop profitability. Current control methods rely on chemical plant protection products, which can potentially harm humans and the environment. Hence, alternative strategies, such as implementing biological control organisms (BCOs), are necessary. In this study, the biocontrol capacity of different *Bacillus* spp. was evaluated to control *Phytophthora* root rot in hydroponically cultivated lettuce. Additionally, mutants of *Bacillus velezensis* GA1 were utilized to investigate the antimicrobial activity of its specialized metabolites. *In vitro* dual culture assays demonstrated

that BCO treatments, including various *B. velezensis* and *B. siamensis* strains, reduced mycelial development of *P. cryptogea* by at least 70 % and inhibited cyst germination. *In planta* assays confirmed the efficacy of *B. velezensis* GA1, along with its specialized metabolites, in reducing *P. cryptogea* infection in hydroponically cultivated lettuce roots when applied at a concentration of 10⁷ CFU/mL, one day before *P. cryptogea* zoospore inoculation (0.1 zoospores/mL). Moreover, changes in lettuce root architecture were observed upon application of *B. velezensis* GA1 and other *Bacillus* spp. In parallel, *in vitro* assays with mutants of *B. velezensis* GA1 deficient in bacilysin production showed a significant loss of biocontrol activity compared to the GA1 wild type. The successful application of *Bacillus* spp. and their specialized metabolites in hydroponic lettuce cultivations demonstrates their potential as sustainable alternatives, offering hope for mitigating economic losses and promoting environmentally friendly agricultural practices.

P067

AGRICULTURAL COMPOSTED PRODUCTS AS PLANT PROTECTION AND DEFENSE INDUCERS IN TOMATO

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Composts are natural products usually produced from crop residues and contribute to plant nutrition by enriching the soil with organic matter and nutrients. Additionally, they contain high microbial populations containing non-pathogenic fungi, yeasts, and bacteria that can potentially act as biocontrol agents and induce the plant defense system. The aim of this study was to assess the biopotential of two composted products (Ikorganic), based on 1) onion crop residues and green prunings and b) onion crop residues, mushroom litter substrates and green prunings. The evaluation of the two composts was based on their efficacy to reduce the disease severity against plant pathogens in greenhouse studies, as well as to induce the plant defense system. Composted products' efficiency in promoting plant growth and reducing diseases' severity was evaluated via root of foliar application of the two composted products, followed by artificial inoculation of tomato plants with the soilborne pathogen *Verticillium dahliae* or the phytopathogenic bacteria *Pseudomonas syringae* pv. *tomato*, under greenhouse conditions. Subsequently, the biomass of total plants and fruits for treated plants, mock and control plants were calculated. Additionally, total RNA was extracted from tomato leaves and the expression levels of five tomato defense genes, in various time points post pathogens inoculation, were defined using RT-qPCR method. Elevated levels of expression of *PR1b*, *PIN2*, *PAL*, *PR3* and *LOX* genes were detected on tomato leaves, treated with the composts, especially in plants that were artificially inoculated with *Pseudomonas syringae* pv. *tomato* post composts' application. The study's findings from greenhouse experiments and Real Time PCR, will be presented.

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P068

EFFECT OF BACILLUS BIOLOGICAL CONTROL STRAINS AGAINST CUCUMBER PLANT PATHOGENS *PODOSPHAERA XANTHII* AND *PSEUDOPERONOSPORA CUBENSIS* IN VIVO AND IN PLANTA

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Cucumber (*Cucumis sativus* L.) is an important crop distributed worldwide. Powdery mildew (PM), mainly caused by the fungus *Podosphaera xanthii* and downy mildew (DM) caused by the oomycete *Pseudoperonospora cubensis* are major pathogens in cucurbit crops. Large amounts of chemicals are used annually for their control with the known side effects. In this study, three bacillus endophytic strains Cal.r.19 (*B. subtilis*), Cal.r.29 (*B. velezensis*) and Cal.l.30 (*B. halotolerans*) with plant growth promoting activity and strong biological control activity against soil borne pathogens, isolated from the medicinal plant *Calendula officinalis*, were tested *in vitro* and *in planta* for their potential to control PM and DM in cucumbers. *In vivo* bioassays were performed to evaluate Bacillus strains effectiveness against both PM and DM. Their phytotoxicity and efficacy was also tested in greenhouse experiments in potted cucumber plants. Foliar sprays of spore/cell suspensions at various concentrations were carried out with and without the addition of an adhesive agent in order to determine the maximum application concentrations without any side phytotoxic effect. Cal.l.30 showed an overall efficacy of > 90% against PM after 3 weekly applications, in artificially inoculated plants. Cal.r.29 showed an efficacy of ca 40 % against DM in cucumbers, 11 days post inoculation. Since data on biological control of obligate pathogens is limited, our results are encouraging and the effectiveness of these Bacillus beneficial strains against powdery and downy mildews in cucumber plants support the significant role of endophytic bacteria in plant disease management.

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P069

EVALUATION OF BIOLOGICAL CONTROL EFFICACY OF BACILLUS STRAINS AGAINST FUSARIUM STEM AND ROOT ROT OF CUCUMBER UNDER GREENHOUSE CONDITIONS

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Cucumber (*Cucumis sativus* L.) is an important crop cultivated in the Mediterranean Basin and worldwide. One of the most destructive diseases especially when the temperatures in the greenhouses are around 18°C is caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum* (FORC). The pathogen causes stem and root rot in cucumber plants. It is widely known that the few chemical agents available have limited or no effect along with the known undesirable consequences. In this study, two endophytic strains Cal.f.4 and Cal.l.30, both belonging to the *Bacillus halotolerans* group, with plant growth promoting activity and strong biological control effect against soilborn pathogens, isolated from the medicinal plant *Calendula officinalis*, were used for greenhouse experiments for an eco-friendly disease control in two different commercial varieties of cucumber plants. Cucumber plants of both varieties (SS1 and SB2) with four true leaves were bacterized with foliar spraying of suspensions of *B. halotolerans* strains Cal.f.4 and Cal.l.30 as a single-strain application and as a mixture (1:1) under greenhouse conditions with a completely randomized block design. FORC application was done by root drenching of a conidial suspension ($10^6 \times 1$ conidia/ml). Additional bacterial applications were applied by foliar sprays every 10 days approximately for 30 dpi. Plants were observed during three months' period and disease symptoms, plant height and number of leaves were recorded. Results indicated that disease symptoms appeared much later in all plots with bacterial applications, compared to the control plants that showed collapse of stems and subsequently plants died. At the end of the experiment (three months after pathogen inoculations) biocontrol applications showed about 67% protection against FORC, for both bacterial strains applied either separately or in mixture. Variety cucumber SB2 showed moderate resistance against FORC compared to SS1 which appeared to be more susceptible.

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Trichoderma SPECIES CONTROLLING POTATO SCAB IN BRAZIL

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Potato scab, caused by *Streptomyces* spp., represents a significant global challenge impacting the quality and yield of potatoes. In Brazil, this disease has led to substantial economic losses. This study addresses the need for effective and sustainable control methods, focusing on the biocontrol potential using *Trichoderma* spp. *T. conigiopsis* LBF 11/48 and *T. asperuloides* LBF 04/04. The experiments comprised *in vitro* evaluations, greenhouse and field assays. Both *Trichoderma* isolates exhibited antagonistic behaviour, inhibiting the growth of six phytopathogenic *Streptomyces* species (*S. scabiei*, *S. europaeiscabiei*, *S. cavigabiei*, *S. turgidicabiei*, *S. reticuliscabiei* and *S. brasiliiscabiei*) in the *in vitro* tests. Greenhouse experiments demonstrated a significant biocontrol potential against *S. scabiei* IBSBF 2950 with *T. asperuloides* LBF 04/04 reducing 53.6 % of disease incidence and 30.5 % of severity compared to the positive control. *Trichoderma conigiopsis* LBF 11/48 and Ecotrich® (a commercial product based on *T. harzianum* used for comparison) also demonstrated effectiveness, albeit with slightly lower rates of inhibition. *Trichoderma conigiopsis* LBF 11/48 showed a 34.2 % reduction in incidence and an 18.3 % decrease in severity, while Ecotrich® exhibited a 39.7 % reduction in incidence and a 30.5 % decrease in severity. *Trichoderma conigiopsis* LBF 11/48 and the commercial Ecotrich® product also exhibited efficacy but with lower values of inhibition XX, XX and XX,XX. Additionally, growth promotion assessments showed the positive impact of *Trichoderma* isolates on fresh mass and tuber production. *T. conigiopsis* LBF 11/48 exhibited the highest rate in the production of tubers, showing an increase of 23.9 % compared to the untreated control and 46.1 % compared to the treatment with the pathogen. Field assays confirmed the biocontrol effectiveness of *Trichoderma* isolates, resulting in reductions in disease incidence ranging from 15-8 %. However, no statistically significant differences were observed in severity, underscoring the complexity of these interactions. *T. conigiopsis* LBF 11/48 and *T. asperuloides* LBF 04/04 demonstrated notable potential in controlling potato scab emphasizing the importance of biological control agents for sustainable diseases management and plant growth.

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The spongy moth, *Lymantria dispar* L. (Lepidoptera: Erebiidae) is a pest of deciduous forests, responsible for severe defoliation. Despite its important pest status, few studies have dealt with the wide-ranging surveillance of the adult male population utilizing different pheromone traps. The current study aimed to carry out an extensive comparison of different trapping devices for the male adult of *L. dispar*, which was run in parallel in different areas of 5 European countries, for two consecutive years, i.e. 2022 and 2023.

Therefore, the effect of trap type on the catches of male adults of *L. dispar* was examined in 18 sites in Europe, two in Slovenia (Ginjevec 1 and 2), two in Spain (Montnegre – Corredor and Les Gavarres), twelve in Greece (Pindos, Drama, Petralona 1 and 2, Kouri, Portaria, Lykodromio, Stavroupoli, Kryoneri cemetery, Kryoneri 1 and 2, Saint Nikolaos), one in Hungary (Telki), and one in Croatia (Koška). Seven different trap types, G trap and eGymer 6 - 1, were examined through 2022 and 2023 from June to September.

The male adult captures in most of the areas examined started at the end of June and lasted until the middle of August. Trap type was found to be significant for most of the sites tested. Among the trap types evaluated, the G trap was found superior to the other traps, in many experimental sites when the numbers of *L. dispar* adults were high, while in Pindos no differences in trap types were noticed. Captures of *L. dispar* in pairs of various trap types showed a positive and significant association in the majority of investigated combinations, suggesting that most of them exhibited comparable population fluctuations.

It is recommended by our findings that the G trap should be used to monitor the adult male population of *L. dispar*.

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P071

INFLUENCE OF TRAP TYPE ON THE CAPTURES OF LYMANTRIA DISPAR L. (LEPIDOPTERA: EREBIDAE): TRIALS FROM DIFFERENT EUROPEAN COUNTRIES

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P072

POTENTIAL OF PHEROMONE AND YELLOW STICK TRAPS FOR MANAGEMENT TUTA ABSOLUTA IN FIELD AND GREENHOUSES

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In the Georgia one of the important axis of development of agriculture is vegetable production. Currently, this sector

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suffers from several phytosanitary problems such pest insects.

The South American tomato moth, *Tuta absoluta* (Povolny) (Lepidoptera: Gelechiidae) an invasive pest, cultivated and wild plants from *Solanaceae* family, which i introduced in Georgia from Turkey in 2011. Insect is one of the most economically important pests of tomato and is posing a serious threat to the fruits production across the Mediterranean and Black Sea regions. In recent years the rapid growth of *T. absoluta* population has caused the significant damage of tomato in Georgia. During vegetation season *T. absoluta* can develop 12-10 generations and consequently necessary to develop control measures. The aim of this work is to monitoring *T. absoluta*, using Pheromone traps and Color Sticky Traps for the established level of pest in the field and greenhouse farms. The studies were carried out in Shida Kartli (600-550 msl) and Akhaltsikhe (1100-1000 msl) Region of the Western Georgia, 2022-2020. These two regions are different geographically and climatically and we to assume that the biological particularity of this insect are also different. Pheromone TUA-Optima® for trapping of *Tuta absoluta* we get from Russell IPM and installed with Delta traps and yellow traps separately. The high capture rate observed in field of Shida Kartli, with these pheromones at the density of 1 trap/500m² in average 184 adults, (min=70, max=460), in Akhaltsike, 36 adults (min=3, max=65). in average 442 adults/trap were caught in the greenhouse with 1 trap/350m² of Shida Kartli (min=142, max=560), and 312 adults/trap (min=132, max=538) were observed in Akhaltsike. Yellow sticky traps 1 traps/50 m² show significant high results, where in field condition attractive adults in average 318 (min=25 max=549) and in greenhouses 356 (min=37 max=581) were observed.

P073

EVALUATION OF FUNGICIDES FOR THE CONTROL OF SEED-BORNE BACTERIAL PATHOGENS

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Seed-borne bacteria impact plant health and yield, and bacterial-contaminated seed is a source for the transboundary spread of pathogens. Due to a lack of effective seed-dressing agents, eliminating seed-borne bacteria involves the hot-water treatment, which is laborious, sensitive to temperature settings, and known to affect seed viability. This study evaluated mancozeb and carbendazim for their bactericidal properties by testing against eight seed-borne bacterial species (*Bacillus cereus*, *Bacillus subtilis*, *Ralstonia solanacearum*, *Xanthomonas manihotis*, *Pantoea stewartii*, *Xanthomonas glycinea*, *Xanthomonas oryzae* and *Xanthomonas vignicola*). in three crops (cowpea, maize, and soybean). Seed dressing with mancozeb resulted in 100 % suppression of seed-borne bacteria and serves as the broad-spectrum reagent for seed dressing to eliminate seed-borne bacterial infection. Mancozeb is widely available, and doubles as a reagent for preventing seed-borne and bacterial contamination. To our knowledge, this is the first study to validate the effectiveness of mancozeb to control seed-borne bacterial contamination.

P074

BIOLOGY, STATUS, AND MANAGEMENT OF FALL ARMYWORM IN THE PHILIPPINES: A SYSTEMATIC REVIEW

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The introduction of fall armyworm (FAW) *Spodoptera frugiperda*, a type of transboundary plant pest and disease (TPPD) in the Philippines has caused significant threats to the agricultural sector. This study provides a qualitative systematic review of the biology, incidence status, and pest management of FAW in the country using the PRISMA 2020 guidelines. Thirteen (13) unique peer-reviewed journal articles from 2018 to 2023 were selected from electronic searching databases and expanded search and were classified through three phases of a filtering process involving identification, screening, and eligibility assessment. FAW is an understudied topic in the Philippines, with the majority of research in Luzon, and little to no published studies in Visayas and Mindanao. Although there is adequate morphological information, there is only one report on the molecular identification of *S. frugiperda* strains. Protogyny is observed in fall armyworms, deviating from the common sexual bimaturation (SBM) in lepidopterans which could serve as a fitness-enhancing strategy yet further exploration needs to be conducted. The primary host preference of fall armyworm is corn (*Zea mays*), although with a significant aversion to Bt corns. Ecological adaptability and a broad range of host utilization were evident, suggesting the presence of refuge host species like common weeds surrounding cropping areas. Pest management mainly focuses on the utilization of entomopathogenic fungi such as *Metarhizium sp.* (*Metarhizium rileyi* and *Metarhizium anisopliae*) and *Beauveria sp.* (*Beauveria bassiana*), biochemical compounds such as wettable powders (kaolin clay), and Bt corn hybrid accession Bt Cry1Ab. However, organic pesticides, Bt, and insecticide resistance are yet to be explored. The high homogeneity of data and potential publication bias are accounted for since the majority of the studies have the same collaborating authors. There is a need for more studies across the country to achieve a comprehensive assessment.

P075

GENETIC DIVERSITY, DISTRIBUTION, AND STRUCTURE OF WHITEFLY BEMISIA TABACI SPECIES ON CASSAVA IN NORTHERN DEMOCRATIC REPUBLIC OF CONGO

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Cassava Mosaic Disease and Cassava Brown Streak Disease (CBSD) are two viral diseases that threat cassava production in East and Central African countries. These diseases are transmitted by vector, the whitefly *Bemisia tabaci* or by exchanges of cuttings. *Bemisia tabaci* is a complex of more than 40 morphologically undistinguishable species identified in the world. This study aims to i) identify the different species that colonise cassava in the north of the Democratic Republic of Congo (DRC), ii) study their genetic diversity, and iii) see how this diversity might be structured in space or suffer from invasion from neighbouring countries (East) where CBSD has a high prevalence. To do so, a large sampling was done in 43 sites from East to West DRC. Then, both nuclear and mitochondrial markers were used to identify the species and study the genetic diversity of the collected populations. Two *B. tabaci* species were found: SSA1 and SSA3. In the surveyed provinces, SSA1 was the main species (95.34 %), structured into two genetic clusters along the east-west transect, and SSA3 was restricted in western provinces. Our results confirm SSA1 as the most abundant and adapted species on cassava throughout Sub-Saharan Africa and may be responsible for the spread of cassava viral diseases in DRC.

P076

EFFECT OF FEEDING TIMES OF BEMISIA TABACI ON TOSRV AND TOCV-INFECTED TOMATOES SPRAYED WITH INSECTICIDE ON VIRUS ACQUISITION AND TRANSMISSION

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Tomato golden mosaic and tomato yellowing diseases, caused by the begomovirus tomato severe rugose virus (ToSRV) and de crinivirus tomato chlorosis virus (ToCV), respectively, are the most important diseases affecting tomato (*Solanum lycopersicum*) crops in Brazil. Both viruses are efficiently transmitted by *Bemisia tabaci* MEAM1 in semipersistent and persistent circulative manners, respectively. This work aimed to evaluate the effect of different feeding times of *B. tabaci* in mixed-infected tomato plants sprayed with two different doses of the insecticide cyantraniliprole (0.5 ml/L and 1.25 ml/L) on virus acquisition and transmission and insect mortality. For this, 50 adults of *B. tabaci* MEAM1 were confined to infected tomato plants 48 h after insecticide spraying for 24, 8, 4, 2, and 48 h virus acquisition access periods (AAP). Plants were inside insect proof-cages. After each virus AAP, the infected tomato plant was eliminated, and five healthy tomato plants were introduced into the cage for a virus inoculation access period (IAP) of 48 h, when the insects were eliminated. ToSRV and ToCV infection rates were evaluated 30 days later by PCR and RT-PCR, respectively. Insect mortality was evaluated by confining 30 adults of *B. tabaci* MEAM1 to infected tomato plants 48 h after insecticide spraying, inside individual cages, counting dead insects at 24, 48, 72, and 96 h after confinement. The experiments were performed three times for each insecticide dose. Neither

of the two doses of the insecticide cyantraniliprole efficiently controlled the primary transmission of ToSRV and ToCV by *B. tabaci* MEAM1. Insect mortality was low in the first hours of exposure, reaching almost 100 % only after 72 h. This time is enough for some viruliferous insects to transmit both viruses, suggesting that chemical control of the vector, widely used by producers, is not efficient in controlling the primary transmission of these viruses.

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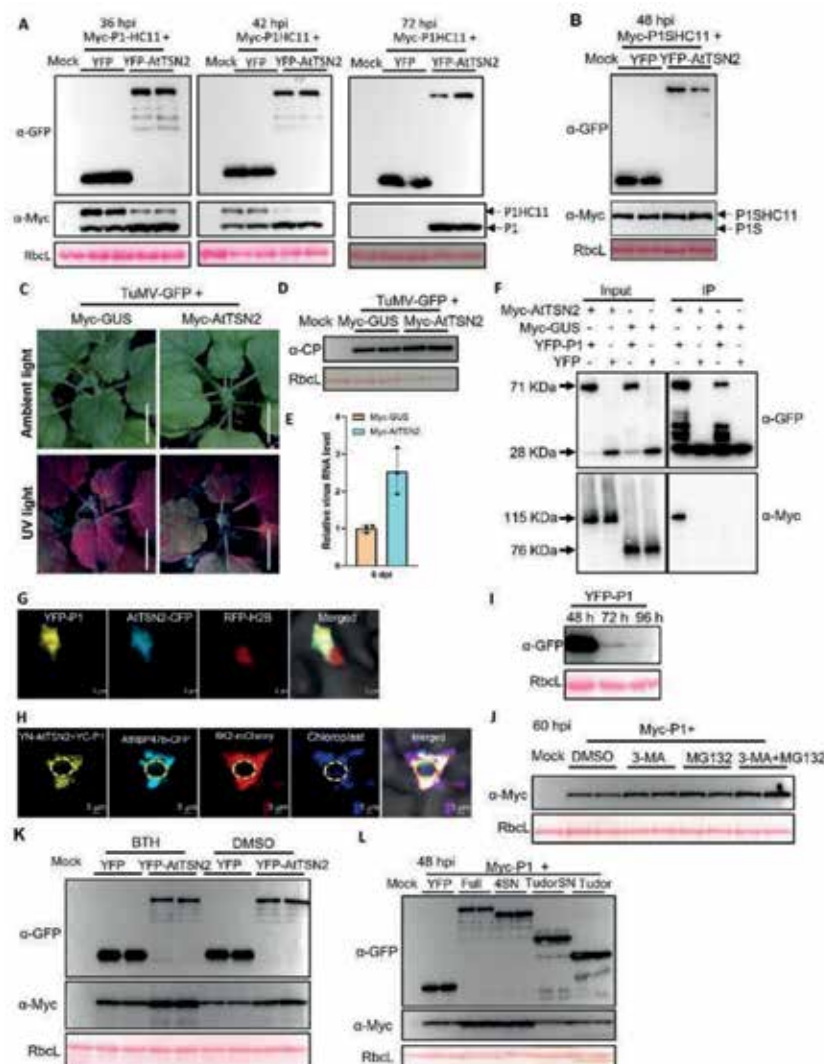
FIRST REPORT OF BIDENS MOTTLE VIRUS INFECTING ZINNIA ELEGANS JACQ. IN GUANGXI, CHINA

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Zinnia elegans Jacq., also known as common Zinnia, is an widely cultivated ornamental plant in China and many other nations. In april 2022, plants of Zinnia displaying symptoms of mosaic and leaf rolling were observed in the garden of the base of Guangxi academy of agricural sciences. To identify the causal pathogens, four plant samples with typical symptoms were collected and total RNA was extracted from pooled samples (n=4) using RNAiso Plus following the manufacturer's instruction. The sRNA library was constructed and then the high throughput sequenced (HTS) were performed using the illumina Nextseq500 platform. Total of 13,693,651 reads of raw data were obtained from the pooled samples and 12,716,092 reads of clean data were obtained. Then the clean reads were assembled by spades (the Kmer value was 17) and the assembled contigs were blast in NT and NR database. Only 10 contigs were annotated as virus sequences and showed high nucleotide identity (98-97 %) with bidens mottle virus. To verify the HTS results, specific primers were designed according to the HTS contigs and RT-PCR were applied for the viruses detection. The amplicons only detected positive in the diseased samples, but not in healthy plant. To further confirm the BiMoV infection Zinnia, ten healthy Zinnia plants were inoculated by mechanical inoculation of leaves sap extracts in phosphate balanced solution (0.05mol/L, pH 7.2). Typical symptoms were observed on 7 plants and RT-PCR were confirmed the infection of BiMoV. The results above revealed that BiMoV can infected zinnia plants both in natural and artificial conditions. Phylogenetic tree constructed on the basis of coat protein sequences revealed that BiMoV-GX isolate clustered a small branch with BiMoV-PL5 (AB601906) and BiMoV-Kunming, indicating their close evolutionary relationship. To our knowledge, this is the first report of BiMoV infecting Zinnia in China, and zinnia is a natural host of BiMoV.

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TSN2 FACILITATES POTYVIRUS INFECTION BY PROMOTING THE CLEAVAGE AND BLOCKING THE DEGRADATION OF THE VIRAL P1 PROTEASE

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Turnip mosaic virus (TuMV) in the *Potyvirus* genus, is the most devastating RNA virus in oil crops and cruciferous crops, which usually leads to destructive economic losses worldwide. Application of resistance genes is one of the effective strategy to control the virus in the field. In this study, using anion exchange chromatography, cation

exchange chromatography, gel filtration chromatography and mass spectrometry analysis, we identified a host factor, Tudor staphylococcal nuclease (TSN), which is involved in the P1 proteolytic process that is crucial for viral infection and host specifically. Overexpression of AtTSN2 promoted the cleavage ability of the TuMV P1 protein on P1-HC-Pro and stabilizes the free P1 protein to prevent its rapid degradation. The 4SN domain of AtTSN2 regulated the P1 protease cleavage activity and the protein stability of free P1, but the second SN domain (SN2) is mainly involved in controlling the cleavage activity of the P1 protease. The infection of TuMV induced the expression of TSN. Meanwhile, the viral infectivity was significantly enhanced in *AtTSN2*^{OE} transgenic plants, and vice versa in *AtTSN2* RNAi mutants. AtTSN2 interacted with P1 and co-localized with the marker proteins of stress granules (SGs) in viral replication complex (VRC). The P1 protein during viral infection was degraded through autophagy and 26s proteasome pathways, but AtTSN2 was able to block the autophagic degradation of P1. This study unveils the function and mechanism of AtTSN2 in the infection of TuMV, which may provide a novel TuMV recessive resistance gene in *B. napus* and cruciferous crops and a novel target for the prevention and control of potyviruses.

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P079

DEVELOPMENT OF INFECTIOUS CLONES AND GENE SILENCING VECTOR BASED ON BLACKBERRY CHLOROTIC RINGSPOT VIRUS AND BLACKBERRY YELLOW VEIN ASSOCIATED VIRUS

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Blackberry yellow vein disease (BYVD), caused by a combination of at least two viruses, is responsible for a notable decline in blackberry production in the Southeastern United States. Among the most prevalent viruses in the complex are blackberry yellow vein associated virus (BYVaV) and blackberry chlorotic ringspot virus (BCRV). The contribution of each virus to disease development and their interaction is poorly understood. Hence, we developed infectious clones and virus-induced gene silencing vectors (VIGS) for each of these viruses to initiate studies to advance our understanding of BYVD. To build the infectious clones, genomic RNAs, two for BYVaV and three for BCRV, were cloned into a PjL89 agrobacterium binary vector. Infectious clones were modified into VIGS vectors by adding a short insert of a targeted gene, phytoene desaturase (PDS) or green fluorescent protein (GFP), right at the start of the 3' UTR of RNA 2 and RNA 3, for BYVaV and BCRV, respectively. Constructs were delivered to plants through a syringe agroinfiltration method. Systemic tissue of *Nicotiana benthamiana*, for both viruses, and *N. occidentalis*, for BCRV, was used to determine virus infection through RT-qPCR. Down-regulation of the VIGS-targeted gene was assessed either by the presence of a photo-bleaching phenotype or the lack of green fluorescence under UV illumination in systemic leaves, for PDS and GFP, respectively. Both infectious clones induced systemic infection in their respective experimental hosts. The BYVaV-based VIGS vector induced silencing of GFP, while the BCRV-based VIGS induced silencing of both PDS and GFP. The outputs of this research will facilitate future investigations on the molecular biology of BYVD and its management, as well as functional genomics studies on the hosts of BYVaV and BCRV.

P080

COMPARATIVE ANALYSIS OF PATTERN-TRIGGERED IMMUNITY AND EFFECTOR-TRIGGERED IMMUNITY GENE EXPRESSION IN SUSCEPTIBLE AND TOLERANT CASSAVA GENOTYPES TO SACMV INFECTION

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South African cassava mosaic virus (SACMV) causes cassava mosaic disease which reduces the production yield of the cassava (*Manihot esculenta* Crantz) crop in many tropical and subtropical regions. Recent evidence suggests a mutual potentiation of pattern-triggered immunity (PTI) and nucleotide leucine-rich repeat (NLR) immunity (ETI) in antiviral immune responses. This study compared ETI and PTI defence strategies to confer resistance to SACMV in cassava. Identification of PTI, ETI and SA-responsive genes in SACMV-challenged susceptible T200 and tolerant TME3 cassava

germplasm was achieved by whole transcriptome sequencing (RNA-seq) and by reverse transcriptase quantitative PCR (RT-qPCR).

RNA-seq data and gene expression analysis revealed reduced expression of several components of PTI-associated signalling genes during SACMV systemic infection in susceptible T200 cassava germplasm. In addition to suppression of PTI responses, reactive oxygen species (ROS) production was greatly reduced in symptomatic cassava genotypes. In contrast, transcription of ETI signalling, and SA biosynthesis and signalling genes, were upregulated during SACMV systemic infection.

These results indicate that PTI signals are necessary for systemic acquired resistance against SACMV and suggest that SACMV targets PTI receptors to subvert PTI-mediated antiviral immunity in cassava

During early viral infection, replication-associated protein (AC2) establishes virus infection by inhibiting host PTGS mechanisms and nuclear shuttle protein (NPS) becomes the main pathogenetic determinant at the late stage of infection by suppressing PTI responses. The recovery phenomenon is achieved owing to suppressed viral NSP gene expression and activation of ROS-dependent basal immune response through NIK-pathway and possibly TNFs-mediated immune response.

P081

IDENTIFICATION OF PATHOGEN CAUSING TOMATO WHITE TIP AND CURL LEAF DISEASE AND ITS PATHOGENICITY IN GUANGDONG, CHINA

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Lisianthus enation leaf curl virus (LELCV) is a new species of the Genus *Begomovirus* discovered in 2015. In 2022, a new disease of tomato plants exhibiting leaf small, curling and leaf tips whitening symptoms occurred in guangzhou city, guangdong province, China. Total DNA was extracted from two disease samples respectively, and used as template for PCR detection with degenerate *Begomovirus* primers AV494/CoPR. The expected ~570 -bp fragment was obtained in all two symptomatic samples. The full genome sequence of isolate Tomato-2022 was obtained by RCA amplification, gene cloning and sequencing from positive samples by PCR detection. The genome of the virus isolate Tomato-2022 contained only DNA-A, with a size of 757 2 nt, and encoded six ORFs. The isolate Tomato-2022 had high nt similarity with LELCV isolates deposited in GenBank, and had the highest nt similarity with LELCV Taiwan *Eustoma grandiflorum* isolates (LC091539, LC091538) at 98.33 %. The constructed Tomato-2022 infectious clone pGreenII-0229 1.6A was inoculated into *N. benthamiana* and tomato by Agrobacterium-mediated injection. At 18 dpi, *N. benthamiana* and tomato plants showed obvious curling symptoms. At 30 dpi, *N. benthamiana* showed veins swelling and leaves severe curling, and the leaves of tomato plants showed curling and leaf tips whitening symptoms. The results of PCR showed that LELCV could be detected in both the inoculated plants.

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Tomato- 2022 is a new isolate of *Lisianthus enation* leaf curl virus, and also the first isolate found in mainland China. The virus isolate is the pathogen of tomato curl leaf and white tip disease.

P082

RECENT INSIGHTS INTO THE BIOLOGY AND EPIDEMIOLOGY OF TOMATO FRUIT BLOTCH VIRUS, A POTENTIAL NEW RISK TO TOMATO CROPS

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Fruit blotch virus - ToFBV, (*Blunervirus solani*, genus *Blunervirus*, family *Kitaviridae*) is the proposed name for a recently identified virus causing uneven and blotchy ripening along with dimpling and dark spots on tomato (*Solanum lycopersicum* L.) fruits. From the first identification, in -2018 2019 in Italy and Australia, ToFBV was reported in Brazil and Spain and retrieved from stored samples dating back to 2012 its presence in Italy. High Throughput Sequencing (HTS) analyses were leading to a growing number of records in several European and Mediterranean countries (Greece, Portugal, Slovenia, Switzerland, and Tunisia), and ToFBV was recently included into the EPPO Alert list as a potential new threat for tomato crops.

Studies on ToFBV biology and epidemiology have been conducted to increase knowledge and awareness of its phytosanitary risk. The ToFBV bacilliform particles are known to accumulate mainly in the pericarp rather than in other tissues of infected tomato fruits. Seeds are only contaminated on their external tegument; however, the virus appears unable to replicate in the emerging seedlings, thus excluding its seed transmissibility. The ToFBV mechanical inoculation on indicator plants also failed.

A likely candidate ToFBV vector is the tomato russet mite *Aculops lycopersici* Massee (family Eriophyidae). High population densities of *A. lycopersici* were found in several infected tomato cultivations worldwide and the only arthropod sequences, found in HTS RNA-seq datasets from ToFBV-positive tomato samples, belonged to *A. lycopersici*. Eriophyid collections in Italian tomato fields were performed to assess the potential rate of ToFBV acquisition by *A. lycopersici*. Laboratory transmission trials are ongoing as well. Such studies were facilitated by the development of molecular tools allowing simultaneously virus detection and eriophyid identification at species level.

P083

MECHANICAL TRANSMISSIBILITY OF TWO TOMATO LEAF CURL NEW DELHI VIRUS ISOLATES IN CUCURBIT SPECIES

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Tomato leaf curl New Delhi virus (ToLCNDV), a bipartite member of the genus *Begomovirus* in the family *Geminiviridae*, has emerged as a serious constraint on various crops since its rapid spread and extent of outbreaks in different countries, mainly in Asia and the Mediterranean region. In 2019, we identified a new isolate of ToLCNDV in Pakistan and analyzed together with ToLCNDV-ES which were previously reported in Italy. These two isolates were categorized as different strains, and our research focused on their molecular pathogenic differences via infectivity, pathogenic phenotype induction and transmission modes. Previously, we successfully constructed two ToLCNDV infectious clones using tandem repeat fragment method and figured out the differences in infectivity and symptoms development in tomato and cucumber plants. In a current study, the mechanical transmissibility of two ToLCNDV isolates has been investigated in cucurbit species which showed infectious via agro-inoculation assay. The results revealed that the Italian ToLCNDV (ToLCNDV-ES) showed a severer pathogenicity than Pakistani isolates (ToLCNDV-In) in squash zucchini hosts. The exchanged sub-genome components between two isolates suggest that DNA B played a determinant in symptom development through mechanical transmission. Additionally, we have been generating several chimeric clones to identify the region determining the mechanical transmissibility of ToLCNDV in squash zucchini. Our results provide a foundation for elucidating the viral determinant and molecular mechanisms underlying ToLCNDV infection by mechanical transmission.

P084

ANALYSIS OF CONSTRAINTS ON VIROID QUASISPECIES EVOLUTION

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RNA viruses and viroids replicate with high mutation rates, forming quasispecies, a population of variants centered around dominant sequences. The mechanisms governing quasispecies remain unclear. This study aims to understand the role of plasmodesmata, which regulate viroid movement, in regulating the structure of a viroid quasispecies.

The progeny of the potato spindle tuber viroid intermediate (PSTVd-I) strain from mature guard cells lacking plasmodesmal connections and from *in vitro*-cultivated mesophyll cell protoplasts from systemic leaves of early-infected tomato (*Solanum lycopersicum*) plants were sequenced. Coinfection and single-cell sequencing experiments were performed to test whether plant cells can be repeatedly infected. To study

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the impact of initial population composition on PSTVd-I quasispecies, we conducted coinfections with PSTVd-I and variants. Two inoculum ratios (10:1 or 1:10) established quasispecies with or without PSTVd-I as the master sequence. Mutant pools were prepared for some loops and stems of the PSTVd secondary structure, and the role of RNA 3D structure in regulating PSTVd-I quasispecies was analyzed by infection assay.

Remarkably, more variants accumulated in guard cells compared to whole leaves. Similarly, after extended cell culture, we observed more variants in cultivated mesophyll protoplasts. The same plant cell can be infected multiple times by the same or different PSTVd sequences. In the absence of the master sequence, the percentage of novel variants increased. Moreover, variants with an RNA 3D structure are more likely to accumulate in the quasispecies after cell-to-cell movement and long-distance trafficking.

These results suggest that cell-cell communication, initial population composition, and RNA 3D structure can shape the structure of PSTVd quasispecies.

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P085

EFFICIENT VIRUS-INDUCED GENE SILENCING IN SOYBEAN PLANTS THROUGH A NOVEL RECOMBINANT GEMINIVIRUS-MEDIATED TRANS-REPLICATION OF A BETASATELLITE

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Plant viruses have emerged as promising tools in agricultural and medical biotechnology. Understanding the expression strategies of plant viruses and their interactions of hosts and other viruses or satellites will facilitate the development of plant virus-derived vectors. In our previous research, we identified soybean yellow leaf curl virus (SbYLCV), a novel recombinant geminivirus that may belong to a new genus of the family *Geminiviridae* and was later demonstrated to be highly associated with soybean stay-green symptoms. To investigate the interaction of SbYLCV with betasatellites associated with begomoviruses, we selected six different betasatellites and assessed their ability to be trans-replicated by SbYLCV. Our findings revealed that SbYLCV selectively trans-replicated one of the six different betasatellites in both *Nicotiana benthamiana* and soybean plants. To leverage this interaction, we constructed a novel SbYLCV-DNA β silencing vector by replacing the pathogenic factor β C1 with a multiple cloning site. The modified vector efficiently silenced the endogenous *PDS* and *Su* in *Nicotiana benthamiana*, resulting in obvious phenotypic mutations. Significant silencing of the endogenous *Su* was also observed in *N.*

tabacum. Furthermore, the system enabled silencing of the endogenous *PDS* of soybean plants, leading to leaf chlorosis and photobleaching phenotypes. Through optimization of the inoculation method and the insertion of fragments with different lengths, we improved the performance of the silencing system in soybean plants. Notably, the modified silencing system efficiently silence the ribosomal protein 6 gene and simultaneously inhibited the expression of two endogenous genes in soybean plants. Moreover, the silencing signal triggered by SbYLCV-DNA β could transmit to root tissues. The establishment of the SbYLCV-DNA β vector system provides a valuable tool for the study, analysis, and discovery of gene functions in soybean and other plants.

P086

SCREENING OF ALLIUM PLANT GENETIC RESOURCE COLLECTION FOR PRESENCE OF POTYVIRUSES, CARLAVIRUSES AND ALLEXIVIRUSES

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Garlic (*Allium sativum* L.) and potato onion (*Allium cepa* var. *aggregatum* L.) are substantial vegetables for agriculture and phytotherapy. Like most vegetatively propagated plants, cultivated garlic and potato onions may accumulate several viruses that can cause severe crop losses and reduce yield amount and quality. In Latvia are long-standing garlic and potato onion cultivation traditions. During several centuries, diverse breeding lines and landraces have been developed, representing independent introduction sources. The present study supported by the National Fruit Crop and Vegetable Breeding Program (No.-1654/23/11-10.9.1e) is aimed to screen 49 potato onion and 93 garlic genotypes in the genetic resources collection for presence of seven viruses (garlic virus A (GarV-A), garlic virus B (GarV-B), shallot virus X (ShVX), shallot latent virus (SLV), garlic common latent virus (GCLV), leek yellow stripe virus (LYSV) and onion yellow dwarf virus (OYDV)) by RT-PCR. The total RNA was extracted using a Genomic DNA Purification Kit (Thermo Fisher Scientific) adapted for RNA extraction. RT-PCR was performed with the OneStep RT-PCR kit (QIAGEN) according to the manufacturer's instructions. The most common viruses in garlic were LYSV, GCLV and SLV, but in potato onions, they were ShVX and SLV. All tested garlic plants were infected with LYSV. GarV-A, GarV-B and OYDV were not detected in tested potato onion plants. This is the first study where garlic and potato onion viruses have been tested by laboratory techniques to evaluate the health status of the national germplasm in Latvia. Our findings indicate the urgent need for virus elimination so ensuring safe maintenance of national *Allium* germplasm. The development of meristem-tip culture technique has been started to obtain and establish virus-free *Allium* germplasm.

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IMPROVING A DOUBLESEX ALTERNATIVE SPLICING MEDIATED RICIN TOXIN-A SYSTEM FOR BACTROCERA DORSALIS CONTROL

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Oriental fruit fly, *Bactrocera dorsalis*, causes severe economic losses in many Asian countries and the number of females directly related to the degree of damage. In order to solve the problem, a system combine ricin toxin A chain gene and intron 3 of *doublesex* gene (RTA-Bddsx) has been developed focusing on female-specific lethal effect. However, a defect has been found in original design that the *actin 5C* promoter constantly expression of RTA killed all transgenic female flies and hard to keep those lines. In this study, a heat shock promoter has been used for selectively expression of RTA. This improvement allows modified flies can cultivate normally at 22 °C, but induce female suppressing after heat shock treatment. A heat shock promoter of *Drosophila melanogaster* was first identified and used to construct pB-Hsp-LERQ plasmid for the RTA expression; subsequently, embryo injection has been performed and a series of Ds-red fluorescent screening performed. Several lines with germ-line transmission have been identified and established; the transgenic flies can be cultivated at 22 °C with normal sex ratio but showed female-lethal effects after heat shock treatment. In addition, many genomic DNA analyses will be conducting to confirm the integrity of the exogenous RTA in those transformed flies and many functional assays now also be done for optimum maintenance conditions including different temperature tests. Data collection covers the hatching rate, pupation rate, emergence rate, and the sex ratio of adult flies. We believe these results are an important assessment to future application. It improves the sterile insect technique (SIT) method and provide a better choice to *Batcorder dorsalis* control.

P088

DIFFERENTIAL RESPONSE OF ADULTS OF TWO GEOGRAPHICALLY ISOLATED POPULATIONS OF BACTROCERA OLEAE (DIPTERA: TEPHRTIDAE) TO THERMAL AND HYDRIC STRESS

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The olive fruit fly, *Bactrocera oleae* (Diptera: Tephritidae) is a major pest of olives in many olive-growing areas globally. The response of *B. oleae* to various environmental stressors has been extensively studied. However, the response of populations from different climate zones has not been addressed. We estimated the cold/heat tolerance and desiccation/starvation resistance of two olive fruit fly populations obtained from Greece and Israel. Specifically, five days after adult emergence, we acclimated the adults

for five days at 15, 20, 25, 30°C, and then we estimated their critical thermal limits (CT_{min} and CT_{max}). Resistance to extremely low relative humidity (<10 %) (desiccation) and to food deprivation (starvation) were also assessed. Significant effects of population and thermal acclimation on CT_{min}, but not on CT_{max} were found. The population from Greece expressed lower CT_{min} than that from Israel. The lower the temperature of acclimation, the lower the CT_{min} values. Females and males responded similarly both at low and high temperatures. Regarding desiccation resistance, all tested factors and their interactions significantly affected adult survival. Regardless of the thermal acclimation regime, adults from Greece responded better than that from Israel, and regardless of the population females survived longer than males. Population, acclimation, and all interactions (except population*acclimation) affected adult survival under food starvation. Females and males responded similarly to starvation. Both for desiccation and starvation assays, adults acclimated at 30°C survived for a significantly longer period compared to those acclimated at 15°C. Our findings broaden the existing knowledge on stress physiology of *B. oleae* and can be used for the development of population dynamics and invasion models providing data for important parameters that determine its persistence and thriving of populations to marginal for their existence environments.

P089

TRUNK INJECTION: AN ALTERNATIVE PLANT PROTECTION METHOD IN WALNUT AND CHERRY PRODUCTION

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Our research group has been working with trunk injection plant protection method development for four years. Choice of topic was justified by the fact that new environmental challenges need for sustainable solutions, especially for large woody plants where the spray drift is relevant. This problem can be achieved by trunk injection. Our aim was to develop endotherapy treatment, during which the active ingredients are directly injected into the trunks of the walnut and cherry trees, thus protecting them against their most important pests (*Rhagoletis completa* and *Rhagoletis cerasi*), in addition to being safe to use. Trunk injections were made in spring with abamectin, acetamiprid, emamectin benzoate, flupyradifurone and cyantraniliprole in different doses. The evaluation was carried out using entomological methods (counting of maggots in the pericarp of the fruits) and chemical methods (analytical method: determination of active ingredient residues with UHPLC-MS/MS). During our work, we searched for answers to the following questions: 1 (When should be the trees injected?); 2 (What is the minimum effective dose against these pests?); 3 (Is there a second

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year effect of endotherapy?); 4 (How homogeneous is the distribution in the canopy?); 5 (Can fruits be consumed from a food safety point of view?); 6 (Is this technology safe for bees?); 7 (Is it possible to replace the trunk injection with trunk spraying?). According to our measurement data, this technique can be used effectively and safely for the purpose. We provide detailed information about our results in the poster.

P090

MANAGEMENT OF FALL ARMYWORM (*SPODOPTERA FRUGIPERDA*) ON SUMMER MAIZE IN LEBANON

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Currently, fall armyworm (FAW), *Spodoptera frugiperda*, is the most important insect pest of maize planted in spring and summer in Lebanon. This led to the excessive use of insecticides to combat FAW using 2 to 3 unsafe pesticides weekly. To save maize crops, a field trial was carried out during the summer of 2023 in the farmers' field to evaluate environmentally safe and low-risk insecticides recommended by FAO and Lebanese Ministry of Agriculture to combat FAW using data from pheromone traps. The pheromone used was Z-9tetradecenyl acetate (1.96 mg), Z-7dodecenyl acetate (0.02 mg), and E-7 dodecenyl acetate (0.02 mg) produced by Sanidad Agrícola ECONEX S.L., Spain. The Trap (unitrap, universal trap) was placed five days after planting in farmer's field planted with maize cv. Nour on 25 July 2023. Foliar insecticides were sprayed using the recommended dose: 2 sprays with Emamectin benzoate + Alpha-cypermethrin; 2 sprays with Indoxacarb + Alpha-cypermethrin; and one spray with Deltamethrin. The number of captured FAW male and non-FAW moths were counted two times/week starting 31 July 2023. The results obtained showed that the peak of FAW population was during August 22 - September 15. In addition, FAW damage on maize production in the field sprayed with recommended insecticides based on the data from pheromone traps was almost unobservable compared to neighboring farmers' fields, who followed their own practices. Adjusting planting date and pheromone trap based safe pesticide application is recommended to reduce the impacts of the pest on the quality and quantity of green cobs.

P091

INTEGRATED MANAGEMENT OF FABA BEAN VIRUSES AND THEIR VECTORS IN LEBANON

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Faba bean viruses and their aphid vectors are major problems for Lebanese farmers, resulting in low faba bean productivity. Besides the direct harm, aphids (e.g. *Aphis craccivora*, *A. fabae* and *Acyrtosiphon pisum*) transmit devastating legume virus diseases. To save the crop and restore farmers' confidence in growing faba bean, a field trial was conducted in the farmers'

fields to evaluate options for managing viruses and their aphid vectors during the 2023/2022 growing season. The options evaluated were: (i) three imported faba bean varieties (Italian, Moroccan and Spanish), (ii) two sowing dates (24 November 2022 and 5 January 2023), and (iii) faba bean seeds treatment with Celest top (25g/L difenoconazole + 25 g/L fludioxonil + 262.5 g/L thiamethoxam) at the rate of 1 cc/kg of seeds with untreated seeds as control. The experiment was conducted using split-factorial design in two replications. Results showed that the number of infected plants with viruses (yellowing, stunting, mosaic, mottling) and aphids were higher at the 2nd sowing date compared to the 1st sowing date, whereas viruses and aphids incidence were lower in the plots planted with treated seeds compared to the plots planted with untreated seeds and for both sowing dates. Moreover, no aphids were recorded in the plots planted with treated seeds for all three faba bean varieties sown in November (1st sowing date). Serological tests of the plants associated with virus symptoms revealed that 48 % of the plants were infected with Bean yellow mosaic virus (BYMV), 32 % with Chickpea chlorotic stunt virus (CpCSV) and 19 % with Faba bean necrotic yellows virus (FBNYV). FBNYV and CpCSV are transmitted by aphids in a persistent manner only, whereas BYMV is transmitted by aphids in a non-persistent manner as well as by seeds.

P092

BIODIVERSITY OF FUSARIUM IN BANANA PLANTS IN THE CANARY ISLANDS (SPAIN)

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Bananas are one of the most important crops in the Canary Islands. Its production is distributed in different bioclimatic zones of the archipelago and covers more than 9,000 ha. Different diseases affect the banana crop in the subtropical areas, among which *Fusarium* Wilt of Banana (FWB) stands out for its severity. The aims of this work were to determine the diversity of the genus *Fusarium* in the different islands and production regions of the Canary Islands and to confirm the main causal agent of FWB in this subtropical area. Sixty-five farms located on five islands were visited (Figure 1). On each farm, pseudostem and rhizome samples were collected from three symptomatic and three healthy plants. All samples were planted onto Potato Dextrose Agar (PDA) and incubated at 25°C for 6-4 days. All fungal colonies with *Fusarium* morphology were subcultured and purified on the same medium. The identification at the species level was performed by phylogenetic analysis of the *tef-1a* gene, while the identification of the different races of *Fusarium oxysporum* f. sp. *cubense* (Foc) was performed using specific primers of the Secreted In Xylem (SIX) genes. A total of fifteen *Fusarium* species were identified. Three species (*Fusarium phialophorum*, *F. joanfreemanieae* and *F. crassum*)

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were observed in all the islands, while seven species were minority and were detected only on one of the five islands. In asymptomatic plants *Fusarium joanfreemaniae* was the dominant species. Whereas in the symptomatic plants the main species was *Fusarium phialophorum*. All of the isolates obtained of this species (550) were positive in Six8-b (specific to Subtropical Race 4 -STR4) and negative in Six-1a (TR4). A high diversity of *Fusarium* species in the Canary Island was detected and the causal agent of FWB was the Foc-STR4 (*Fusarium phialophorum*).

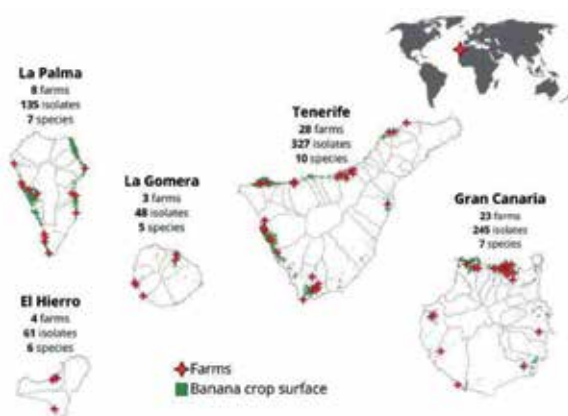


Figure 1. Map of sampling collection.

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EFFECT OF PGPR BACTERIA ON GROWTH AND BIOPROTECTION GREENHOUSE CROPS AGAINST BOTRYTIS CINEREA

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Grey rot caused by *Botrytis cinerea* is a predominant disease of vegetable crops, particularly those grown in sheltered greenhouses. This pathogen is described as an aggressive necrotroph because of its ability to invade host plant tissues, causing severe damage and economic losses. In this respect, protection via biological agents seems to be one of the most promising alternatives. It is within this framework, that this study was carried out with the aim of (i) evaluating the efficacy of epiphytic and endophytic bacteria in the biological control (in vitro and under glass) of gray mold in greenhouse crops (tomato and pepper) and (ii) studying their biostimulatory effect on growth (in vitro). The results obtained showed that four *B. cinerea* isolates were virulent and capable of inducing necrosis on tomato and chili stems, with the surface area of necrosis increasing over time. After validation of direct confrontation tests, five PGPR bacterial strains were selected: *Bacillus subtilis* (57), *Bacillus amyloliquefaciens* (S15c), *Bacillus siamensis* (Ki71), *Enterobacter cloacae* (TE8), and *Halomonas cupide* (Ni71). In tomato, *Enterobacter cloacae* (TE8) and *Halomonas cupide* (Ni71) strains showed antifungal potential against *Botrytis* canker in both preventive and curative treatments. In chili, *Bacillus siamensis* (Ki71) and

Enterobacter cloacae (TE8) strains were the most effective in inhibiting stem canker progression. The bacteria screened were also capable of producing hydrolytic enzymes (protease, cellulase and amylase), diffusible volatile organic compounds and biofilm production. Inorganic phosphate solubilization, nitrogen fixation and auxin production were the most common PGP activities detected in the strains. With the exception of *Bacillus amyloliquefaciens* (S15c), the other strains were unable to solubilize potassium. The mechanisms of action already mentioned may well be responsible for the strains' antagonistic power towards *B. cinerea*.

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VERTICILLIUM DAHLIAE IN FLAX: QUANTIFICATION OF SOIL INOCULUM, ISOLATE CHARACTERIZATION AND EFFECTS OF CROPPING FACTORS

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The fungal pathogen *Verticillium dahliae* has become a limiting factor in the production of high quality fiber flax in the region of Flanders, Wallonia, and Northern France. This has sparked interregional research towards the detection, characterization, and biology of *V. dahliae* in this crop. To quantify the pathogen in soil, a method using density flotation, DNA extraction and qPCR was validated during interlaboratory assays, evaluating factors such as sample pre-processing and qPCR assay. The pathogen was then quantified in the soil of nearly 300 fields over a two year period and correlated to yield, together with cropping factors such as soil texture, tillage method, seeding density, fertilization, and crop rotation. We also performed a genetic and pathological characterization of isolates from flax soil and hay using genotyping-by-sequencing (GBS). Factors that had an important effect on the pathogen quantification were sample pre-processing and homogenization, DNA quantification of the standard curve, qPCR assay and mastermix used. Flax yield was most affected by the factor year, which differed strongly due to weather differences. Most cropping factors had a limited effect on yield, or showed an interaction with the factor year. *Verticillium* quantity in the soil did have a negative effect on fiber yield. A small negative effect on flax yield was linked to a preceding crop of potato, which is known to increase the *V. dahliae* inoculum. The isolates from the fields of each of the three regions belonged to either one or two clearly distinct GBS groups, which corresponded with Vegetative Compatibility Groups (VCG) 2B and 4B. Little intra-group genotypic diversity was observed. Controlled inoculation assays did not reveal a difference in pathogenicity on flax between representative isolates of the two groups. This research provided methodological insights and revealed knowledge about the biology of *V. dahliae*, which can contribute to its management.

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FUSARIUM OXYSPORUM F. SP. LACTUCAE AS THE CAUSAL AGENT OF LETTUCE WILT IN GREECE: IDENTIFICATION, PATHOGENICITY AND DEFENSE-RELATED GENES' EXPRESSION

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Fusarium oxysporum f. sp. *lactucae* is the most significant pathogen of lettuce cultivation causing significant yield losses in intensive cropping systems worldwide. To date, the specific pathogen has been differentiated into 4 races based on the pathogenic ability of given isolates using a set of differential lettuce cultivars which most times is supported by molecular analysis. In this study, 84 isolates of *Fusarium oxysporum*, obtained from soil-grown lettuce plants exhibiting wilt symptoms, were characterized as belonging to race 1 of *F. oxysporum* f. sp. *lactucae* based on sequence analysis of the translation elongation factor -1a (*TEF-1a*) gene and the rDNA intergenic spacer (rDNA-IGS) region. Four representative isolates were confirmed to be associated with race 1 based on the reaction observed upon pathogenicity tests with a set of differential lettuce cultivars. Furthermore, artificial inoculations on the most cultivated lettuce cultivars in Greece revealed that the tested cultivars varied regarding their susceptibility to *F. oxysporum* f. sp. *lactucae* race 1. Two cultivars (cvs.); "Cencibel" and "Lugano" were found to be highly susceptible, while cvs. "Sandalina" and "Starfighter" were the most resistant ones. Expression analysis of 10 defense-related genes (*PRB1*, *HPL1*, *LTC1*, *SOD*, *ERF1*, *PAL1*, *LOX*, *MPK*, *BG*, and *GST*) was carried out on artificially inoculated lettuce plants of the four above cultivars at different time points after inoculation. In resistant cultivars, a higher induction rate was observed for all the tested genes in comparison with the susceptible ones. Taking into account that the use of resistant cultivars is considered as the main method of controlling *Fusarium* wilt of lettuce, the results of this study are expected to contribute towards the implementation of an integrated management program to control this disease in lettuce cultivation.

(Oxford Nanopore Technologies). Selected fungal-specific primers were used to amplify three different genomic DNA loci (ITS, beta-tubulin, and 28S LSU) originating from olive twigs. A multiplex metabarcoding approach was initially evaluated using healthy olive twigs, and further assessed with naturally infected olive twig samples. Bioinformatic analysis of basecalled reads was carried out using MinKNOW, BLAST+ and R programming, and results were also evaluated using the BugSeq cloud platform. Data analysis highlighted the approaches based on ITS and their combination with beta-tubulin as the most informative ones according to diversity estimations. Subsequent implementation of the method on symptomatic samples identified major olive pathogens and endophytes including genera such as *Cladosporium*, *Didymosphaeria*, *Paraconiothyrium*, *Penicillium*, *Phoma*, *Verticillium*, and others.

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FUSARIUM WILT IN LETTUCE: EPIDEMIOLOGY, ALTERNATIVE HOSTS AND NON-CHEMICAL CONTROL STRATEGIES

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Fusarium wilt of lettuce, caused by *Fusarium oxysporum* f.sp. *lactucae* (Fol), poses a serious threat to the soil-grown lettuce industry. In Europe, Fol race 1 is common in warmer climates while race 4 is more prevalent in colder climates. In our applied research project FOSSY we investigate three major research lines: (1) introduction and spread of Fol, (2) evaluation of alternative lettuce varieties and crops to mitigate Fol soil inoculum, and (3) exploration of non-chemical control strategies. (1) Race-specific qPCR assays were designed to study the epidemiology of Fol in Belgium. Analysis of numerous samples, ranging from swabs of boxes to water samples, revealed the significance of agricultural hygiene practices. Farm equipment can easily spread Fol if not disinfected properly. To underscore the importance of limiting the spread, a variant of race 4 (Fol +4) was recently discovered. Fol +4 can cause symptoms in lettuce varieties semi-resistant to race 4. The genomic basis underlying the differences in pathogenicity profile was determined using whole genome sequencing and data analysis. (2) The qPCR assays were used to evaluate inoculum build-up in tolerant lettuce varieties and alternative crops. Fol soil inoculum may be maintained when crops that support (asymptomatic) development of Fol are included in crop rotation. (3) Techniques to reduce Fol soil inoculum were evaluated. Regular sheet steaming proved ineffective as Fol soil inoculum was not sufficiently suppressed. Steaming with active drainage resulted in stronger reduction of the pathogen.

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NANOPORE-SEQUENCING METABARCODING FOR IDENTIFICATION OF PHYTOPATHOGENIC AND ENDOPHYTIC FUNGI IN OLIVE (OLEA EUROPAEA) TWIGS

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Metabarcoding approaches for the identification of plant disease pathogens and characterization of plant microbial populations constitute a rapidly evolving research field. Fungal plant diseases are of major phytopathological concern; thus, the development of metabarcoding approaches for the detection of phytopathogenic fungi is becoming increasingly imperative in the context of plant disease prognosis. We developed a multiplex metabarcoding method for the identification of fungal phytopathogens and endophytes in olive young shoots, using the MinION sequencing platform

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Chitin as a soil amendment gave varying results. Finally, during greenhouse experiments, several registered and non-registered commercial products containing biological control agents (BCA's) were tested, but didn't reduce Fol symptoms significantly. Experimental BCA's showed potential *in vitro*, but need field validation. As no single method resulted in complete reduction of Fol soil inoculum, development of an Integrated Pest Management strategy is necessary.

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VIRUSES WITHIN VERTICILLIUM SPP

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Verticillium sensu stricto is an anamorphic genus within the pathogenic plant fungi of Ascomycetes that causes verticillium wilt, one of the most harmful vascular diseases that affects numerous crops worldwide. Among *Verticillium* species, *Verticillium nonalfalfae* is particularly important for hop production, as it may destroy the entire plantation upon infection with the lethal pathotype. Mycoviruses are omnipresent viruses, that infect fungi. In *Verticillium dahliae* and *Verticillium albo-atrum*, five mycoviruses have been identified so far: *Verticillium dahliae* chrysovirus 1 (VdCV1), *Verticillium dahliae* partitivirus 1 (VdPV1), *Verticillium dahliae* magoulivirus 1, *Verticillium dahliae* RNA virus 1 (VdRV1), and *Verticillium albo-atrum* partitivirus 1 (VaaPV1). *V. nonalfalfae* isolates were obtained from the culture collection of the Slovenian Institute for Hop Research and Brewing. Small RNAs (sRNAs) were isolated with mirVana miRNA Isolation Kit. sRNA libraries were prepared using the Ion Total RNA-Seq kit. Barcode-labeled cDNA libraries were sequenced on an Ion PI chip v3 using an Ion Proton Sequencer (Ion Torrent; Life Technologies). According to the Ion Torrent sequencing pipeline, raw reads had removed adapter sequences. The sRNA-seq data were analyzed using the VirusDetect pipeline and CLC Genomic Workbench, and validated with RT-PCR and Sanger sequencing. sRNA-seq resulted in 10,468,293-4,387,811 reads per pool. sRNA-seq revealed that VaaPV1, with a bipartite genome, naturally infects *V. nonalfalfae*. The total number of reads mapped to VaaPV1 RNA1 segment ranged between 7510-965, while the total number of reads mapped to VaaPV1 RNA2 segment ranged between 18467-1237. In silico predicted results were validated by RT-PCR and Sanger sequencing.

The effects of VaaPV1 on fungus virulence will be confirmed by: (i) curing infected strains; (ii) virus transmission on other strains (hyphal anastomosis or transfection of fungal protoplasts with purified virus particles); (iii) pathogenicity assay on hops.

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RACES AND PATHOTYPES OF VERTICILLIUM DAHLIAE: MOLECULAR CHARACTERIZATION AND VIRULENCE PATTERNS ON DIFFERENTIAL TOMATO CULTIVARS, COTTON, AND EGGPLANT

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Verticillium dahliae is responsible for vascular wilts in several economically important crops. In this fungus, three pathogenic races have been documented (R1, R2 and R3), distinguished by their pathogenicity on differential tomato cultivars. Also, two pathotypes, the non-defoliating (ND) and defoliating (D), with the latter causing defoliation on cotton, olive and okra, have been described. The objectives of this study were to investigate virulence diversity within and between races and pathotypes subgroups of *V. dahliae*, and to determine the correlation between molecular classification and pathogenicity. To achieve this, 32 isolates from various hosts were characterized with race-specific and multiplex pathotype-discriminating PCR assays. In addition, pathogenicity assays on differential tomato cultivars, cotton and eggplant were carried out with 15 selected isolates. Furthermore, quantification of the pathogen was assessed on tomato stems using "TaqMan" qPCR. On the molecular level, each isolate was assigned on a single pathotype, and in almost all cases, race-specific primers differentiated the selected isolates. However, two isolates could not be assigned to any race. Overall, R1 isolates were strictly correlated with the ND pathotype and non-R1 isolates were assigned either on the D or ND pathotype. Pathogenicity assays on tomato revealed cultivar-dependent differentiation within R1 isolates in disease response but also in pathogen biomass. Moreover, low, and moderate virulence of D isolates was observed on tomato and eggplant, respectively. Pathogenicity assays on cotton underlined that defoliation and virulence should be noticed as separate traits, since maximum virulence was accomplished by ND isolates. To the best of our knowledge this is the first report of R3 occurrence in *V. dahliae* populations in Greece. In conclusion, the results obtained in this study showed that a "molecular toolbox" when assisted with phenotypic assays can support a precise description of race and pathotype variation of the pathogen population, important traits to design control strategies.

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AN ECO-FRIENDLY ALTERNATIVE TO MANAGE AILANTHUS ALTISSIMA, ONE OF THE WORST INVASIVE PLANT SPECIES OF THE WORLD

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Ailanthus altissima is a deciduous tree of Simaroubaceae family, native to China. Since 2019, this species has been included in the list of Invasive Alien Species of European Union. The potential of *Verticillium* species as biological

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control agents (BCAs) to counteract *A. altissima* might deserve attention, given the failure of other conventional strategies (i.e., mechanical and chemical methods). Here a strengths, weaknesses, opportunities and threats (SWOT) analysis was performed in order to identify the internal (resources and experience readily available) and external factors (typically uncontrollable items) that are favourable or not to *Verticillium*-based products as BCAs. Specifically, two cases regarding *V. nonalfalfae* (United States and Austria) and one concerning *V. dahliae* (Italy) were evaluated by considering that all fungal species were isolated from naturally infected *A. altissima* trees and are specialized on the target. *Verticillium*-based products resulted completely effective in *A. altissima* management, being able to kill plants within few years, can be transmitted through root graphs guaranteeing the transmission to clonal stands, and are eco-friendly (without chemical residues release). Unfortunately, a common weakness of these BCAs is the risk to infect other hosts, since preliminary tests revealed the susceptibility of ten woody and few horticultural/floral species to *V. nonalfalfae*, and few crops to *V. dahliae*. This factor further obstacles the process to recognize at regulative level the *Verticillium*-based products, known to be particularly difficult and already identified as the main threat. To overcome these issues, appropriated indications of safe spaces in which the BCAs can be applied without risks, should be provided. This may also increase the opportunities of the use of *Verticillium*-based products where the presence of potential non-target species is scarce, as in industrial and archaeological areas, and railways traits. Further researches added to active communicative campaigns are necessary to promote these BCAs to different stakeholders.

P101

DIVERSITY OF PECTOBACTERIUM SPECIES RESPONSIBLE FOR CAUSING BACTERIAL SOFT ROT ON BRASSICA PARACHINENSIS IN SOUTH CHINA

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Bacteria from *Pectobacterium* genus, which were previously denoted for soft-rot *Erwinia*, are widely spread and caused a broad spectrum of bacterial soft rot diseases (soft rot, blackleg, and stem wilt) in a wide host range of important crops. Chinese flowering cabbage (*Brassica parachinensis* L.) known as Choi-sum is an important leafy vegetable in China. However, its growth is seriously affected by various diseases, such as bacterial soft rot. Bacterial soft rot caused by *Pectobacterium* genus is a common disease on Choi-sum, but the causal pathogen of bacterial soft rot on Choi-sum is ambiguity. In the years 2021 and 2022, seventy-three *Pectobacterium* sp. strains were isolated from Choi-sum showed bacterial soft rot symptom collected from seven counties in Guangdong province, China. The results of physiological and biochemistry, Multilocus sequence analyses and pathogenicity assays show that sixty-three strains were *Pectobacterium brasiliense*, four strains were *Pectobacterium carotovorum*, three strains were *Pectobacterium*

quasiaquaticum, one strain was *Pectobacterium aroidearum* and one strain was *Pectobacterium colocasium*. Further, the virulence of the six selected isolates GDPB1, GDPC5, GDPAr1, GDPcol1, GDPQua1 and GDPQa2 were carried out by using injection. The result showed that GDPB1, GDPC5, GDPAr1, GDPcol1 and GDPQua1 caused soft rot symptoms on Chinese flowering cabbage, potato, tomato and Chinese watermelon, while GDPQa2 caused soft rot symptoms on Chinese flowering cabbage, potato and tomato. This result will help us to develop an effective prevention and control measures for bacterial soft rot on Chinese flowering cabbage.

P102

THE DEFENSIVE ROLE OF SECONDARY METABOLITES AND SMALL RNAs IN HOP (HUMULUS LUPULUS) DURING VERTICILLIUM NONALFALFAE INFECTION

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MicroRNAs control gene expression at the post-transcriptional level and thus regulate a variety of biological processes. MicroRNAs also ensure that the regulation of genes involved in pathogen defense and stress response is precisely controlled. Little is known about hop miRNAs and their role in the response to *Verticillium nonalfalfae*.

Resistant and a susceptible hop cultivar were artificially inoculated with the phytopathogenic fungus *Verticillium nonalfalfae* and the profiles of phenolic compounds and fungal colonization were determined at different time points after inoculation. Analysis of RNAi genes (three Argonaute genes, two Dicer-like proteins and two RNA-dependent RNA polymerase genes) showed that *AGO2* was significantly down-regulated in both cultivars at day 3 post-inoculation, which may result in reduced post-transcriptional gene silencing mediated by AGO-2bound small RNAs. Decreased expression of *RDR6* in the resistant cultivar at day 3 may indicate suppression of biogenesis of trans-acting siRNAs. In a study on miRNA, we identified 56 known and 43 novel hop miRNAs. In response to infection with *V. nonalfalfae*, we detected changes in the expression of five known and two novel miRNAs in the susceptible cultivar and six known miRNAs in the resistant cultivar. The differentially expressed miRNAs silence 49 transcripts that are involved in protein localization and pigment synthesis in the susceptible variety, whereas they are involved in the regulation of transcription factors and hormone signaling in the resistant variety. Using the RLM-RACE method, we confirmed the binding and cleavage sites of miR160a and miR319c-f. The upregulation of miR160a in the resistant cultivar could suppress auxin response factors leading to stabilization of Aux/IAA repressors and consequently downregulation of the auxin signalling pathway. Similarly, upregulation of miR319c-f could lead to suppression of genes that inhibit root growth (Teosinte branched 1), branching, and vascular tissue development.

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EARLY DETECTION OF RICE LEAF FOLDER INFESTATION IN RICE USING MULTISPECTRAL UNMANNED AERIAL VEHICLE IMAGERY AND DEEP LEARNING

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The impact of rice leaf folder infestations on rice production is significant, establishing early detection systems becomes crucial. This study aims to develop a system to assist agricultural decision-making by integrating visible/multispectral UAV imagery with deep learning techniques to detect infestations in rice.

The research utilized visible/multispectral images captured by UAVs over rice fields, applying Grad-Cam for image classification, detection, and segmentation tasks. Features from the multispectral images were used to train deep learning models to recognize early signs of infestation.

The model demonstrated excellent performance in identifying infested seedlings during the training phase, with accuracy, precision, and specificity were 93.2 %, 97.9 %, and 99.3 %. Independent validation also showed considerable potential. However, there remains potential for further refinement.

The integration of multispectral UAV imagery with deep learning demonstrates significant potential for the early detection of rice leaf folder infestations, which could lead to a reduction in the use of insecticides. Additionally, by diminishing reliance on chemical pesticides, it promotes sustainable agricultural practices. This method is promising for monitoring and managing other crops, optimizing irrigation and fertilization decisions to improve efficiency and reduce labor in precision agriculture.

P104

A QPCR-BASED ASSAY FOR THE QUANTIFICATION OF OVERWINTERING CHASMOTHECIA OF ERYSIPE NECATOR IN GRAPEVINE BARK

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Powdery mildew (PM) disease causes serious losses in Mediterranean vineyards, where suitable environmental conditions promote repeated conidial infections during the grapevine-growing season. The frequency and severity of these infections are directly linked to the amount of primary inoculum of *Erysiphe necator*; i.e. the chasmothecia developed at the end of summer on leaves that overwinter embedded in the bark of trunk. Numbers of chasmothecia in a vineyard is currently estimated through their counting in special funnel traps or in bark samples, which is difficult and time consuming. In this work, we set up a protocol to extract and quantify the chasmothecia of *E. necator* in grapevine bark samples

based on a qPCR assay. Moreover, we observed PM severity and ascocarp production on leaves in a season and primary infection in the following season, on different grapevine CVs. with known levels of powdery mildew susceptibility. The qPCR showed a significant relationship between the *E. necator* DNA in bark samples and the primary infection ($R^2 = 0.970$) and the disease severity development ($R^2 = 0.776$), as well as the chasmothecia development on leaves ($R^2 = 0.455$). The results demonstrate that the methodology can be used for the quantification of chasmothecia improving current protocols based on visual counting, proving the interrelationships between PM epidemics and overwintering chasmothecia. Quantification analysis can be useful for refining PM disease prediction models and the subsequent fungicides application.

P105

USING HYPERSPECTRAL DATA TO MONITOR THE RESPONSES OF A TUSCANY (ITALY) TOMATO CULTIVAR UNDER ABIOTIC AND BIOTIC STRESSORS

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Advancements in the ability to rapidly detect plant responses to stress are required to improve crop management practices and meet the global challenge of food security. The present study aimed to investigate the capability of full-range (-350 2500 nm) reflectance spectroscopy to discriminate the tomato cultivar 'Pisanello' (typical of Tuscany, Central Italy) from other two ('Goldmar' and 'Isbylia'), which are closely related from a phenotypic point of view, also in presence of abiotic and biotic stress. Plants were grown in soilless culture using rock wool, and treated as follows: (a) control with electrical conductivity (EC) equal to 3 mS cm⁻¹; (b) salinized, with EC equal to 6.0 mS cm⁻¹; (c) treatment with low nutrient level (K), having a nutrient solution composition similar to the control but with a 70 % reduction in K concentration. Some of these plants were also tested positive for tomato mosaic virus (ToMV). Leaf and fruit hyperspectral data were collected at different times. Permutational multivariate analysis of variance (PERMANOVA) of leaf and fruit spectral signatures (2400-400 nm) showed a number of significant effects, both as singular and interactive (e.g., 'cultivar × time × salt treatment' and 'cultivar × time × K treatment' at both leaf and fruit level). PERMANOVA also showed a highly significant effect of ToMV infection on leaf hyperspectral profiles. According to PERMANOVA, partial least discriminant analysis showed an excellent accuracy for the discrimination of most of the experimental theses, in some cases even more than 90 %. Variations of spectral vegetation indices and leaf traits derived from spectra by developed partial least squares regression-models highlighted the differential responses of tomato cultivars to the investigated stresses. In conclusion, we found that hyperspectral information successfully identified crop status in multi-factor experiments and detected specific responses to stress conditions prior to the onset of visual symptoms.

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PRELIMINARY EARLY DETECTION OF TOMATO SPOTTED WILT VIRUS USING IOT DIGITAL ELECTRONIC NOSE IN COMBINATION WITH ARTIFICIAL INTELLIGENCE ALGORITHMS.

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The early detection of plant viruses poses significant challenges to agricultural sustainability and productivity. The spread of invasive pests is accelerated by globalization and changes in climate conditions, increasing the need of prompt and early detection. Traditional methods for virus detection often involve complex laboratory procedures, making them time-consuming and costly. Combining new technologies [i.e., digital electronic nose and artificial intelligence (AI)] could be promising in the field of agricultural prevention. In recent years, the use of this low-cost, portable, IoT and open-source digital electronic nose technology (smell inspector), has emerged as a promising alternative for rapid and non-destructive detection of plant viruses making it particularly suitable for large-scale screening of plants in agricultural settings. The smell inspector sensors detect volatile organic compounds (VOCs). In this study, the VOCs emitted by plants, representing their chemical signatures, indicate the presence of viral infections, allowing the differentiation between healthy and infected ones with high accuracy and sensitivity. Generally, AI allows the analysis of a large amount of data by considering many variables combined with each other. The simultaneous use of smell inspector and AI could provide real-time and on-site detection without the need for extensive sample preparation or specialized training. The potentiality showed by smell inspector devices for early detection of viral infections, could trigger a prompt intervention and establishment of disease management strategies to minimize crop losses. In this preliminary study, it was evaluated the application of digital electronic nose in combination with AI algorithm to detect tomato spotted wilt virus (*Orthotospovirus tomatomaculæ*) in tomato plants. Early disease detection is expected to be able to control the disease spread, to facilitate management practice, including standardization of detection protocols, optimization of sensor arrays and validation of results across different plant species and environmental conditions, and further to guarantee accompanying economic benefits.

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STRATEGIES FOR SUCCESSFUL MANAGEMENT OF CERCOSPORA LEAF SPOT AND RHIZOCTONIA ROOT ROT IN SUGAR BEET

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Cercospora leaf spot (CLS), caused by *Cercospora beticola*, and Rhizoctonia crown and root rot, caused by *Rhizoctonia solani*, are major diseases affecting sugar beet production in the USA and other sugar beet growing areas of the world. Cercospora leaf spot disease pressure has been increasing in the recent years partly due to varietal susceptibility, favorable weather conditions for CLS development, and widespread insensitivity of *C. beticola* populations to commonly used triazole fungicides such as tetraconazole and mefentrifluconazole. Fungicide application remains critical for effective management of CLS. Typical recommendation is to apply triazole fungicides in combination with a broad-spectrum tank-mixing partner. Based on the results from three years of field trials, we observed synergy among select tank-mix combinations. We have identified new tank-mix partners such as sulfur and phosphite that can enhance the efficacy of triazole fungicides for managing CLS. Currently, we are assessing the impact of these tank-mix partners on in vitro sensitivity of *C. beticola* isolates to these triazole fungicides. Rotation crops such as soybean, edible beans and corn grown in the rotation can also serve as hosts for *R. solani*. In fields with elevated Rhizoctonia inoculum levels, substantial stand loss, yield and quality losses can still occur on the moderately resistant cultivars. Several field trials were conducted to assess the effectiveness of at-planting (Succinate DeHydrogenase Inhibitor seed treatments or in-furrow) and postemergence fungicides. Seed treatments were very effective in protecting stands earlier in the season where as in-furrow fungicides were effective beyond the first few weeks. Application of postemergence fungicides as band and broadcast method were equally effective against Rhizoctonia. Integrated disease management utilizing resistant varieties, crop rotation and use of fungicides will be effective in managing these two important diseases. These recommendations are widely adopted by the sugar beet growers to produce a high-quality sugar beet crop.

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FRAC - WORKING TOGETHER GLOBALLY TO MANAGE FUNGICIDE RESISTANCE

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Resistance of agricultural pathogens against fungicides is a long standing and expanding problem for crop disease control. Effective resistance management is essential to ensure long term efficacy of fungicides for sustainable agriculture. Established in 1981, the Fungicide Resistance Action Committee (FRAC) serves as a specialist technical group within CropLife International, the association of crop protection companies and consists of different teams and publishes specific documents to address resistance management. The FRAC Mode of Action (MoA) Expert-Panel is continuously reviewing information on cross-resistance and MoA of new and established fungicides. The FRAC Code List and the corresponding FRAC Poster provide the latest classification scheme for antifungal active ingredients based on their respective MoA and cross-resistance pattern. This knowledge serves as basis for developing

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appropriate resistance management strategies. FRAC-Working-Groups, Expert-Fora and Task-Forces are platforms to regularly review available sensitivity monitoring data and to unanimously agree on resistance management guidelines for all fungicides within respective MoA/cross-resistance groups in relevant crops. Use recommendations for major fungicide classes are published on the FRAC website and are regularly updated. Dissemination and implementation of advice is done in efficient ways by FRAC regional and national organizations. Also, information is passed freely between FRAC and national action groups (FRAGs) including members of research institutions, advisory bodies and other experts involved. This forms a strong network in providing fact-based recommendations to growers and advisors.

P109

FITNESS, VIRULENCE AND FUNGICIDE TOLERANCE OF DIFFERENT FUSARIUM HEAD BLIGHT CAUSAL AGENTS

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Fusarium head blight (FHB) is an important cereal disease, caused by numerous fungal pathogens belonging to *Fusarium* genus. In this study, different *Fusarium* strains isolated from durum wheat and bread wheat in Central Italy were assessed for their fitness (conidial production, conidial germination rate, optimal growing temperature), their virulence and sensitivity to tebuconazole fungicides. The following species were evaluated: *F. graminearum*, *F. poae*, *F. avenaceum*, *F. culmorum*. Additionally, a new species recently isolated in Italy, *F. nodosum*, was included. Fitness parameters were determined by *in vitro* assays, while virulence was assessed through artificial inoculation on durum wheat and bread wheat seedlings, as well as through a fusarium head blight assay. Sensitivity to fungicide was evaluated through the determination of EC₅₀ (strains were grown in media amended with increasing fungicide dosages and the effective concentration reducing mycelial growth to 50 % was determined). The relationship among fitness, virulence and fungicide resistance was analyzed through linear correlation. The isolates were compared to the highly virulent control strains *F. graminearum* wt 3824, which exhibited the highest tolerance to fungicide (EC₅₀ = 0.03 % w/v) compared to the other strains isolated from the field. *F. nodosum* strains showed low virulence, suggesting that their role in FHB needs further elucidations. Our study highlights a relationship between virulence and fungicide tolerance. Given the importance of evaluating fungicide tolerance for crop and cereal production, further investigation following the contribution of this study is warranted.

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ETIOLOGY OF AN EMERGING ALMOND DECLINE SYNDROME IN INTENSIVE CROP SYSTEMS IN SOUTHERN SPAIN

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Almond (*Prunus dulcis*) crop has shift from traditional crop systems in marginal areas to an intensive farming system in regions with more favorable climatic and edaphic conditions, but this change has increased the incidence of diseases. Since 2016, gummosis, wood discoloration, decline, and progressive death of almond trees have been observed in intensive plantings in Andalusia region (southern Spain). Therefore, the main objective of this study was to elucidate the etiology of this complex disease to developed future studies on its epidemiology and on integrated disease management.

Surveys were conducted in 27 commercial almond plantations, from which more than 500 fungal isolates were collected. A total of 23 representative isolates were selected by a polyphasic approach based on multi-gene phylogenetic analysis together with recognizable phenotypic characters, such as colony, conidia or fruiting body morphology. Their pathogenicity was tested in almond plants of cv. Soleta by inoculating detached and/or attached shoots using the wounding and mycelial plug deposition method. The lesion length of each shoot was measured at three weeks or three months after inoculation, respectively.

Fungal species belonging to Botryosphaeriaceae, Diaporthaceae, Diatrypaceae, Nectriaceae, Pleurostomataceae, Togniniaceae and Cytosporaceae were identified, being Botryosphaeriaceae species the most frequent and aggressive. *Lasiodiplodia viticola* showed the highest canker lesions in detached shoots, followed by *Neofusicoccum mediterraneum* and *Diplodia corticola*. Likewise, *Di. corticola*, *N. parvum* and *N. mediterraneum* were the most aggressive in attached shoots.

We are facing a complex disease associated with a wide diversity of canker-causing fungi with different levels of frequency and aggressiveness. So far, the role that each of them may play in the disease cycle remains uncertain. Thus, it is necessary to determine the effect of the interaction between these fungal species, as well as the influence of abiotic factors in the development of the disease.

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WILT AND CROWN ROT OF ALMOND (*Prunus dulcis*) CAUSED BY *Fusarium oxysporum* IN SOUTHERN PORTUGAL

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In spring 2017, a severe outbreak of wilting and crown rot of almond cv. Lauranne grafted in Rootpac 20 rootstock was observed in a commercial orchard in southern Portugal. In this work, the etiology of the disease was determined through molecular, phenotypic and pathogenic characterization.

The diagnosis approach was first conducted by field observations and fungal isolation from the trunk and rootstock of the affected trees. Three representative *Fusarium*-like isolates were identified by phylogenetic analysis combining *tef1* and *rpb2* sequences. Fungal colonies and conidia were characterized on potato dextrose agar (PDA) and on Synthetischer Nährstoffarmer agar, respectively. Temperature effect on mycelial growth was evaluated on PDA from 5 to 35 °C. The pathogenicity of *F. oxysporum* isolates was conducted by inoculating one-year-old almond plants ('Lauranne') grafted on GF-677 or Rootpac 20 rootstocks.

The incidence of the disease was 100 %, but different degrees of disease severity were observed, ranging from partial wilting to death of trees. The trees showed wilting and leaf necrosis, gum secretion and canker formation in the trunk, and occasionally death. Internal symptoms were observed in an ascending direction from the root towards the trunk showing necrosis and xylem discoloration. Only *Fusarium*-like colonies were consistently isolated from affected tissues, and they were identified as *F. oxysporum sensu stricto*. The colonies showed floccose, abundant, white-violet mycelia. Microconidia were abundant, oval, elliptical or reniform, and usually 0-septate; and macroconidia falcate, thin walled, usually 3-septate. Chlamydospores were observed. The optimal growth temperature was 26.4 °C. A significant reduction in plant growth, wilting, and xylem discoloration was observed, with Rootpac 20 being more susceptible than GF-677. Plant infections were also reproduced using naturally infested soils.

The pathogenic role of *F. oxysporum* causing almond decline was demonstrated.

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EFFECT OF FUNGAL-CANKER PATHOGENS ON ECOPHYSIOLOGICAL PARAMETERS OF OLIVE TREE

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Olive branch dieback is primarily attributed to various fungal agents, notably from the genus *Neofusicoccum* within the Botryosphaeriaceae family. These fungal species induce cankers in lignified tissues, leading to branch dieback and irreversible damage. The significance of olive branch dieback in the Spanish olive crop gained attention in the early 2000s. The re-emergence of this disease can be attributed to several factors, including crop intensification, the prohibition of burning pruning residues, and periods of drought. Despite its importance, the tree's response to this disease remains understudied. This work investigates the impact of *N. mediterraneum* and *N. parvum* on 10 major Spanish olive cultivars. In a 13-year-old experimental orchard, 2-year-old branches were inoculated with both pathogens using mycelium plugs. Stomatal conductance and chlorophyll fluorescence were monitored every ten days, while leaf water potential was measured twice post-inoculation in five cultivars using a Scholander Camera. Disease severity, including growth reduction and lesion length, was also assessed. Additionally, each cultivar's specific leaf weight and wood density were determined. Both *Neofusicoccum* species significantly reduced vegetative growth and stomatal leaf conductance, 60 % and 28 %, respectively. Both pathogens exhibited similar virulence levels, but further investigation is needed to understand the impact of woody pathogens on plant ecophysiology.

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STUDY OF THE CAUSAL AGENT OF HAZELNUT BACTERIAL BLIGHT IN THE WEST PART OF GEORGIA

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The symptoms associated with Bacterial blight are presented in hazelnut nurseries in the west part of Georgia. *Xanthomonas arboricola* as a main causal agent of bacterial blight considered the most severe disease. Considering the significant decrease in yield, spreading diseases are recognized as a serious problem for local farmers.

The study aimed to detect *Xanthomonas arboricola* the causal agent of bacterial blight disease of hazelnuts in the west part of Georgia.

Within this project the visual observation of 2000 samples was carried out in two regions of Georgia: Samegrelo (Municipalities: Zugdidi, Khobi, Tsalenjikha and Senaki) and Guria (Municipality-Ozurgeti). Selected 538 samples with doubtful symptoms for *Xanthomonas arboricola* were collected for further laboratory testing.

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Georgian and Italian varieties “Anakliuri” and “Tonda di Giffoni” were tested on the *Xanthomonas arboricola* infection using Das-Elisa and TaqMan® triplex real-time PCR assays. Selective microbiological medias (yeast extract, peptone) were used to obtain pure bacterial culture. DNA from bacterial pure culture was extracted by DNA Purification Kit (Invitrogen™, USA).

It was revealed that *Xanthomonas arboricola* was the main causal agent of bacterial blight diseases in Hazelnut. 11.2 % of the tested variety “Anakliuri” was infected with bacterial blight, and comparatively low distribution was evaluated in the hazelnut cultivar “Tonda di Giffoni” (5.8 %).

Zugdidi municipality was shown to be the most infected region in Samegrelo with *Xanthomonas arboricola* (14.7 %) compared to the Guria region (9.3 %).

This study is important for the local farmers to monitor diseases and improve further management strategies.

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AN EFFECTIVE TECHNIQUE BASED ON TWO CRUCIAL PERIODS TO CONTROL KIWI BACTERIAL CANKER

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Bacterial canker caused by *Pseudomonas syringae* pv. *actinidiae* (*Psa*), is a most destructive disease in kiwi production worldwide. Control methods were mostly relied on chemical sprays such as copper and removing infected materials in period of high-incidence. However, it was inadequate in controlling the disease, often leading to trunk canker even orchards devastation. Therefore, we shift the strategy from “treatment-focused” to “prevention-oriented”. Our research revealed *Psa* could invade multiple host organs and moved systemically within vascular tissues. Cool temperature promoted *Psa* infection and spread quickly and above 24°C correlated with the decline of *Psa* populations and symptoms. Microbiome analysis showed *psa* population of kiwi trees fell to bottom in summer, then gradually rising up to peak in early spring, further elucidating the influence of temperature on annual epidemic dynamics and pathogen population fluctuations. We uncovered cool conditions exacerbate disease development by both suppressing ethylene-mediated resistance pathways and enhancing pathogen virulence. Based on the novel insights, we developed a technique based on two crucial periods to prevent pathogen invasion, colonization, and systemic movement within the host. Biological agents such as Zhongshengmycin, Kasumin, Polymyxins and developed Compound products were sprayed to the trunks twice at the period “After fruit-picking and before leaves-falling” to protect trees from bacterial ooze-induced mortality the following spring. Additionally, trees are treated twice “before and after florescence” to minimize *Psa* accumulation. This convenient approach, requiring only four treatments annually, dramatically reduces pathogen load in trees by 87.3 % and prevent the tree mortality rate from 30

% to less than 3 %. The system has been implemented nearly 80 % of kiwi-growing regions in China, offering an effective solution for managing kiwi bacterial canker. This work was supported by the National Key Research and Development Program of China (2022YFD1400200).

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REGULATION OF TYPE III SECRETION SYSTEM IN THE CAUSAL AGENT OF KIWIFRUIT BACTERIAL CANCER

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Kiwifruit Bacterial Canker, caused by *Pseudomonas syringae* pv. *actinidiae* (*Psa*), is a significant global threat to the kiwifruit industry. Currently, *Psa* is classified into five biovars (biovar 1,2,3,5,6). *Psa* biovar 3 (*Psa*3) is the most virulent biovar and the only one biovar in Chinese kiwifruit cultivation areas, despite lacking toxins. However, research on *Psa*3 is limited, highlighting the need for further investigation into the factors and pathways contributing to its pathogenicity. Through comparative genomics and molecular biology experiments, a conserved locus upstream of the *hrpR* gene in the Type III secretion system (T3SS) cluster was identified as crucial for full pathogenicity on kiwifruit and the elicitation of HR on non-host *Nicotiana benthamiana*. The T3SS is a key pathogenicity factors of *Psa*3, but their regulation and specific mechanisms are not fully understood. The two-component system RegAB was found to be a novel regulator of T3SS, with RgeA directly inhibiting the expression of key T3SS regulator genes, *hrpR* and *hrpS*, thereby controlling virulence in a T3SS-dependent manner. The DNA-binding affinity of RegA, and therefore its repressor function, is enhanced by phosphorylation. Additionally, c-di-GMP, PilZ domain-containing proteins, and the T6SS were identified as regulators contributing to the pathogenicity of *Psa*, probably via effects on bacterial competition, biofilm formation, and environmental adaptability. Moreover, a complicated relationship exists between T6SS and T3SS. While all known T3SS effectors play a critical role in plant-pathogen interactions, the specific effectors in *Psa* and their modes of action remain poorly understood. we shown that HopAU1 interacts with the Calcium-sensing receptor, HopZ5 interacts with GF14C, ultimately triggering plant immunity. Above all, these findings underscore the importance of T3SS and emphasize the varied signaling pathways and molecular mechanisms of T3SS regulators. This study was supported by the Natural Key Research and Development Program (2022YFD1400200).

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MANAGEMENT OF POWDERY MILDEW IN GARDENS; CAN NOVEL PRODUCTS HELP TO REDUCE DISEASE INCIDENCE?

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Powdery mildew is a common fungal disease affecting a wide range of ornamental and edible plants. Recently there has been an increasing preference for non-chemical methods of disease control, especially in gardens. Our research aims to determine whether different novel products (citric acid, phosphite fertiliser (a.i. potassium phosphite) and potassium bicarbonate) can reduce powdery mildew, and we compare them with the anti-mildew fungicide products widely available to gardeners to treat powdery mildew (Mildew Clear for Edibles- a.i. sulphur, and Provanto Fungus Fighter Concentrate- a.i. tebuconazole).

In May 2023 potted plants of the susceptible courgette cv. 'All Green Bush' were pot grown outside (Figure 1.). A powdery mildew resistant courgette cv. 'Ambassador' was chosen for comparison. Plants were inoculated under high humidity 6 days after treatment followed by two more treatments. Plants were assessed weekly for powdery mildew infection (percentage of plants infected and number of leaves affected), growth (weight of fruits), SPAD (leaf chlorophyll concentration) and chlorophyll fluorescence (Handy PEA), and treatments were compared.



Fig. 1. Early stages of powdery mildew infection 9 days after inoculation.

After three treatments over the three-week period a protective effect lasted for up to 2 weeks, with phosphite fertiliser, sulphur and tebuconazole causing significant reductions compared to non-treated controls. Phosphite fertiliser shows promise as a more environmentally friendly alternative to sulphur and tebuconazole giving reasonable reduction in powdery mildew infection when compared to non-treated controls. Interestingly 9 weeks after inoculation all plants had produced less fruit weight when compared to non-treated controls, apart from tebuconazole giving similar total weights.

Future work aims to examine the potential of phosphite fertiliser as a useful product for gardeners to manage powdery mildew.

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MELATONIN: THE ROLE IN PROTECTING RICE AGAINST BACTERIAL LEAF STREAK

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The Rice Bacterial Leaf Streak (BLS) caused by *Xanthomonas oryzae* pv. *oryzicola* (Xoc), is one of the most important rice bacterial diseases in China. Melatonin (MT) has a strong antibacterial activity. In our previous work, melatonin treated with rice could enhance rice plants against Rice Bacterial Blight (BB), caused by *Xanthomonas oryzae* pv. *Oryzae* (Xoo). In this study, we wanted to further investigate whether melatonin treatment in rice can enhance its resistance against rice bacterial leaf streak. Firstly, the chaff of rice seeds of Nipponbare was removed. Secondly, the rice seeds were disinfected in 70 % ethanol for 30 seconds, washed with sterilized ddH₂O for five times, and dried on sterilized filter paper for 10 min at room temperature. Thirdly, the rice seeds were placed on 2/1 MS medium amended with different melatonin concentrations (10 - 500 µg/mL) in an intelligent light incubator (light for 14 hours, dark for 10 hours, temperature at 1 ± 28 °C). After 7 days, the young rice seedlings were transplanted from 2/1 MS medium to a rice field. To test rice resistance, *Xanthomonas oryzae* pv. *oryzicola* strain RS105 was firstly activated on NA solid medium (beef extract 3g/L; sucrose 10 g/L; yeast extract 1g/L; peptone 5 g/L; pH=7.2 20g agarose powder added per 1L) at 28 °C. Single colonies were transferred to NA liquid medium (beef extract 3g/L; sucrose 10 g/L; yeast extract 1g/L; peptone 5 g/L; pH=7.2) and shaken to proliferate. The bacterial cells were then centrifuged and the concentration of RS105 was adjusted to OD₆₀₀=0.6 - 1.0. For pathogen inoculation, the final cell suspension was inoculated into the leaves of 7-week-old rice plants by leaf-needling method. The lesion size was measured 10 days after inoculation. Thirty leaves were inoculated for each Xoc strain in each treatment. The same experiment was performed three times. Rice leaves inoculated with ddH₂O were used as a negative control group. Rice leaves inoculated with RS105 in the absence of melatonin were used as a positive control. All rice plants grew well in the rice field. Compared to the control group, the lesion length was decreased when plants were treated with melatonin. The best performance was obtained at a concentration of 200 µg/mL. This work provides an easy way for inducing a far long-term resistance in rice by using melatonin treatment. This method is easy to operate, very effective, with a low investment cost. It can be applied in many fields such as agricultural production, scientific research, and environmental protection to reduce food losses caused by plant diseases.

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OLIGONUCLEOTIDE INSECTICIDES WITH LOW CARBON FOOTPRINT FOR GREEN PLANT PROTECTION

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Climate change creates favourable conditions for the growth of insect populations. Today, the world is seeing an increase in the number of insect pest infestations associated with a long-term increase in the average temperature of climatic systems. For example, local invasions of *Icerya purchasi* Maskell, a citrus pest recognized worldwide, have increased in size and number in recent years. Controlling this pest is complicated because not all chemical insecticides are effective, and their use is undesirable since citrus fruits are used for food and chemical agents cumulatively harm human health. We demonstrated the successful use of short antisense DNA fragment of the 28S ribosomal RNA gene called "oligoICER-11" (5'-ACA CCG ACG AC-3') to control cottony cushion scale, and we propose the use of green oligonucleotide insecticides with a low carbon footprint for large-scale implementation in agriculture and forestry. Using the contact oligonucleotide insecticide oligoICER11- at a concentration of 100 ng/μL on *I. purchasi* larvae resulted in a mortality of 70.55 ± 0.77 % within 10 days, while in thiamethoxam-treated group at a concentration of 0.8 g/L it comprised 35.33 ± 2.08 %. Application of the olinscides oligoICER 11 - was highly effective against *I. purchasi*, while the thiamethoxam had only a moderate insecticidal effect. Thus, the high efficacy of olinscides, along with their low carbon footprint during production (Table 1), make them attractive candidates for large-scale use to control insect pests without contributing to climate change. The research was funded by a grant from the Russian Science Foundation "Development of oligonucleotide insecticides for plant protection against insect pests from the suborder Sternorrhyncha (order Hemiptera) based on short antisense oligonucleotides of ribosomal genes" (project no. 20052-16-22).

Insecticides against <i>I. purchasi</i>	Corresponding compound	Reagent and solvents	t CO ₂ /t ratio
Synthesis of oligonucleotide insecticides (DNA insecticides, olinscides) on a solid-phase carrier on an automatic DNA synthesizer does not lead to greenhouse gas emissions			
Oligonucleotide insecticide (oligoICER-11)	Antisense oligonucleotide	Amidites, tetrazole, 1-methylimidazole, triethylamine, acetic or propionic anhydride, pyridine, iodine, acetic acid, trichloroacetic acid, dichloromethane, acetonitrile	~ 0
Synthesis of frequently used chlorothiazole neonicotinoid insecticides results in greenhouse gas emissions			
Chlorothiazole (Thiamethoxam)	Thiamethoxam	S-methyl-N-nitrosothiourea, methylamine, N-methyl-N'-nitroguanidine, formaldehyde, formic acid, tetrahydro-1,3,5-oxadiazine, 2-chloro-5-chloromethylthiazole, dimethylformamide, potassium carbonate	0.351

Table 1. Comparison of greenhouse gas emissions associated with the manufacture of reagents for the synthesis of oligonucleotide insecticides and chlorothiazole neonicotinoid insecticide against *I. purchasi*.

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THE ROLES OF BENZOXAZINOID IN WHEAT RESPONSE TO COMBINED ABIOTIC AND BIOTIC STRESSES

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Plants produce specialized metabolites to survive under hazardous environmental conditions and to minimize damage caused by insect herbivory. Benzoxazinoids (BXDs) are specialized metabolites produced by important cereal crops, such as wheat, maize, and rye. These metabolites are mostly known for their role in plant defense against herbivores. It was recently discovered that BXDs also accumulate under different abiotic stresses such as drought, high salinity, and cold conditions. However, the function of these molecules in drought tolerance remains largely unknown, and whether they play a dual role in combined drought tolerance and herbivore resistance. Here we show the potential roles of BXDs in bread wheat (*Triticum aestivum*) seedlings response to combined drought and *Rhopalosiphum padi* aphid stresses using several approaches: aphid bioassays, feeding behavior analysis, BXD levels, callose deposition, and gene expression. Under drought conditions, aphids showed poor fecundity, reduced body weight, and changes in feeding behavior compared to control. Differential expression of genes involved in an aphid's stress response, osmoregulation, defense response, detoxification, and olfactory system were also observed. Under combined or individual drought and aphid conditions, increased levels of BXDs were observed in the leaves, while their levels were reduced in the phloem sap of wheat seedlings. Interestingly, increased levels of both callose deposits and biosynthetic genes were observed in the leaves under applied stress conditions. In addition, BXD biosynthetic and MYB transcription factor genes were upregulated. Thus, we suggest that drought-induced callose might interfere with aphid feeding by triggering BXD accumulation and the MYBs might play a regulatory role in wheat response to combined stresses. To conclude, a deeper understanding of BXDs' function in wheat response to biotic and abiotic stresses could lead to better breeding strategies for stress tolerance in wheat.

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HIGH-THROUGHPUT SCREENING SYSTEM BASED ON BIOLUMINESCENCE REPORTER GENES TO SELECT THE PLANT ACTIVATORS IN ARABIDOPSIS THALIANA

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Plant activators (PAs) are a group of pesticides that can induce plant immunity to pathogens by activating plant defense systems. They are considered to be low environmental impact pesticides, because unlike conventional fungicides PAs induce disease resistance in plants without antibiotic activity.

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In addition, PA-mediated defense induction is also effective for control of viral diseases. Although PAs are considered to be ideal agrochemicals for plant disease management, the selection of compounds based on induction of plant defense systems requires tedious and time-consuming steps such as the analysis of plant defense related gene expression. To overcome the problem, we developed a high-throughput screening (HTS) system using luciferase bioluminescence reporter assay using 96- or 384- multiwell plates. The promoter from plant defense related genes are fused to the firefly luciferase gene (Fluc) and introduced into plants by Agrobacterium-mediated transformation. Transgenic seeds harboring promoter-Fluc fusion genes are germinated in the wells of multi-well plates and then treated with chemicals. Positive reactions are observed as the bioluminescence from the gene expression of the promoter-luciferase fusion gene in response to the chemicals with PA activity. As a result of estimating the reporter activity of several defense related gene promoters in transgenic Arabidopsis seedlings, we found that the Pathogenesis Related protein 1a from tobacco BY-2 and the Vegetative Storage Protein 1 promoter from *Arabidopsis thaliana*, show clear induction of Fluc activity in response to treatment with chemicals. Using the HTS system, we have identified several PA candidates that induce salicylate or jasmonate dependent defense gene induction pathways. To improve the *in planta* system for the HTS of PAs, we established the dual-color luciferase assay system using click beetle luciferase reporter genes.

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CHITOSAN NANOPARTICLES LOADED WITH JASMONIC ACID: AN INNOVATIVE APPROACH TO CONTROL BOTRYTIS CINEREA

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Nowadays, there is a growing need for environmentally friendly and sustainable compounds to manage plant diseases. Induction of defense responses in plants by elicitors is a promising eco-friendly strategy to combat plant fungal pathogens. Chitosan, a biopolymer with elicitation properties, can be formulated in nanoparticles. Here, we show that chitosan nanoparticles loaded with jasmonic acid (JA-CNPs) can significantly induce defense responses and reduce *B. cinerea* (Bc) incidence on *Arabidopsis thaliana* leaves. First, we evaluated phytotoxicity of JA-CNPs in *A. thaliana* ecotype Col-O by measuring root length, as an index of growth inhibition. It was found that at the concentration of 5 ppm JA-CNPs in the growth medium, a reduction of root growth at a maximum of 15.8 % was observed. To assess the induction of resistance, a GUS-staining experiment was carried out in *PDF1.2:GUS A. thaliana* transgenic line

at 10, 5 and 20 ppm JA-CNPs concentrations. The 5 ppm concentration displayed induction of the JA marker gene *PDF1.2*, that was remarkably upregulated after artificial inoculation of the leaves with a suspension of 10^5 conidia $\times$ mL^{-1} . *In-vitro* bioassays were carried out to test if different concentrations (0.5, 1, 5, 10, 50, 100 ppm) of JA-CNPs had a direct effect on conidial germination. No reduction in the conidial germination was observed on Col-O leaves, 18h after artificial inoculation with Bc. When 12-d-old Col-O plants were grown in solid medium embedded with 5 ppm JA-CNPs, disease incidence was reduced by 20.3 % 4 days after artificial inoculation with Bc. JA-CNPs spray application onto leaves of 30-d-old Col-O plants resulted in significant reduction of lesions growth by 47.7 % compared to the water control, when droplet inoculation was performed one day post treatment application. Overall, these findings demonstrate the potential of JA-CNPs to be used as new plant defense inducers against *B. cinerea*.

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SIRNACIDE: A NOVEL RNAI-BASED OOMICIDE AGAINST PLASMOPARA VITICOLA

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The oomycete *Plasmopara viticola*, also known as downy mildew or Peronospora, is a widespread pathogen in viticulture. As the pathogen becomes increasingly resistant to chemically synthesized active substances and the future application of copper formulations in viticulture will be limited, the efficient control of *P. viticola* poses new challenges for the agriculture.

The EUREGIO project SIRNACIDE aims to develop a new type of fungicide against *P. viticola*, which is based on the process of RNA interference. By using sequence-specific siRNA (short interfering RNA), target genes in various structures of the pathogen are silenced and the further growth of the organism will be prevented.

The identification of genes that are crucial for the survival of *P. viticola* is done by analyzing the gene expression during different life stages of the pathogen. Subsequently siRNAs targeting the most efficient genes are designed and synthesized. The siRNA is transported by vesicles of organic origin, which protect the siRNA from external environmental influences and enable the diffusion with the pathogen. A set of vesicles decorated with various substructures is prepared and their diffusion with *P. viticola* is checked via confocal microscopy using a fluorescent dye. In the target organism, the siRNA binds to the complementary mRNA of the pathogen, leading to its degradation and thus inhibiting the translation and formation of new proteins. Following the development of appropriate formulations, the efficacy of the novel solution is tested on potted grape vines via spray applications.

RNAi-based fungicides represent a promising alternative to chemical fungicide, however knowledge about this technology is still rare. The SIRNACIDE project provides further insights in the efficiency and applicability of RNAi-based compounds.

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USE OF FLUDIOXONIL TO CONTROL COBWEB DISEASE (CLADOBOTRYUM MYCOPHILUM) IN MUSHROOM CULTIVATION (AGARICUS BISPORUS)

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The fungal pathogen *Cladobotryum mycophilum* cause cobweb disease being a major threat to *Agaricus bisporus* mushroom production. The number of active substances authorized has decreased drastically lately, which is increasing the disease presence.

With the aim of finding active substances for the mushroom industry, efficacy trials were carried out with Geoxe[®] 50 WG (Fludioxonil 50 % w/w, Syngenta) in controlled infection trials.

Under the accreditation of Officially Recognized Trials, (EOR, Spain) a trial was performed with 5 different doses of Geoxe[®], one chemical control treatment (Prochloraz 46 % WP w/w, Sporgon[®], BASF) and water treatment control.

Treatment	Code	Dose (g/m ²)
Fludioxonil	F-A	0,5
Fludioxonil	F-B	0,3
Fludioxonil	F-C	0,2
Fludioxonil	F-D	0,1
Fludioxonil	F-E	0,05
Prochloraz	C	1
Water	W	1 litre

Mushroom compost phase II blocks, were placed on shelves in groups of 5 (total group surface 1 m²). Three replicates per treatment were done. Six days after casing, treatments were applied. A controlled number of *C. mycophilum* conidia (10⁶ sp/m²) was sprayed nine days after casing. The mushrooms were handpicked in two successive flushes. Total yields are represented in Figure 1. Best yield results were obtained with treatments F-B and F-C followed by treatment C.

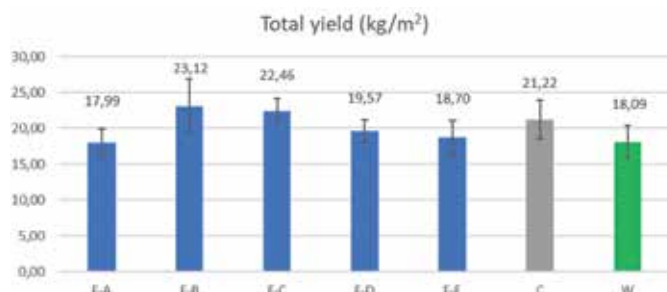


Figure 1. Total yield obtained per treatment at the trial (kg/m²).

Fungicide incidence was recorded by number of daily new infection spots found. Fungicide efficacy was calculated by Abbott's formula (Figure 2). All treatments except F-D gave better efficacy than the chemical control. In case of the highest dose applied (F-A) the efficacy was 87.50 %.

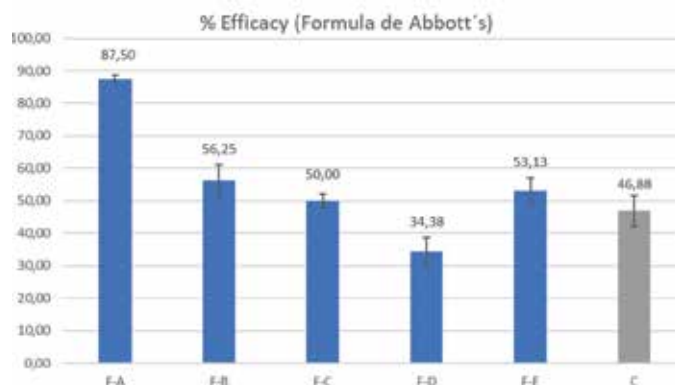


Figure 2. Fungicide efficacy (%).

Taking into account that the fungicide used as a control for this experiment (Prochloraz) has also been banned during this study, we believe that the results obtained are very important in the face of the lack of authorized active materials in mushroom cultivation.

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IN VITRO EVALUATION OF BLAD ACTIVITY AGAINST PHYTOPHTHORA CINNAMOMI

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BLAD, a 20.4 kDa, 173 amino acid residue polypeptide which inhibits fungal and oomycete growth, is the main subunit of BCO (Blad-containing oligomer), a *Lupinus* bioactive, 210 kDa polypeptide oligomer. *Phytophthora cinnamomi* is a major plant destructive pathogen with a wide range of hosts and is considered one of the main causes of *Quercus* spp. decline in the Southwest of the Iberian Peninsula. We selected five Portuguese isolates of *P. cinnamomi* (PH 13_98, PH 107, PH 194, PH 1247 and PH 4005) from three hosts (*Quercus suber* L., *Quercus rotundifolia* Lam. and *Castanea sativa* L.) to test the activity of BLAD/BCO in six different concentrations (0.2, 0.4, 1, 2, 3 and 4 g/L). Isolates were cultivated: a) in PDA medium, at 24 C ± 1 C, in darkness (colony radial growth was measured at 3, 5, 7, 9 and 11 days after the inoculation, in order to determine their radial growth rate); b) and in liquid culture (clarified V8 medium), at 24 C ± 1 C, in darkness (mycelial biomass dry weights were determined after 12 days of cultivation). We concluded that BLAD/BCO exhibits activity against *P. cinnamomi*, attaining 100 % inhibition for the higher concentrations tested in the five isolates, when the radial growth was evaluated, and a maximum of 97.8 % inhibition when the dry weight was evaluated.

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METAGENOME OF SOILS FROM FRUIT TREE ORCHARDS IN NE SPAIN AS A PREDICTIVE TOOL OF PHYTOSANITARY STATUS IN PERENNIAL AGROECOSYSTEMS

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A high percentage of agricultural soils in the European Union cope with degradation processes, mainly because of unsustainable management practices, such as intensive tillage, excessive fertilization, erosion, etc. Under this scenario, soil microbiological diversity is crucial for the productivity and sustainability of agricultural systems since microbial communities (prokaryotes and eukaryotes) support and modulate basic and essential services for their functioning. The abundance and population structure of the microbiome informs us about aspects such as the phytosanitary status of crops, the capacity for resilience against disturbances, or the presence/absence of microorganisms that allows the plant to cope with biotic or abiotic stresses. In this sense, it has been commonly accepted that detailed studies of the microbial diversity associated with different compartments (soils, rhizospheres, accompanying floras, etc.) are a very useful indicator of the sustainability and functioning of the whole agroecosystem. This study has characterized by means of high-throughput sequencing methods, the microbial biodiversity (fungi and bacteria) from soils of pome and stone fruit trees (apple, pear, peach, fig, plum and cherry) crops in different productive areas of the province of Zaragoza (NE Spain), in the frame of a broader study to evaluate some soil indicators related with its health and sustainability, including their phytosanitary status. The results obtained suggest that the microbial communities associated with the soil of the surveyed plots have, in general terms, a medium-high degree of biodiversity, taking as reference diversity values observed in studies of this type (bare soil of woody agroecosystems). Considering the fungal taxonomic composition globally, most of these soils presented a certain degree of microbiological exhaustion, defined by the major presence of soil-borne pathogens in relation to the rest of the saprophytic or symbiotic microorganisms characterized.

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METAGENOMIC INSIGHTS INTO THE GRAPEVINE ENDOPHYTIC MICROBIOME-PHAEOMONIELLA CHLAMYDOSPORA INTERACTIONS: IDENTIFICATION OF TAXA CORELATED TO HEALTH STATUS OF PROPAGATION MATERIAL

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In the metagenomics era, the development of next generation sequencing technologies has led to extensive investigations of the interactions between microbiomes and plant pathogens, in several pathosystems. In grapevine, a few studies have been conducted that associate the beneficial microbiome with plant health status, concerning grapevine fungal diseases. This study aimed to investigate the fluctuation of *Phaeomoniella chlamydospora* biomass from the nursery to the field and to explore alterations in the endophytic microbiome between vines with low and high pathogen concentration, throughout the propagation process. Quantification of the pathogen's biomass was carried out in three types of propagation material (canes, grafted unrooted and grafted rooted vines in various rootstock/scion combinations), using a dual-labeled probe assay. Subsequently, next generation sequencing was carried out in selected vines with low and high pathogens' concentrations. The qPCR assays showed that *P. chlamydospora* biomass was highly increased in grafted vines compared to canes, hypothesizing that the production process negatively affects the beneficial microorganisms present in the initial propagation material. Bioinformatic analysis including non-metric multidimensional scaling followed by differential abundance analysis in fungi and yeasts, revealed that several Amplicon Sequence Variants (ASVs) such as *Aureobasidium* and *Filobasidium*, two taxa widely used in biological control of plant pathogens, were more abundant in canes compared to the grafted rooted vines. Furthermore, *Clonostachys rosea* and *Filobasidium* were significantly more abundant in vines with low biomass compared to those with high concentrations. A specific pattern was observed regarding the bacterial microbiome, with *Pseudomonas*, being consistently more abundant in vines with low pathogens' biomass in almost all rootstock/scion combinations. These findings suggest that the propagation material production process highly impacts the composition of the grapevine microbiome and could stimulate research regarding the discovery of effective biological control agents and the development of microbial consortia against trunk pathogens.

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THE RHIZOSPHERE OF WILD TOMATO GENOTYPES IS ENRICHED FOR PATHOGEN-SUPPRESSIVE BACTERIA

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Crop domestication allowed humans to select for desirable agronomic characteristics and marked a major turning point in the human history. However, this artificial selection, which was accelerated during the last decades by intense breeding activities, resulted in most modern cultivars to exhibit low genetic diversity and thereby limited resistance to devastating diseases. Tomato is a major crop which relates to several wild *Solanum* species. Like many other domesticated species, this crop suffered a drastic genetic erosion which we hypothesized that also impacted on the composition and function of its microbiome. To test this, we employed plant-soil feedback experiments in which the soil was first conditioned with 99 core accessions (donors) belonging to 6 wild species and then tested for suppressiveness to the pathogens *Fusarium oxysporum* f.sp. *radicis-lycopersici* (*Foxrl*) and *Clavibacter michiganensis* subsp. *michiganensis* (*Cmm*), using in this second phase the cultivar *MoneyMaker* (MM) as recipient genotype. We found that genotypes belonging to the wild species *S. chilense*, inherit a soil legacy that enhances plant survival to *Foxrl* and *Cmm*. In particular, MM survival to *Foxrl* in *S. chilense*-primed soil was increased by 1.92-fold compared to MM-conditioned soil whereas the average *Cmm* disease index was reduced by 25 %. By employing culturomics, we found that soils primed with *S. chilense* accessions were enriched for *Foxrl*- and *Cmm*-suppressive bacteria the majority of which belong to the genus *Bacillus*. Synthetic communities of these strains were also effective in suppressing *Foxrl* and *Cmm* in planta. Collectively, our data demonstrates the impact of crop domestication on the functional potential of the root microbiome and further highlights the plant microbiomes as novel targets in crop breeding.

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VIRULENCE OF INTERSPECIFIC FUSANTS OF VERTICILLIUM SPECIES ON EGGPLANT

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Using parasexual recombination between strains which differ in virulence on a host plant species, we are continually investigating genomic regions determining the host range of *Verticillium* species. For this study, we attempted genetic recombination between *V. dahliae* and other *Verticillium* species to elucidate the genetic factors involved in virulence on eggplant.

Protoplast fusion between a strain of *V. dahliae* that is highly virulent on eggplant and less virulent on other species (*V. longisporum* A1/D3 or *V. alfalfae*) was performed. Antibiotic (Hygromycin B and G418) resistance genes were used as markers. Fungi having resistance on both antibiotics were screened as recombinants. Whether these recombinants carry chromosomes from each parental strain was resolved through assay by PCR using strain-specific DNA markers. The virulence of each recombinant on eggplant and other plant species was assayed by root-dipping inoculation.

Single spore isolates of most recombinants appeared to have the entire genome of both parental strains of each species. These "fusants" grew as well as the parental strains on agar medium. Genomes of both parental species were maintained after several months of culturing. In our earlier study of intraspecific recombination between strains of *V. dahliae*, karyogamy and subsequently haploidization occurred quickly after protoplast fusion. However, in interspecific combinations of this study, haploidization occurred only slightly. It is particularly interesting that all fusants between *V. dahliae* and *V. longisporum* lost pathogenicity on eggplant, although they were still pathogenic on okra. In contrast, fusants between *V. dahliae* and *V. alfalfae* were not only pathogenic on okra, but also on eggplant.

These results indicate that some genetic factor in the genome of *V. longisporum*, but not in the genome of *V. alfalfae*, suppresses pathogenicity on eggplant completely.

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REDUCTION OF VERTICILLUM DAHLIAE BY ONION EXTRACT: A BIOSTIMULANT AND ANTIFUNGAL EFFECT

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Verticillium wilt caused by fungus *Verticillium dahliae* is consider one of the most threatening diseases of olive trees, and is widely distributed in all countries of the Mediterranean basin. In recent years, *V. dahliae* has caused significant economic losses, severely affecting mainly

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irrigated plantation. Furthermore, the current climate emergency scenario has been related to a higher incidence of verticillium wilt. Therefore, the search for more effective and sustainable solutions to maintain productivity is a priority for the olive sector. The present work evaluates the biostimulant and antifungal activity of an onion extract standardized in organosulfur compounds (OE) in two experimental irrigated olive orchards in which the presence of *V. dahliae* was confirmed. To this end, leaf samples from control and OE-treated smallholdings were processed. The accumulation of malondialdehyde (MDA) and reduction of Fe^{+3} to Fe^{+2} were determined as stress markers. Detection and quantification of *V. dahliae* was carried out using qPCR. Additionally, weight, volume and moisture and fat content of the fruits were also analysed. Results suggest that the OE reduces lipid peroxidation, as a significant decrease in MDA was observed in the treated olive trees. Application of OE also reduced the severity of the infection, with a significant decrease in the fungal population of the leaf samples. Lastly, a direct correlation between the application of the extract and the fat percentage of the fruit was established. Even though further *in planta* trials are needed, it can be concluded that onion extract standardized in organosulfur compounds might be promising tool in the integrated control of Verticillium wilt.

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ALEUROCANTHUS WOGLUMI: MANAGING A POTENTIAL CLIMATE CHANGE-INDUCED BIOSECURITY THREAT

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Citrus blackfly, *Aleurocanthus woglumi* Ashby, is an agriculturally important polyphagous pest due to the economic losses it causes to citrus and many other plants. *A. woglumi* is found in tropical and subtropical regions but is presently unknown in Europe. As climate changes, the areas at risk from biological invaders shift and so it is important for biosecurity managers to be aware of the emerging threat patterns, as they institute phytosanitary measures. In our study, we used a published CLIMEX bioclimatic niche model to estimate the potential distribution and climate suitability patterns for *A. woglumi* under recent historical and medium-term future conditions. We incorporated data from three global climate models, namely ACCESS-CM2, CNRM-CM1-6, and EC-Earth3, considering a business-as-usual greenhouse gas emissions scenario (SSP3.7), and including a spatially explicit irrigation scenario by using the Global Map of Irrigated Areas. Under historical climate conditions, *A. woglumi* could potentially establish in very limited areas in southern European peri-coastal locations. The impact of irrigation in southern Mediterranean countries is significant, as it substantially increases the risk of *A. woglumi* establishment

if introduced, making citrus-growing countries serve as a potential hotspot for *A. woglumi*. Under the future climate scenarios, the potential range in Europe by the year 2050 extends further North and into higher elevations. Our findings emphasize the influence of future climate scenarios on the invasion risk posed by *A. woglumi* in Europe, shifting it from a concern to a threat. Therefore, adaptive biosecurity measures are essential, encompassing horizon scanning, enhanced targeted surveillance, periodic updates of risk assessments, and adjustments to regulations. Our results encourage the Mediterranean countries most likely to be affected, to scale up partnership and cooperation for prevention, control, outreach, and research activities.

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A NEW GREASY SPOT-LIKE DISEASE CAUSED BY ZASMIDIUM FRUCTICOLA WAS REPORTED IN SOUTHERN CHINA

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The price of citrus fruits is not only determined by the internal quality of the fruit, but also affected by the cosmetic quality. A good appearance quality could attract consumers' desire to buy. Recently a cosmetic defect caused by unknown agents was outbreak on Orah citrus (a hybrid) and Shatangju (Citrus reticulata Blanco cv. Shatangju) etc. in Southern China. In severely diseased orchards, the incidence of fruit exceeded 50 %, even up to 100 %, and severely infected fruits completely lost the marketability. Initially, black ink-like mold patches were firstly appeared on the fruit surface, the lesions gradually turned gray with the necrosis of infected peel. With the expansion of the fruit, radial cracks were occurred on the necrotic patched peels. The patches vary in size and could occur in any part of the fruit. As the symptoms looked like ringworm, the name of "white scaled blotch" was given by local formers. To explore the causing agents, multiple field investigations were carried out and samples were collected from more than 50 orchards in the Guangxi and Guangdong provinces during 2020 to 2023. The presence of mycelia and fungal spores on the necrotic blotches were demonstrated by scanning electron microscope, then the suspected pathogenic fungi was isolated from the infected pericarp, and its pathogenicity was confirmed with Koch's postulates. Based on morphology and phylogenetic analysis of the combined sequence data of LSU, ITS and rpb2, the pathogenic fungi were identified as some species in *Zasmidium*, including *Zasmidium fructicola*. Greasy spot-like symptoms were observed on leaves of Shatangju, but not on Orah citrus. The determination of etiology lays a foundation for the study of epidemiology and to guide prevention and control of this disease.

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THE COMPLETE GENOME OF *Stenotrophomonas indicatrix* REVEALS A NEW PLANT PATHOGENIC BACTERIUM IN MAIZE

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Stenotrophomonas indicatrix has been isolated from a wide range of environmental samples, including rotten fruit and the sunflower flower microbiota. During the last decade, yellow stripe symptoms with whorl on the apical meristem have been observed on leaves of maize grown between 2200 and 2600 masl. Recently, this bacterium has been reported as plant pathogenic on maize in Mexico. Therefore, in this research, we studied the complete genome of *S. indicatrix* to clarify its relationship with *S. maltophilia*, another bacterium recently reported as a causal agent of maize seedling rot. For this study, strains were isolated from symptomatic tissue and identified by phylogenetic reconstruction of the concatenated 16S rDNA and *gyrB* sequences. The pathogenic potential of these isolates was assessed by inoculating 6-week-old “chalqueño” plants with the injection method. After 15 days, symptoms of yellow stripe were observed at the base of the maize leaves, later whorling in the apical meristem. The most aggressive isolate was re-isolated for whole genome sequencing, which was performed using a NextSeq kit (300 cycles) on Illumina’s NextSeq platform, following the guidelines described by the manufacturer, resulting in 400 Mb of data (paired reads of 150 × 2 bp long paired-end reads) for each sample. The findings of this research provide information on the genomic functions of *S. indicatrix*, and it is the first genome sequencing of *S. indicatrix* as a plant pathogen, which highlights the importance of considering this new bacterium in maize breeding programs in Mexico. Also, this study provided the basis for future research on the management of bacterial diseases in maize.

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CITRUS PATHOGEN CONTROL BASED ON RNA INTERFERENCE INDUCED BY THE HOST PLANT

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The citrus industry has economic relevance for many countries around the world, even Brazil. However, this crop is affected by several plant pathogens, including fungi, such as *Colletotrichum abscisum*, responsible for the Postbloom fruit drop (PFD) disease. This pathogen causes the fruit drop at the beginning of its development, consequently, economic losses are associated. The fungus control is mainly based on chemical products, which have a high environmental impact and induce fungal resistance. Alternative methods such as gene silencing induced by interference RNA can be an interesting tool for pathogen control. Nevertheless, studies are required to guarantee the efficiency and safety for each

pathogen. Therefore, a *C. abscisum* mutant expressing DsRed fluorescent gene was used to analyze the gene silencing effect induced by citrus plants transformed with RNAi construction, via *Agrobacterium tumefaciens*. Successful citrus transformation was confirmed in 27 plants demonstrating the efficiency of the method. Transgenic plants were acclimatized at greenhouse conditions, and after flowering they were inoculated with fluorescent *C. abscisum* to evaluate the silencing by RT-qPCR. Among them, sixteen transgenic plants induced the fungus uptake and gene silencing of DsRed gene at different levels. These results indicate the potential use of Host-Induced Gene Silencing (HIGS) technology in citrus, due to RNAi delivery efficiency, and its probable application to other eukaryotic plant pathogens, decreasing the fungal resistance occurrence and environmental impacts.

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STUDY ON ITALIAN ONION CULTIVARS/ECOTYPES TOWARDS ONION YELLOW DWARF VIRUS (OYDV) INFECTION

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Onion yellow dwarf virus – OYDV (genus *Potyvirus*, family *Potyviridae*) is the most widespread potyvirus in onion (*Allium cepa* L.). Due to the severe symptoms, OYDV represents one of the most limiting biotic stresses of onion cultivation worldwide. Few information on resistant varieties and genetic source thereof is available. In Italy onion production is limited but it is represented by a wide diversity in terms of cultivar and ecotypes. The OYDV-susceptibility/tolerance of 14 Italian onion cultivars was assessed in two annual field trials, by rating symptoms severity and growth parameters, and by recording the occurrence of post-harvest secondary infections. The same onion cultivars were genetically characterized through SSR analyses, and the expression analysis of two genes belonging to the eukaryotic initiation factors (EIF) plant family genes, reported to be involved in potyvirus replication in several hosts, were performed to correlate the OYDV-susceptibility/tolerance to the different genetic profiles. Two susceptible and one tolerant onion cultivars were identified based on the symptom expression and the overall impact of virus infection on plant physiology. Although differences in growth parameters between infected and healthy plants of all cultivars were limited to the first-year trial, the OYDV-infection was correlated to a higher incidence of secondary infections in the post-harvest phase with an increase of water in infected bulbs compared to the healthy cultivars. SSR analysis identified separated clusters accounting for susceptible and tolerant cultivars; moreover, the isoform *EIF4e* iso1 showed higher levels of expression in OYDV/onion pathosystem in the cultivar resulted tolerant in

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this study. In conclusion, this study defines the genetic profile of Italian onion cultivars giving useful information about susceptibility and tolerance features that could be used in future studies addressed to identify resistance traits in onion *versus* OYDV. In addition, the OYDV-infection was correlated with the establishment of post-harvest secondary pathogens on bulbs during long-term storage, a crucial phase of onion production.

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IDENTIFICATION OF CONSERVED RECEPTORS FOR LEAF BLIGHT PATHOGENS IN WHEAT

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Bread wheat (*Triticum aestivum*) is a staple cereal crop and faces persistent threats from fungal leaf blight pathogens, significantly impacting global grain yields and quality. Among them, *Zymoseptoria tritici* which causes Septoria tritici blotch (STB) is considered to be one of the most destructive wheat diseases in Europe. Additionally, *Ramularia collo-cygni* causing ramularia leaf spot (RLS) is an emerging barley pathogen. Recent reports have found that *R.collo-cygni* could infect wheat also as an alternative host, posing new challenges. Interestingly, both fungi belong to the Dothideomycetes class and share notable similarities in some secreted proteins termed effectors. These small secreted proteins play a crucial role in overcoming the first line of plant defense known as PAMP-triggered immunity (PTI). Recent studies in other fungal pathogens suggest that these fungal effectors could target plant immune receptors, particularly pattern recognition receptors (PRRs), which are key players of the PTI. Indeed, a core fungal effector, NIS1, targeting the receptor-like kinase (RLK) signaling pathway, a PRR superfamily, has been identified.

In our study, we aim to identify conserved wheat receptors upregulated in response to *Z.tritici* and *R.collo-cygni* and identify any corresponding effectors that target these. To achieve this, candidate receptors have been identified from RNA sequencing of wheat leaf samples infected with either *Z.tritici* or *R.collo-cygni* at early time points. Any direct interaction between candidate receptors and effectors will be tested with yeast-2-hybrid experiments, followed by *in planta* validation through co-immunoprecipitation. Finally, silencing the receptors via virus-induced gene silencing (VIGS) will be coupled with disease assays.

Given the lack of effective control methods against STB and RLS, there is an urgent need for a better understanding of the interactions between wheat and these fungi. By identifying conserved effector wheat targets, our study aims to provide insights into resistance mechanisms and develop new control methods.

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TOWARDS IDENTIFYING THE PATHOGEN EFFECTOR RECOGNIZED BY THE WHEAT TANDEM KINASE WTK1

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Wheat yellow rust caused by the fungal pathogen *Puccinia striiformis* f.sp. *tritici* (*Pst*) reduces crop production. *Pst* secretes effector proteins into plant cells to facilitate pathogen colonization. This research focuses on the identification of *Pst* effector molecule associated with the yellow rust resistance gene *Yr15* derived from wild emmer wheat. *Yr15* encodes a unique protein comprising tandem kinase and pseudokinase domains, designated Wheat Tandem Kinase 1 (WTK1). According to the model developed by our team, the pseudokinase domain within the tandem kinase proteins serves as a decoy. It interacts with the pathogen effectors to counteract the suppression of receptor-like kinases by initiating a counter-offense mechanism. This mechanism is activated by tandem kinases, effectively "switching on" a defensive response.

To detect the Avr-Wtk1 effector, an approach involving chemical and UV mutagenesis is being employed. The mutagenized urediniospores are screened using wheat plants containing the *Yr15* gene. The genomic sequences of mutants will be elucidated and compared with those of the wild race to identify potential candidates.

Concurrently, we are pursuing an alternative strategy selected *Pst* candidate effectors and screening them in plant protoplasts for recognition by WTK1. Effector candidates from *Pst* were selected from the reference genome annotation and a differential expression analysis of all genes encoding secreted proteins during the infection cycle based on the following filtering criteria: i) presence of a predicted secretion signal peptide; ii) small peptides; iii) expression in haustoria; and iv) the presence of kinase docking motifs. We assume that the kinase docking motif interacts with the WTK1 pseudokinase domain.

Characterizing *Pst* effectors opens novel avenues for resistance breeding, allowing for targeted modifications and advancements in understanding rust resistance genes in wheat.

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GENOME-WIDE IDENTIFICATION AND CHARACTERIZATION OF NB-LRR GENES IN WILD OLIVE AND THEIR EXPRESSION IN RESPONSE TO VERTICILLIUM DAHLIAE INFECTION.

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Verticillium wilt of olive (VWO), caused by the phytopathogenic soil fungus *Verticillium dahliae* Kleb, is one of the most important and harmful diseases of the olive tree. The availability of wild olive (*Olea europaea* L. subsp. *europaea* var. *sylvestris*) germplasm banks is key for generating improved varieties. Its relevance has been demonstrated with *V. dahliae*, where the resistance to the defoliating pathotype has been evaluated in different collections of wild genotypes. Plants have evolved hundreds of resistance (R) genes to defend themselves against pathogens. Most of plant R genes encode proteins with the nucleotide-binding and leucine-rich repeat (NB-LRR) domains that interact with pathogen effectors to induce defense responses. Genome-wide identification and analysis of NB-LRR genes have been performed in several species. However, it is still unknown in wild olive. We have identified 202 putative NB-LRR genes in the wild olive genome, performed phylogenetic studies, as well as its chromosomal distribution, gene structure, protein motifs and gene ontology. The gene expression of NB-LRR genes in wild olive clones AC18 and AC15, previously shown as tolerant and susceptible to *D. V. dahliae*, respectively, reported a group of NB-LRR differentially expressed. Among them, we selected *OeuNB-LRR94*, whose expression was higher in the tolerant AC18 clone, and mainly expressed in the stem. Furthermore, it was observed that the expression of *OeuNB-LRR94* was induced in the tolerant AC18 clone upon infection with *V. dahliae*. Constitutive heterologous expression of *OeuNB-LRR94* in *N. benthamiana* resulted in an apparent increase of the resistance against the *V. dahliae* infection, but further research is required to disentangle the mechanism underlying this improvement of resistance. In summary, these data suggest that the NB-LRR gene family is a potential target for the genetic improvement of olive tree against verticillium wilt.

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RAPID SCREENING FOR RESISTANCE TO CONIOTHYRIUM GLYCINES THE CAUSE OF RED LEAF BLOTCH OF SOYBEAN USING DETACHED LEAF ASSAY

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Red leaf blotch, caused by the fungus *Coniothyrium glycines*, is a hugely important disease of soybean in Africa. The fungus is native to Africa and the disease can cause over 50 % yield losses. The use of host resistance provides among the most economical means to manage the disease. However, as there are no known sources of resistance, we are actively screening soybean germplasm for resistance. The whole plant method is laborious and requires large areas of space. The objective of the presented work was to evaluate the efficiency of a detached leaf assay, in lieu of screening whole plants. Soybean cultivars were grown under sterile conditions in pots in the greenhouse to supply healthy leaves. At two weeks after planting, the first trifoliate leaves were harvested, cleaned, and placed on moist filter paper in a 15 x 90 mm Petri dish or on moist paper towels in a 12 x 20 cm clear plastic 'clam shell' box. The leaves were spray inoculated with a spore suspension of *C. glycines* at 1.5×10^6 spores/ml. Two weeks after inoculation, lesion formation and disease severity were assessed, and results compared to corresponding whole plant assays. We noted highly comparable similarities between the lesion formation and severity on the detached leaves as occurred on inoculated whole plants. In addition, the fungus could infect, sporulate, and maintain pathogenicity on the detached leaves. We conclude that the detached leaf assay under laboratory conditions will improve the efficiency of screening for higher throughput soybean germplasm screening.

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SCREENING COMMON BEAN GERMPLASM FOR RESISTANCE TO TWO MAJOR DISEASES IN UGANDA

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Angular leaf spot (ALS) caused by *Pseudocercospora griseola* and Southern blight caused by *Sclerotium rolfsii* are important diseases of common bean in Uganda. Losses due to both diseases may exceed 60 %. Smallholder farmers often fail to control these diseases due to resource limitations. We envisage that providing resistant bean varieties will lead to sustainable management of the above diseases.

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We aimed to study the field diversity of *P. griseola* and identify resistance sources among advanced breeding lines. Additionally, we sought to identify sources of resistance to Southern blight among a diverse germplasm.

We planted 100 lines (advanced breeding lines from CIAT, ALS differentials and local checks) in farmers' fields in an area of high ALS infection and assessed disease at pod initiation and pod filling. We screened 143 lines for *S. rolfii* resistance in the screenhouse using 2 strains with varying virulence. ALS and Southern blight were assessed using the published scales of 1 to 9 and 1 to 6, respectively.

Several lines including Mexico54 (resistant parent) succumbed to ALS. However, lines such as NUA 184, SEQ10006 and AFR735 were resistant to ALS (scores of ≤ 3.0). Eleven lines from the Southern blight nursery at NARO Uganda, 2 from the CIAT Root Rot nursery in Kawanda Uganda and the Pythium root rot resistant parent RWR719 were resistant to *S. rolfii*. However, no differences were observed in the reaction of the lines to the highly and less virulent *S. rolfii* strains.

Likely sources of resistance to Ugandan ALS races identified by the current study need to be validated in additional locations. There is a high chance of identifying durable sources of resistance to Southern blight in our germplasm. However, the observed resistances need to be characterized before deployment in breeding programmes.

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DIVERSITY OF PSEUDOKINASES AMONG PLANTS

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Pseudokinases play diverse roles in organisms, from scaffolding for signaling complexes and modulating kinase activity to acting as molecular switches. Plant pseudokinases participate in essential functions including the regulation of growth, response to stress, and immune signaling pathways. The current approach of discriminating between functional and pseudokinase domain sequences is based on the presence of a handful of conserved catalytically crucial residues. This simplification is inadequate and fails on approximately 10 % of experimentally confirmed pseudokinases, including CASK, WNK1, and Titin.

Here, we propose an improved method to identify pseudokinase domains and discriminate them from functional kinases. We curated a dataset of over 70 experimentally validated pseudokinases, including nine plant-derived proteins and 18 structures with resolved X-ray structures. Using this dataset, we created a structure-oriented multiple sequence alignment (MSA) and a dedicated Hidden Markov Model (HMM) to enhance the sensitivity of pseudokinase domain searches. We also used the obtained MSA for training an interpretable machine learning (ML) classifier to distinguish between functional and non-functional kinase domains and gain insight into sequence

determinants of pseudokinases.

We apply our methodology to plant kinases assembled across over a hundred plant species to gain insights into plant kinome complexity. This methodology advances pseudokinase identification and enhances our understanding of their roles in plant biology. This methodology can inform new research directions in plant adaptive mechanisms and signaling pathways.

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TEMPERATURE EFFECTS ON SYMPTOM EXPRESSION OF BLACK ROOT ROT DISEASE CAUSED BY *BERKELEYOMYCES* SPP

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The black root rot pathogen *Thielaviopsis basicola* was recently reclassified into two species: *B. basicola* and *B. rouxiae*. Generally, diseases caused by these pathogens occur during cool seasons. However, black root rot of lettuce caused by *B. rouxiae* usually occurs during hot seasons in Japan. For this study, the relation between temperature and symptom expression of black root rot disease on lettuce and other plants was investigated.

Mycelial growth of black root rot pathogen was investigated on PSA medium at 35–5°C. Seedlings of lettuce (*Lactuca sativa*), okra (*Abelmoschus esculentus*), cotton (*Gossypium arboreum*), and tobacco (*Nicotiana benthamiana*) were inoculated by endoconidial suspension (10^6 spores / mL) of black root rot pathogen. Inoculated plants were cultivated at 15, 20, 25, and 30°C for a month. The disease severity of each plant was evaluated on a scale of 0–4.

The optimum temperature for mycelial growth of *B. rouxiae* and *B. basicola* on PSA medium was 25°C. Inoculation tests of *B. rouxiae* on lettuce showed that root rot symptoms became severest at 25°C. Therefore, optimum temperatures for pathogen growth and symptom expression were consistent for lettuce. However, symptoms on other plants became severest at lower temperatures of 20–15°C. In those cases, the optimum temperature for symptom expression did not accord with that for pathogen growth. In *B. basicola*, the optimum temperatures for symptom expression on each plant species were similar to those of the sister species *B. rouxiae*, although no symptom was observed on lettuce or tobacco. Results demonstrate that the optimum temperatures for symptom expression on plant species are probably determined not only by the pathogen characteristics but also by those of the plants.

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MOLECULAR CHARACTERIZATION OF SPORE-FORMING AIR AND SURFACE BACTERIA AND STUDY OF THEIR PATHOGENICITY ON POTATO TUBERS

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Post-harvest diseases are caused by pathogens that may occur during harvesting, handling, storage, marketing and distribution. The presence of aerospores in cold storage facilities can cause bacterial diseases in stored products, limits their storage time and affects their organoleptic properties. Our study focused on characterizing bacterial spores present in two potato warehouses in Tunisia. A total of 110 bacterial isolates were established and were tested in vivo for their pathogenicity on potato tubers at room temperature and under storage conditions at 4°C. 69 % of the tested bacteria exhibited pathogenic properties. Molecular analysis through amplification and partial sequencing of the *rpoB* and 16S rRNA genes of the most devastating species belonged to the *Bacillus* genus. Strains of "*Pantoea agglomerans*," "*Panearthrobacter*," and "*Bacillus subtilis*" were described for the first time as pathogens of potatoes. The pathogenic isolates were able to produce plant cell-wall degrading enzymes such as, pectinase, cellulase, protease and others enzymes such as amylase and gelatinase. Virulence genes encoding amylase (*Amy α*), chitinase (*chi*), cellulase (*EG*), and sporulation protein (*spoVG*) were detected by PCR. This study identified the potato pathogens during the storage phase and elucidated the key virulence factors involved in the pathogenicity of airborne spores collected from the air and surfaces.

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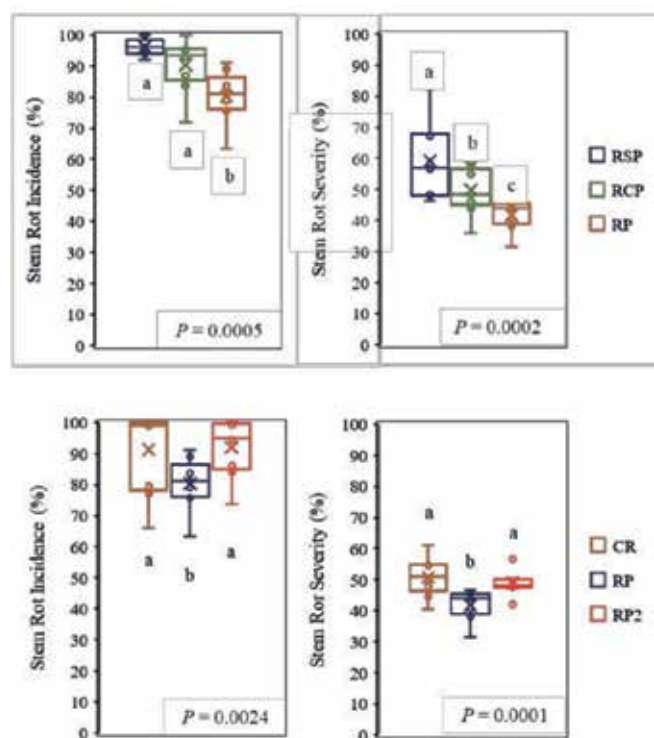
EFFECTS OF PASTURE LENGTH AND STUBBLE ON SOILBORNE DISEASES AND GRAIN YIELD OF RICE IN RICE-PASTURE NO-TILLAGE ROTATIONS.

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Rice production in Uruguay is historically based on rotations with pastures. Market demands have led to changes to intensify the traditional rice-pasture (RP) system (two years of rice followed by three-four years of pasture) by shortening pasture phases, and planting on stubble from the previous crop. Soilborne rice diseases (stem rot and sheath spots) are among the main yield limiting factors in intensified systems by producing sclerotia that remain viable for years in soils. The objective of this study is to assess the effects of intensification by pasture length reduction and cultivation on rice stubble on the occurrence of soilborne diseases and the impact on rice yield. Using four rotations and RP as control, two comparisons were made in a long-term field experiment during 2022-2019. First, rotations with 1.5 (RSP), 2.5 (RCP) and 3.5 (RP) years of pasture before rice were compared

to investigate the effects of pasture length on disease occurrence and rice yield. Stem rot severity was increased in RSP and RCP, compared with RP and yield was lowest for RSP in two years and for RCP in one year of the study. Second, a contrast was performed by comparing continuous rice (CR) with the first and second rice (RP2) of RP used to investigate disease occurrence and rice yield responses to cultivation on rice stubble. Stem rot occurrence was increased in CR and RP2, and sheath spots in CR, compared with RP. The yield was lower for CR in three years, and RP2 in two years compared with RP. Reducing a single year in the pasture length and the cultivation of rice on stubble favors the development of diseases that can affect the rice yield in these systems. Additional management practices for disease control and yield improvement should be implemented in situations of crop intensification.



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PHYLOGENETIC ANALYSIS OF LETTUCE BLACK ROOT ROT PATHOGEN REPORTED IN JAPAN

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In 2016 Japan, lettuce black root rot disease caused by *Berkeleyomyces rouxiae* occurred suddenly. For this study, we investigate pathogenicity on lettuce and genetic relations of various *B. rouxiae* isolates including pathogens of lettuce and other plants to elucidate the reasons for black root rot of lettuce occurrence in Japan.

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The pathogenicity of Japanese isolates of *B. rouxiae* on lettuce was assayed by root dip inoculation. Whole genome sequences of the two Japanese isolates were analyzed to develop microsatellite markers. Phylogenetic analyses of Japanese and American isolates of *B. rouxiae* were conducted using these microsatellite markers. A network diagram was constructed.

The results of inoculation assays indicate that Japanese isolates from non-lettuce plants are nonpathogenic on lettuce. Although *B. rouxiae* is known as a polyxenic pathogen, the host range of each isolate appears to be different. Phylogenetic analysis revealed that Japanese isolates which are pathogenic on lettuce are related closely to one another, and are distinct from Japanese isolates from non-lettuce plants. These results indicate that the occurrence of lettuce black root rot disease in Japan possibly was not caused by isolates which have been known as the pathogens of non-lettuce plants in Japan before occurrence of lettuce disease. Lettuce black root rot disease has also occurred in the United States. Therefore, we investigated the relationship between Japanese and American isolates. Black root rot pathogen, known as *Thielaviopsis basicola* in the past, was reclassified recently into two species: *B. basicola* and *B. rouxiae*. For this work, the American isolates were identified as *B. rouxiae*, similar to Japanese isolates. However, we found that Japanese and American isolates of the lettuce black root rot pathogen are phylogenetically different. This result indicates that the occurrence of lettuce black root rot disease in Japan is probably unrelated to the American pathogen.

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ENSURING THE PHYTOSANITARY QUALITY OF BEANS, TROPICAL FORAGES AND CASSAVA COLLECTIONS IN THE "FUTURE SEEDS" GENE BANK: REFERENCE STRAIN BANK

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The Germplasm Health Unit of the Bioversity International Alliance and CIAT (GHU-CIAT) ensures that the germplasm of beans, cassava and tropical forages conserved and distributed nationally and internationally by the Germplasm Genebank, and breeding programs are free of quarantine risk pests and diseases. As part of routine activities, GHU-CIAT detects the presence of phytopathogenic fungi using morphological and molecular techniques. In order to ensure the quality and traceability of the results, the GHU-CIAT has created a reference strains bank with those quarantine fungi that can historically be detected in the germplasm evaluated under laboratory conditions. Initially, fungal isolates were identified and characterized morphologically by using conventional taxonomic keys, including photographs of macro and micro morphological features. Additionally, a protocol for DNA extraction was standardized and using Oxford Nanopore Technology, isolates were sequenced and molecularly characterized. Using this methodology,

40 isolates were obtained corresponding to the following genera: *Colletotrichum* sp., *Phoma* sp., *Alternaria*, *Curvularia* sp., *Bipolaris* sp., *Pestalotiopsis* sp., and *Fusarium* sp. Finally, the morphologically and molecularly characterized strains were conserved under controlled temperature and luminosity conditions. Having this kind of reference strain ensures the quality of the results obtained in the diagnosis of phytopathogenic fungi and works as a quality control mechanism which to evidence and demonstrate the accuracy in the identification and detection of quarantine fungi. This study strengthens the traceability for the validation of the diagnostic method and additionally provides information about the genetic variability of the different species detected.

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A SURVEY FOR OOMYCETES INCIDENCE IN NURSERIES AND GARDENS

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Diseases that are caused by Oomycetes and in particular *Phytophthora* species in nurseries are a worldwide problem and a direct threat to biodiversity, as *Phytophthora* can move readily through the nursery supply chain via infected still asymptomatic plants. *Phytophthora* genus with more than 180 species and a wide range of plant hosts can cause a great number of devastating plant diseases with *P. cinnamomi*, a highly aggressive species, parasitizing ca. 5.000 plant species.

A survey on symptomatic and asymptomatic plants in nurseries and gardens was conducted in Greece from 2021 to 2023 to determine mainly the prevalence, distribution, and diversity of *Phytophthora* sp in these environments. A plethora of species were isolated from 34 plant genera in a total of 2,607 rhizosphere and irrigation water samples. Tested plants originated from Greek and Italian nurseries and from gardens in Attica region, Greece. 763 oomycetes strains were isolated using baiting assays and microscopic observations. Based on morphological features, 129 isolates were selected for further identification. 11 different *Phytophthora* sp. and 1 hybrid species (*P. lacustris* x *riparia*) were identified (99 strains tested) based on internal transcribed spacer (ITS), Beta-tubulin (B-tub) and cytochrome oxidase c subunit I and II (COXI and COXII) sequences. Additionally, 25 isolates were identified as *Phytophthora vexans* and 2 as *Ph. litorale* while 3 strains were verified as different *Pythium* species.

Phytophthora nicotianae was the most dominant species (35.36 %) isolated from the rhizosphere of 20 different hosts. *P. virginiana*, *P. parsiana*, *P. lacustris* and hybrid *P. lacustris* x *riparia* were isolated from the irrigation water samples. These species were not detected in the rhizosphere samples. Motif analysis of the ITS region of *P. cinnamomi* isolates, showed differentiation in specific DNA patterns with *P. mediterranea*. Risk analysis based on specific hosts, environments and other plant species in gardens was performed.

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GRAPEVINE TRUNK DISEASES MANAGEMENT: IN VITRO EVALUATION OF A NEW FORMULATION BASED ON EXTRACTS OF YEAST CELL WALL

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Grapevine Trunk Diseases (GTDs) are a group of diseases caused by wood-colonizing fungal pathogens that induce necrosis and discoloration, vascular infections, and white wood. After the banning of some fungicides with good control capability towards GTDs, no effective control measures have been developed. The aim of this study was to elucidate the mechanism(s) of action of a new liquid formulation based on cell wall yeast extract (YE) of *Saccharomyces cerevisiae* against some GTD agents [*Neofusicoccum parvum* (Np), *Eutypa lata* (El), *Fomitiporia mediterranea* (Fomed) and *Phaeomoniella chlamydospora* (Pch)]. All the selected fungi have been inoculated at the centre of Petri dishes containing PDA added with YE (0.25, 0.50, 1.00% v/v). Potato Dextrose Agar without YE has been used as control. Mycelium has been measured every three days starting from 24 h after the inoculation until the colony reached the edge in control plates. The inhibition by YE of Np mycelial development has been also evaluated in liquid cultures by using a conidial suspension (10^3 conidia mL⁻¹). Every two-three days, spore germination and colony formation have been evaluated. Our results showed the ability of YE (regardless of all concentrations) to exert a strong inhibition on mycelium growth of all the tested pathogens (-86, -85 and -81 % compared to the controls, in the case of Np, Fomed and Pch). The highest YE concentration tested completely prevent El-mycelium growth and Np-conidia germination. Based on this first screening on the growth of some important GTD pathogens, results suggest that the YE strain could have an antifungal direct effect against these important pathogens.

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SUSCEPTIBILITY OF DIFFERENT GRAPEVINE VARIETIES TO ESCA DISEASE IN VINEYARDS IN MONTENEGRO

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Esca is a very complex and geographically widespread grapevine trunk disease. Research carried out from 2021 to 2023 confirmed esca disease in the most important wine-growing regions in Montenegro. A survey of 20 vineyards was conducted during the period of intense symptoms appearance (June/July), and typical symptoms were found in 17 vineyards (85 %), while in three vineyards (15 %) no symptoms were

observed. In symptomatic plants, chronic and apoplectic drying of vines was noticed with typical symptoms of necrotic leaf tissue bordered by a clear purple line, forming the so-called "tiger stripe" pattern. Cutting followed by examination of the wooden trunk tissue revealed black dot necrosis in cross sections, and black necrotic lesions and white rot of xylem vessels in longitudinal sections. In such vineyards, empty spaces formed by removal of the drying vines were also noted. In vineyards up to 10 years old, the average disease intensity incidence was 8-6 %, while in some older vineyards the disease incidence was up to 30 %. Esca disease has been observed in all vineyards older than five years, and the disease incidence was significantly increased with the vineyard age. Moreover, the autochthonous Montenegrin wine varieties, Vranac and Kratosija, showed a significantly higher sensitivity to esca disease compared to the introduced varieties Shiraz, Cabernet Franc, Cabernet Sauvignon, Merlot, Malvasia and Marselan. Vranac and Kratosija manifested symptoms in all examined vineyards older than five years, in contrast to the introduced varieties, where only Merlot and Cabernet Sauvignon showed symptoms in 20 % and 14 % of examined vineyards, respectively. Furthermore, table varieties Cardinal, Ribier, Victoria and Afus Ali also showed a high degree of susceptibility to the disease. A three-year monitoring revealed esca disease in the most important Montenegrin wine-growing regions. The obtained results indicate the great economic importance of this disease in Montenegro.

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CAN PULSED LIGHT BE USED TO CONTROL GRAPEVINE DISEASES?

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The cultivated grapevine (*Vitis vinifera* L.) is a perennial woody fruit crop susceptible to numerous diseases, such as mildews and gray mold. To fight against these diseases, repeated applications of phytosanitary products are generally carried out. However, their use has led to an increasing awareness of potential risks associated with them. Nowadays, there is a growing interest in employing alternative methods, such as those based on stimulation of plant defenses (elicitors), use of biopesticides, or even physical control. In this project, we investigate the possibility of using pulsed light technology as alternative solution for controlling grapevine diseases. Pulsed light technology is described as producing a spectrum ranging from UV to infrared. Historically used for decontaminating medical equipment and industrial surfaces, this technology is now being developed for decontaminating post-harvest products. To study if the technology can be suitable for application in plant cultivation concerning disease control, different spectral profiles were firstly defined. Their effectiveness was examined on grapevine to reduce the development of major grapevine pathogens, including downy mildew. The experiments were conducted in laboratory,

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greenhouses, and vineyards. After the determination of the optimal spectra, we will explore the impacts of this light treatment on grapevine physiology and on primary and specialized metabolisms through metabolomic approaches. The effects of this method will also be studied at the pathogen level.

This project is conducted in partnership with a major regional player providing agronomic solutions to agricultural enterprises. An industrial partner well-established in the field of lighting is also involved in the project. We aim to propose an environmentally friendly and effective control solution to reduce the use of phytosanitary products in viticulture.

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OZONATED WATER: A NOVEL STRATEGY FOR CONTROLLING GRAPEVINE TRUNK PATHOGENS DURING GRAPEVINE'S PROPAGATION.

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Grapevine Trunk Diseases (GTDs), caused by various fungal pathogens, are a significant threat to viticultural production worldwide. Managing GTDs involves multiple factors, such as the pathogen, geographic location, climate, grape variety, and agricultural practices. The propagation of grapevines in nursery houses is a major source of disease spread globally. This study explores the effectiveness of ozonated water (2.5 ppm) as an alternative to conventional fungicide (Switch 37.5/25 WG, Syngenta, Greece) in the grapevine propagation process. Specifically, scion and rootstock materials were artificially inoculated with a conidial suspension (2×10^4 conidia mL^{-1}) of *Phaeomoniella chlamydospora* (*Pch*) and *Dactylonectria torrescensis* (*DacT*). The nursery house's propagation protocol (VNB Bakasietas, Nemea) was followed, with the propagation plant materials (scion and rootstock) subjected to ozonation during rehydration steps, while the control group followed the conventional protocol using fungicide. The resulting grafted plants were planted in commercial vineyards, and after six months, various plant growth parameters (dry matter, stem, and root length) were evaluated, along with disease severity via absolute quantification of pathogens using RT-qPCR (Real-Time quantitative PCR). Overall, plants treated with ozonated water exhibited higher growth, with longer roots and stems compared to those treated with fungicide. Moreover, in plants generated from artificially inoculated propagation materials, higher presence ($p < 0.05$) of *Pch* and *DacT* was detected in the fungicide treatment compared to the ozonated water treatment. Specifically, the mean quantity of *Pch* and *DacT* in the ozonation treatment was 3.94 and 4.14 copies μL^{-1} , respectively, while in the fungicide treatment, the quantity was equal to 10.73 and 6.23 copies μL^{-1} , respectively. These findings highlight the benefits of ozonated water treatment, showing improved plant growth characteristics and lower pathogen levels compared to traditional fungicide use, thereby offering a promising and environmentally friendly solution for enhancing vineyard health and productivity.

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BIOLOGICAL CONTROL OF MYCOTOXINS ON MAIZE USING ENDOPHYTIC BACTERIA

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One of the most important consequences of consuming food and feed contaminated with mycotoxins, is harmful effects in both humans and mammals. Aflatoxins and fumonisins, are products of the secondary metabolism, which mainly produced by the species *Aspergillus flavus* and *Fusarium verticillioides* respectively, and they are the most important mycotoxins in maize. Due to the absence of safe chemical means to deal with the spread of these fungi in corn derivatives and feed, as well as means to reduce or/and inhibit mycotoxins contamination, research into alternative eco-friendly solutions to protect grains has been imposed and attracted wide attention from researchers worldwide. In this effort, 10 endophytic bacterial strains isolated from different medicinal plants and olive trees with remarkable antagonistic activity against several phytopathogenic fungi, were evaluated for their ability to inhibit the production of mycotoxins by the aforementioned fungi. Initially, *in vitro* experiments were conducted in Petri dishes using dual culture technique to examine the competitiveness of bacterial isolates against the mycelial growth of two toxigenic fungi belonging to *A. flavus* and *F. verticillioides* species. Subsequently, *in situ* experiments were carried out on corn kernels to further investigate their efficacy against the reduction of aflatoxins and fumonisins produced. Finally, the most effective bacterial strains, were evaluated in maize plants under field conditions. The quantitative analysis of mycotoxins was performed by the enzyme-linked immunoassay ELISA method and the results of the abovementioned experiments will be presented.

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BIOLOGICAL CONTROL OF AFLATOXINS IN MAIZE USING ENDEMIC NON-AFLATOXIGENIC ISOLATES OF ASPERGILLUS FLAVUS

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Aflatoxin contamination of food and feed, produced by *Aspergillus flavus*, is emerging as a significant food safety hazard in Europe. The use of chemical fungicides for mycotoxin management is becoming progressively debated and subject to regulations, prompting the exploration of alternative environmentally friendly strategies. *A. flavus* non-aflatoxigenic strains emerge as the most promising method currently available for mitigating aflatoxin accumulation. This study investigated the aflatoxin-producing potential of 99 endemic *A. flavus* isolates, collected from rotten corn kernel samples from Thessaly and Macedonia during 2016-2014. Initially, all strains underwent assessment for aflatoxin production using thin-layer chromatography (TLC). Among them, 31 isolates were identified as aflatoxigenic,

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while the remaining 63 as non-aflatoxigenic. Subsequently, the 63 non-aflatoxigenic *A. flavus* isolates were tested *in vitro*, for their ability to suppress aflatoxin production when co-cultured with two highly aflatoxigenic strains, BAF47 and AAF24. TLC results demonstrated that overall, 38 non-aflatoxigenic isolates exhibited the ability to inhibit, either partially or completely, aflatoxin production from both toxigenic strains. The inhibitory capacity of the most effective non-aflatoxigenic *A. flavus* strains was later evaluated in maize kernels, *in situ*. ELISA results indicated that all isolates reduced aflatoxin levels, in rates between 62 % to 98 %. Furthermore, the non-aflatoxigenic *A. flavus* isolates were clustered into vegetative compatibility groups (VCGs) to evaluate their suitability to the local environment. This analysis resulted in their classification into 23 distinct VCGs, predominantly concentrated within four major groups. Finally, 35 non-aflatoxigenic *A. flavus* strains were evaluated in maize field conditions. 11 isolates presented significant reductions in aflatoxin contamination in the range of 94 % up to 100 %, compared to the control. Notably, among these, 6 strains belong to the 4 main VCGs. Based on these results, certain native non-aflatoxigenic strains could potentially be employed as biocontrol agents to regulate aflatoxins, aiding in an integrated strategy to safeguard maize crops in Greece.

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TRANSCRIPTIONAL REPROGRAMMING EVENTS REVEALED VARIETY-DEPENDENT RESPONSES OF GRAPEVINE BERRIES UPON INOCULATION WITH *ASPERGILLUS CARBONARIUS*

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This study allows to gain insights into the transcriptional reprogramming of grapevine berries upon *Aspergillus carbonarius* challenge. Three time points after the pathogen inoculation were employed to determine the expression profiles of the induced genes across two grape varieties, Fraoula (red) and Victoria (white). In parallel, we also investigated the responses of these two varieties to the fungus in terms of their susceptibility, and ochratoxin A (OTA) production. Global gene expression profiles and pathways enrichment analysis, based on an RNA-seq approach, revealed an extensive repertoire of differential transcriptional responses between these varieties across the inoculation period. Notably, defense-related genes were induced in a cultivar- and time-dependent pattern. Furthermore, our results showed a differentiation between these two cultivars regarding the quantification of the pathogen and OTA accumulation. By accurately deciphering the transcriptomic mechanisms underlying grapevine responses to *A. carbonarius* infection, this work allows to expand our knowledge at this plant-pathogen interaction, contributing to the development of targeted strategies to enhance grape quality and safety against this necrotrophic mycotoxigenic fungal pathogen.

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BIOLOGICAL CONTROL OF FUMONISINS IN MAIZE USING EPIPHYTIC BACTERIA AND YEASTS

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Maize (*Zea mays*), a crop essential for the production of food for human and animal consumption worldwide, is repeatedly targeted by many pathogenic mycotoxigenic fungi whose presence often results in mycotoxin contamination. Fumonisins -secondary metabolites produced by *Fusarium verticillioides* and *Fusarium proliferatum* – can cause acute or chronic diseases with various toxic effects to both human and animal health including cancer. Fumonisins' contamination is an ever-increasing problem which makes control methods necessary, not only for the economy but also for the benefit of human and animal health. Fumonisin B1 is the most common mycotoxin found in food products, especially in maize. The focus of this study was to evaluate six epiphytic bacteria isolates and two yeasts, shown before that can reduce aflatoxin contamination in maize, in terms of their ability to inhibit fumonisin production by *Fusarium verticillioides*. Based on molecular identification, two of the bacteria classified as *Klebsiella pneumoniae*, three of them as *Pantoea agglomerans* and one of them as *Enterobacter cloacae*. The two yeasts were classified as *Aureobasidium pullulans* species. *Fusarium verticillioides* growth rate was calculated in the presence of bacteria and yeasts in dual culture *in vitro* assays. The results indicated inhibition rates of mycelial growth in rates reaching 40 %. Subsequently, *in situ* experiments on maize kernels, fumonisin B1 levels were inhibited in rates above 70 %; *Pantoea agglomerans* and *Aureobasidium pullulans*, were the species demonstrating the highest inhibition rates. In order to evaluate their effectiveness in reducing fumonisin levels under field condition, the aforementioned species were further applied in a maize field. Field testing proved that solely the bacteria demonstrated around 40 % inhibition rates of fumonisin A contamination. The yeasts were not able to decrease fumonisin levels. In conclusion, the results from the current study, will be a useful tool for the enrichment of biological control methods for the control of fumonisins in maize.

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LOCALIZATION SYSTEM OF OLIVE TREES INFESTED BY *XYLELLA FASTIDIOSA* THROUGH SATELLITE IMAGES

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Xylella fastidiosa is a phytopathogenic bacterium, associated with serious diseases in crops of great economic importance, such as olives. There are no effective control strategies, thus if an outbreak occurs, eradication measures have to be taken to limit the spread of the disease. The aim of this study is

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to create a methodology for the early detection of olive trees infested by the bacterium through satellite images. Both healthy and diseased plants have specific spectral traits depicted through vegetation indices. These indices allow us to detect physiological and biochemical disorders of plants. Three vegetation indices have been selected: 1. normalized difference vegetation index (NDVI), 2. modified anthocyanin reflectance index (mARI) and 3. modified chlorophyll absorption in reflectance index (MCARI). Time series charts are generated using the values of these abiotic stress indicators of the trees, which aid in defining the time at which the bacteria first infected the plant. Besides, using the high-resolution satellite images, the canopy of olive trees is categorized into four groups according to the severity of the disease: 1. healthy trees, 2. trees suffering from plant stress, 3. trees whose canopy has begun to dry out and 4. trees that have completely dried out. This approach concludes in a web-based application that clearly visualizes the results, indicating an early stage of bacterium infection before the symptoms are visible in the olive trees. The specific methodology is implemented in pilot fields in olive groves of Messina and Zakynthos. The early detection of the bacterium is extremely important for the application of the appropriate measures in time, reducing their negative consequences and increasing the yield of the agricultural holdings.

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BIOVEXO, A BBI-JU-H2020 PROJECT ON BIOCONTROL OF XYLELLA AND ITS VECTOR IN OLIVE TREES FOR INTEGRATED PEST MANAGEMENT

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Xylella fastidiosa is increasingly causing diseases in olive trees and various other crops in the Mediterranean region. It has wiped out a number of olive and almond groves in Italy and Spain in only a few years. Unfortunately, the climate of the southern European Union is ideal for *Xylella*. Due to its rapid transmission across cultivation areas, *Xylella fastidiosa* is projected to cause yield losses of 70 % - 35 % in olive harvests and 13 % in almond harvests. Currently, there are no products available on the market that have been proven to be effective against *Xylella fastidiosa*, which is spread by xylem-feeding insects – notably the spittlebug *Philaeus spumarius*. Several chemical insecticides are authorized to control xylem-feeding insects; while for products allowed in organic farming, only temporary authorizations, for limited periods, have been obtained.

11 partners from 5 different countries (industries, SMEs, RTOs, and universities) aim to develop environmentally sustainable and economically viable plant protection solutions that can be deployed as a method of integrated pest management to control *Xylella fastidiosa* and its vector.

After 3 years of BIOVEXO, both V-biopesticides and X-biopesticides have been upscaled and formulated and are currently being applied via endotherapy, foliar spray, or

drip irrigation on large scale field trials (olive and almond trees) as preventive and curative approaches, and also in integrated management, in Italy (Apulia) and Spain (Alicante and Mallorca). The biopesticides' mechanisms of actions are also being investigated. Toxicity and life cycles of each biopesticide are also being assessed.

At the end of BIOVEXO, the two best performing solutions will be brought closer to the market (TRL 8-7).

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ONION EXTRACT: BIOSTIMULANT EFFECT IN OLIVE PLANTLETS UNDER CONTROLLED CONDITIONS AND IN VITRO ANTIBACTERIAL ACTIVITY AGAINST XYLELLA FASTIDIOSA

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Olive trees dominate the landscape of the Mediterranean basin since it is one of the species best adapted to the Mediterranean climate and soil. Nevertheless, the effects of climate change in the Mediterranean basin poses substantial impact to olive trees, since it has been related to a higher incidence of endemic and emerging diseases. Among these, *Xylella fastidiosa* stands out as the main threat for this crop. It is therefore crucial to develop new strategies that increase the resilience of the olive trees against it. The present work aims to evaluate the biostimulant activity of an onion extract standardized in organosulfur compounds (OE), applied both by foliar spray and by irrigation, in 6-month-old olive plantlets in growth chambers. To this end, root and leaf samples from control and treated plantlets were processed. Four different stress markers were determined: accumulation of malondialdehyde (MDA), reduction of Fe³⁺ to Fe²⁺, amino acid content and expression of stress response genes. The length of the roots and the number of leaves were also determined. Additionally, in vitro tests were carried out to evaluate the antimicrobial activity of OE against *X. fastidiosa*. Results suggest that OE increases the reducing capacity while it reduces lipid peroxidation, as a significant decrease in MDA was observed in the treated olive trees. Greater root development was observed in olives treated by irrigation. Lastly, OE inhibited the growth of *X. fastidiosa* in agar diffusion test. Even though further in planta trials are needed, it can be concluded that onion extract standardized in organosulfur compounds is a promising tool to increase resilience of olive trees in an integrated pest management policy, and specifically against *Xylella fastidiosa*.

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INVESTIGATION OF THE INTERFERENCE OF OLIVE LEAF MICROBIOME IN MOLECULAR DETECTION OF XYLELLA FASTIDIOSA

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Performance characteristics of the real-time PCR method of Harper et al. (2010 erratum 2013), recommended in EU phytosanitary legislation as a screening method for detection of *Xylella fastidiosa* (Xf), include analytical specificity evaluated based on its inclusivity using various Xf strains and its exclusivity using a series of non-target known phytopathogens. The EPPO Xf-diagnostic standard reports no cross-reactions for this method, but mentions that late Cq values may occur, and such cases are inconclusive. To clarify such cases, the Laboratory of Bacteriology of Benaki Phytopathological Institute (LB-BPI) investigated, in the frame of the 'Olive Routes' nationally funded project, whether microorganisms other than Xf or known phytopathogens, commonly found in olive leaves, interfere in this method producing late Cq values. To this end, the microbiome present in eleven leaf samples of olive trees (six cultivars) grown under different soil and climate conditions was determined by 16S/ITS amplicon sequencing (Novogene, UK) on genomic DNA from these samples. This NGS technique revealed great diversity in bacterial and fungal genera present in the microbiome, in which those with the highest frequency included: 32 bacterial genera or groups and 35 fungal genera. An *in silico* PCR (Primer-BLAST) was performed to check whether the primers/probe set of the Harper et al. method could hybridize to the above-identified bacterial and fungal genera, focusing on products ≤ 1000 bp due to PCR conditions employed. Also, *in silico* melt curve sequence analysis of these products showed distinctive difference between the peak of Xf and that of the other microorganisms, with the exception of the genus *Bacteroides* in which case agarose gel separation for size-based discrimination would be needed. These results indicate that melt curve analysis could be useful in clarifying late Cq values rapidly and reliably; research is ongoing in the LB-BPI to optimize this method and solve inconclusive cases.

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SEARCHING FOR OLIVE GENOTYPES RESISTANT TO QUICK DECLINE SYNDROME

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The olive quick decline syndrome (OQDS) is a severe vascular disease caused by *Xylella fastidiosa* subsp. *pauca* (Xfp), a Gram-negative, quarantine bacterium for the European Union. It first appeared in 2013 in a grove in the Salento peninsula, Apulia region, southeastern Italy, and quickly spread northward. Currently, the pathogen has affected over 40 % of the Apulian territory, hosting to more than 21 million olive trees. Utilizing resistant olive varieties is the most effective strategy to mitigate the disease's impact. Extensive surveys

were conducted in the infected areas, where OQDS has decimated nearly 90 % of olive plants, to identify genotypes displaying resistance. Seventeen olive genotypes, exhibiting minimal or no symptoms of quick decline syndrome, were monitored for Xfp load over the past six years. Samples were collected in April and October and subjected to qPCR detection following accepted protocols. Some genotypes remained uninfected, while others displayed low Xfp loads or declining trends. Ten SSR markers were utilized to genotype a subset of the tested plants, comparing allelic profiles with 244 olive cultivars from the Mediterranean basin, including Putatively Resistant (PR) genotypes, to identify synonyms (LRM analysis) and explore phylogenetic relationships (Unweighted Neighbor-Joining analysis). Additionally, a paternity test was conducted to ascertain the parentage of unknown genotypes. LRM analysis revealed one case of synonymy and high genetic similarity between P4_C and Proto 12 with the resistant cultivar Leccino. Phylogenetic analysis placed the tested samples close to resistant cultivars Leccino and FS17, as well as some PR genotypes, indicating significant genetic affinity. Notably, sample P4_C appeared to derive from self-pollination of the Leccino cultivar. Some samples showed parentage with the Apulian cultivar Rotondella and the Albanian cultivar Kalinjot oval, closely related to the resistant Leccino. Furthermore, certain samples originated from autochthonous cultivars, underscoring the importance of conserving them to identify Xfp-resistant genetic material.

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EXPLORING THE EFFECTS OF XYLELLA FASTIDIOSA INFECTION ON FOUR OLIVE CULTIVARS THROUGH COMPARATIVE TRANSCRIPTOME ANALYSIS

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Over the past decade, the bacterium *Xylella fastidiosa* subsp. *pauca* (hereafter referred to as *X.f.*), has radically changed the agriculture and landscape heritage of Salento (Apulia, Southern Italy). Several efforts have been made to control and limit the spread of the disease, highlighting the need for a better understanding of its etiological mechanisms. The aim of this project was to identify candidate genes putatively involved in resistance or susceptibility to the bacterium. Therefore, a comparative transcriptome analysis of four olive cultivars known to have different susceptibility to *X.f.* was performed: Leccino and Donna Francesca as tolerant and putative tolerant respectively, and Oliva Rossa and Cellina as susceptible. The four cultivars were grown in a certified greenhouse at DiSSPA of the University of Bari, and three replicates of both mock-inoculated and *X.f.*-artificially infected plants were analyzed

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for each. Total RNA from the leaf and peduncle tissue was used for the synthesis of RNA-seq libraries which were sequenced on the Illumina Novaseq 6000 platform. The two resistant varieties showed genes with differential expression compared to the two susceptible varieties following infection. Data analysis revealed a complex gene expression pattern with more than 800 differentially expressed genes, including several defense-related genes. *X.f.* altered the expression of fewer genes in resistant varieties than in susceptible ones. To date, putative candidate genes identified by RNA-seq analysis are being validated by qRT-PCR. Their identification could highlight metabolic or signaling pathways involved in resistance/ susceptibility to *X.f.* and be useful for further molecular breeding and functional genomics approaches.

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FAUNISTIC STUDY OF AUCHENORRHYNCHA SPECIES IN OLIVE ORCHARDS IN GREECE, WITH INSIGHTS IN THE POTENTIAL VECTORS OF XYLELLA FASTIDIOSA

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Xylella fastidiosa Wells et al. is a gram-negative bacterium, that causes economically significant diseases, including Pierce's disease of grapes, citrus variegated chlorosis (CVC), olive quick decline syndrome (OQDS) in olives, and it is transmitted only by insect vectors of suborder Auchenorrhyncha. It is a quarantine pathogen for Europe, and it was first detected in olives in Italy in 2013 and after that, it was reported in other countries. Since there is the imminent threat of bacterium to invade and expand in Greece, studies of species composition, abundance and seasonal appearance of Auchenorrhyncha in Greek olive orchards are of paramount importance especially that of potential vectors. For this reason, extensive biodiversity studies have undertaken in olive orchards where Malaise traps have been installed and examined in a monthly basis for one year. Moreover, sampling with sweep net was undertaken from the weeds to find not so mobile, hidden species, and to correlate them with host plants. Most dominant species were mainly Typhlocybae species, while many genera of Deltocephalinae were constantly appearing even in small numbers. Further details about the results and extensive discussion are given in the presentation.

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REAL TIME LAMP DETECTION OF XYLELLA FASTIDIOSA IN OLIVE TREES USING THE MEMBRANE-BASED METHOD AND STORED PLANT MATERIAL

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Xylella fastidiosa (Xf) is one of the most harmful bacterial pathogens, that is also responsible for the Olive Quick Decline Syndrome reported in Apulia region, Italy. Real time LAMP is one of the diagnostic assays recommended by EPPO PM24/7

() for the detection of this pathogen using fresh plant material. The latter represents a limit for analysing a great number of samples in a short period of time. Therefore, this study aims to improve real time LAMP using the printed membrane-based extraction method and compare the analysis of printed membranes stored at room temperature with that of plant material stored at 4°C and -20°C.

Stem sections of Xf-infected olive cv Ogliarola were printed onto nitrocellulose membranes. Discs of printed membranes were subjected to rapid nucleic acid extraction and subsequent isothermal amplification using the commercial ICGene PLUS ONE system with the LAMP kit *Xylella fastidiosa* (Enbiotech Srl, Italy). All samples were also analyzed simultaneously by real time PCR. The presence of an endogenous internal control allowed to verify the efficacy of the extraction and amplification of the samples. The same stem portions were analyzed after one month of storage at 4°C and after 5 months at -20°C. The results of real time LAMP were equivalent to those obtained by real time PCR. Regarding the analysis of stored plant material, all samples were Xf positive at 4°C after 1 month and at -20°C after 3 months. Only 2 out of 3 samples were Xf positive after 5 months of storage. In the case of processed printed membranes, all samples showed positive results in the studied period. The membrane-based real-time LAMP method has proven to be easy to use for the extraction step and efficient for long-term storage of samples before analysis. However, for storage of plant material, the period for analysis can be extended to 1 month for fresh and 3 months for frozen material.

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IDENTIFICATION OF SENSITIVE 'WILD TYPE' STRAINS OF PUCCINIA HORDEI AND PUCCINIA TRITICINA ACROSS EUROPE THROUGH AIR SAMPLING

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Maintaining food security worldwide requires effective crop protection. Growers are constantly facing new challenges in fungal pathogen control, namely the reduction of fungicides available for use and constantly evolving pathogens. Strategies are in place to extend the effective use of existing fungicides, such as fungicide application plans and fungal pathogen resistance monitoring. Dwarf rust of barley (*Puccinia hordei*) and brown rust of wheat (*Puccinia triticina*) result in production losses worldwide and are an ongoing threat to cereal production. The Fungicide Resistance Action Committee (FRAC) has classified rust fungi with a low risk for resistance development. However recently Cyp51 mutations and differential expression levels were found in European wheat leaf rust samples, which may lead to resistance to demethylase inhibitor fungicides. Also, recently through rust populations from the UK and France the mutation I187F was reported in subunit SdhC (SdhC-187F). This mutation resulted in a 10 - 5 factor increase in resistance to succinate dehydrogenase inhibitor fungicides. It is there of utmost importance to remain vigilant to resistance development. To gain a better understanding of mutation and resistance development of rusts in barley and wheat across Europe,

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EpiLogic GmbH will harvest rust fungal isolates across Europe using a car mounted air spore trap. The resultant isolates will be screened for mutations, and bioassays will be performed to monitor and compare ED_{50} values. Using correlation analysis between mutation absence/presence and ED_{50} value, we plan to find sensitive 'wild type' strains, necessarily required for a concrete estimation of resistance development in populations of a pathogen. Thereby gaining a better understanding of adaptation to fungicides and mutation presence for dwarf rust of barley and brown rust of wheat across Europe.

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RESISTANCE RISK OF PHYTOPHTHORA COLOCASIAE TO AZOXYSTROBIN IN SOUTHEASTERN OF CHINA

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Qols has been applied to manage taro leaf blight caused by *Phytophthora colocasiae* in southeastern of China for many years. The risk of *P. colocasiae* to Qol and the potential resistant mechanism remains unknown. In this study, the *P. colocasiae* strains sampled from southeastern of China was investigated. The results showed that the azoxystrobin resistant frequency reached 86.5 %, indicating prevalence of resistance in the field. Analysis of the *cytb* fragment showed that only one nucleotide variant in this population, producing two haplotypes. The nucleotide variant leads to codon change at 425 (GGT to GCT) producing A142 (alanine) and G142 (glycine) in Hap_1 and Hap_2 strains, respectively. Further fitness related features showed that no significant difference in temperature sensitivity, mycelial growth rate, sporangia production, zoospore release and aggressiveness between azoxystrobin-resistant and sensitive strains indicating no potential fitness cost for azoxystrobin resistance. Taken together, azoxystrobin resistance need to be taken into consideration to manage taro leaf blight in southeastern of China.

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EUROPEAN MUTATION MAP OF ALLELES CONFERRING RESISTANCE TO COMPLEX III INHIBITORS, ZOAXAMIDE, FLUOPICOLIDE AND OXATHIAPROLIN IN PLASMOPARA VITICOLA POPULATIONS

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Plant diseases can result in important yield losses of agricultural crops each year worldwide. Downy mildew, caused by *Plasmopara viticola*, is one of the most devastating diseases in the common grapevine *Vitis vinifera*. Management strategies of downy mildew include fungicides with different modes of action like single-target fungicides, such as QoI (Quinone outside Inhibitors, e.g. azoxystrobin), QoSI (stigmatellin binding type, e.g. ametoctradin), and more

recently, complex III inhibitors Qil's (Quinone inside Inhibitors; e.g. cyazofamid and amisulbrom), are often introduced in management program. Increasing use of fungicides treatments lead to emerging of resistance and compromise grapevine protection against downy mildew. Monitoring of fungicide resistance in grapevine downy mildew, is one of the strategies supporting disease management decisions. Fungicide sensitivity is carried out in laboratory using bioassays and/or molecular analysis. New molecular technology called long read sequencing by Oxford Nanopore Technologies (ONT) is used to detect and quantify target modification in cytochrome b gene conferring resistance to complex III inhibitors. In this study, samples of downy mildew are collected across Europe by EpiLogic GmbH using a car mounted air spore trap. Then, samples are analysed by CONIDIA CONPHY using a specific ONT amplicon sequencing method. *P. viticola* genes of cytochrome b, β -tubulin, ATPase and Oxysterol Binding Protein are sequenced to detect and quantify alleles (mutations and insertions) conferring resistance to complex III inhibitors, zoaxamide, fluopicolide and oxathiapiprolin respectively. Combining an efficient way of sampling with an innovative method of sequencing give the first and complete **Mutation map** of resistance against fungicides used to control downy mildew in Europe. This unique tool enables the quantification of multiple mutations present on the same gene (multiple resistant phenotypes). This is the first step of a deployment on a wide range of pathogens of vineyards and cereals to manage fungicide resistance at European level.

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FUNGICIDE RESISTANCE RISK SCORE INDEX APPLIED TO DOWNY MILDEW CONTROL IN TUSCANY (ITALY).

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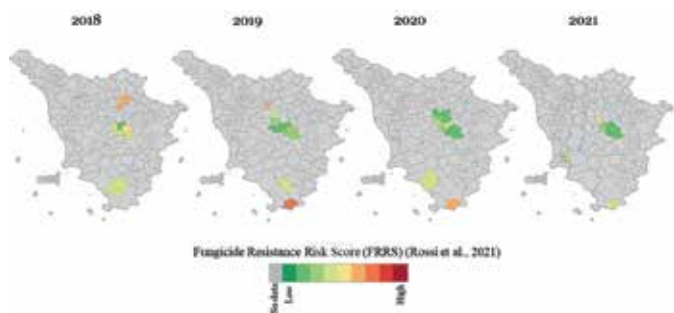
Plasmopara viticola is a high-risk pathogen for fungicide resistance development according to Fungicide Resistance Action Committee (FRAC). Given the well-documented challenge of fungicide efficacy loss in agriculture, anti-resistance strategies should be applied to limit the emergence and/or spread of resistant strains. This study aimed to analyze the risk of fungicide resistance using the Fungicide Resistance Risk Score (FRRS) index developed by Rossi et al. (2021).

For the analysis, a fungicide with medium-high resistance risk development was considered. Italian winegrowers' logbooks from 2018 to 2021 provided data on: i) number of treatments and applications with the same product, ii) dose applied, iii) products sequence, iv) mixture or solo application. Weather data were collected from automatic weather stations and disease pressure index was calculated accordingly. Data were managed to calculate the FRRS for each vineyard, as $FRRS = BRS \times IRS$. BRS is the Behavioral Risk Score, which considers how the product is used by farmers, while IRS is the Intrinsic Risk Score, which evaluates the environmental conditions, pathogen and fungicide characteristics.

FRRS calculated for 89 vineyards across 16 counties in Tuscany, are reported in Figure 1. Green areas represent the lowest risk, while red areas indicate the highest, suggesting

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potential deviations from label prescriptions by farmers or favorable environmental conditions for disease development. Moving from 2018 to 2021 a general improvement can be observed in Tuscany where only one municipality was green in 2018. In the following years the number of green municipalities increased, indicating an improvement in the product usage and, consequently, a reduction in the risk of resistance appearance.



FRRS index may be a useful tool to monitor the risk of resistance development and could be integrated into phytosanitary bulletin to provide alerts to farmers/advisors. Further steps will be the index validation, by combining known resistance strains presence and the riskiest conditions.

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RESISTANCE OF BOTRYTIS CINEREA FROM GREENHOUSE-GROWN TOMATO IN SERBIA TO QoI FUNGICIDES

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Chemical control is the main disease management strategy against gray mold, caused by *Botrytis cinerea*, in greenhouse-grown vegetables worldwide, but the problem lies in pathogens tendency to develop fungicide resistance. Resistance mechanism to QoI fungicides involves mutations in the mitochondrial cytochrome *b* gene. The aim of this study was to provide insights into Serbian *B. cinerea* populations resistance to QoI fungicides.

The total of 19 isolates was obtained from symptomatic greenhouse grown tomato leaves and fruits from major growing areas in Serbia. After DNA extraction (CTAB protocol), the gene-specific PCR primer pair cytb-BcF/cytb-BcR was used for amplification of *cyt b* fragment. PCR-RFLP assay with restriction enzyme *Fnu4HI* was used in order to reveal existence of mutation. Representative QoI-resistant (R) isolate and QoI-sensitive (S) isolate were chosen for *in vivo* assay. Tomato leaves were treated with recommended application rates of commercial formulations of pyraclostrobin (1 kg ha⁻¹) and trifloxystrobin (0.2 kg ha⁻¹) and after 3 hours inoculated with spore suspension (10⁶ conidia ml⁻¹). Nontreated leaves inoculated with spore suspension or sprayed with sterile water were used as positive and negative control, respectively. After incubation period (4 days, AT 23°C) presence of disease symptoms was assessed.

The primer pair cytb-BcF/cytb-BcR amplified ~560 bp PCR fragment in all tested isolates. RFLP analysis showed that 88 % of the isolates possess the *Fnu4HI* restriction site by producing two bands after digestion. This indicates presence of G-to-C point mutation resulting in the glycine-to-alanine substitution on position 143 which is responsible for QoI resistance. Leaves inoculated with R isolate manifested typical symptoms of gray mold in positive control and both fungicide treatments, while leaves inoculated with S isolate showed symptoms only in positive control.

These findings confirm the presence of *B. cinerea* QoI-resistant genotypes in Serbia and represent the starting point for further research in this field.

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SENSITIVITY OF NEOPESTALOTIOPSIS CLAVISPORA ISOLATES TO FLUAZINAM IN VITRO

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In the last ten years, the area under blueberry cultivation in Serbia has increased significantly and is now cultivated on over 2500 hectares. Blueberry production is threatened by a large number of phytopathogenic fungi and pseudofungi, of which species of the genus *Neopestalotiopsis*, especially *N. clavispora*, cause major damage. Symptoms initially appear in the form of reddish spots, which develop into larger necrotic lesions and sometimes even lead to plant failure. Management of the disease is mainly limited to the implementation of preventive measures, which include the selection of healthy and certified planting material, the maintenance of the optimal physiological state of the plant and the cutting of infected plant parts. The lack of registered fungicides to control the pathogen makes the use of chemical measures in blueberry production considerably more difficult.

N. clavispora was isolated in 2020 and 2021 from infected blueberry shoots collected from different locations in Serbia. Based on their morphological, pathogenic and molecular characteristics, all isolates were determined as *N. clavispora*. A total of 28 isolates were selected for sensitivity testing. Sensitivity test was performed under *in vitro* conditions using the mycelial growth assay method. A commercial formulation of fluazinam (Kardinal, 500 g L⁻¹ fluazinam, Galenika-Fitofarmacija a.d.) was used. The EC₅₀ values were calculated using the probit analysis method.

The results showed that the EC₅₀ values were ranged between 0.43 and 1.57 mg L⁻¹, with the average EC₅₀ value for all tested isolates of 0.34±0.91 mg L⁻¹.

Considering that there are no registered fungicides for the control of *N. clavispora* in Serbia, the results obtained in this study indicate the potential possibilities of using fluazinam to prevent this pathogen in blueberries.

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FUNGICIDE SENSITIVITY PROFILE OF MONILINIA FRUCTICOLA ISOLATES IN GREECE

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Brown rot is a devastating disease of stone fruits caused by several *Monilinia* species such as *M. fructicola*, *M. fructigena* and *M. laxa*. *M. fructicola*, although only recently introduced in Europe, has become the major agent of the disease. Brown rot control relies mainly on the use of fungicides belonging in different chemical classes. However, the development of resistance by pathogen populations may reduce the efficacy of these fungicides. The aim of this study was to determine the sensitivity levels of *M. fructicola* isolates originating from peach orchards in N. Greece to fungicides and elucidate the molecular mechanisms of resistance. For this purpose, 137 isolates were collected from infected fruit and their sensitivity to 4 different fungicides was determined. Fungicides tested were including: fenhexamid (HAs), cyprodinil (APs), tebuconazole (DMIs) and fludioxonil (PPs). After EC_{50} measurements the resistance factor values (RF) were calculated and each isolate was clustered based on the RF value, as sensitive, low, moderate and highly resistant. EC_{50} values ranged from 0.03 to 0.66 and 0.03 to 0.77 mg liter⁻¹ for cyprodinil and fludioxonil, respectively, while for fenhexamid and tebuconazole, the respective EC_{50} values ranged from 0.02 to 1.02 and 0.03 to 1.15 mg liter⁻¹. Noteworthy, high resistance frequencies were observed for cyprodinil (21.8 %), fenhexamid (29.9 %), fludioxonil (30.65 %) and tebuconazole (37.22 %), while multiple fungicide resistance was conspicuous among those isolates. The molecular mechanisms for the resistance, are under investigation, considering multiple target site mutations and overexpression of ABC/MFS transporters as a possible rationalization. This information provides an important advancement for risk assessment of chemical fungicide application to manage brown rot of stone fruit.

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EVALUATION OF THE FUNGICIDE SENSITIVITY OF ALTERNARIA SOLANI

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Early blight, caused by *Alternaria solani*, has the potential to reduce significantly yield in potato production globally. Quinone-oxidoreductase inhibitors (QoIs), succinate dehydrogenase inhibitors (SDHIs) and demethylation inhibitors (DMIs) represent three major fungicide groups that are used to control early blight. The sensitivity towards QoIs, SDHIs and DMIs of *Alternaria solani* isolates from different potato growing regions were monitored *in vitro*. Adaptation to QoIs and SDHIs have been detected in the last years and the underlying resistance mechanisms and their effects on efficacy reduction

have been elucidated. Sensitivity to DMIs was firstly described and show a rather stable situation so far. In some isolates low DMI adaptation were determined showing two new *CYP51* haplotypes with different target site mutations. To proof, if such mutations are responsible for this slightly reduced DMI sensitivity, transformation experiments were conducted. The results verified that the *CYP51* mutations are the responsible mechanisms. *In vivo* studies showed that DMIs are still effective on mutated isolates under field conditions and that the mutants were linked to significant fitness costs.

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DETERMINING FUNGICIDE RESISTANCE PROFILES OF BOTRYTIS CINEREA ISOLATES FROM GREENHOUSE-GROWN VEGETABLE CROPS IN CRETE ISLAND, GREECE

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Fungicide resistance is a threat of growing concern in the agricultural sector leading to huge financial damages worldwide. Protein structure alteration because of point mutations in gene targets of plant pathogens is the most common way of developing resistance. *Botrytis cinerea* is a particularly notorious plant pathogen regarding resistance development and indeed fungal populations simultaneously resistant to every botryticide available have often been observed in the fields. Since resistance can directly inhibit the control efficacy of grey mold disease, it is vital that quick and efficient resistance monitoring programs be developed and implemented in control strategies. For this purpose, we conducted a monitoring study of *B. cinerea* sensitivity to fungicides using isolates obtained from tomato and eggplant greenhouses in the region of Crete. In total, 63 isolates were collected and were subsequently tested in substrates amended with discriminatory doses of boscalid, fluopyram, pyraclostrobin, cyprodinil, fenhexamid, fludioxonil and tolnaftate as an indicator for the presence of Multi-Drug Resistance (MDR). Interestingly, 50% of the isolates collected from tomato were resistant to fluopyram and cyprodinil while no MDR was detected. Concerning aubergine, 66 % of the isolates were resistant to pyraclostrobin and 2 isolates were characterized as resistant to fludioxonil, which may imply the presence of MDR. For the detection of the resistance conferring mutations, the *sdhB* gene was sequenced, while a Tetra- primer ARMS PCR was used for the detection of the mutations in *pos5* gene. Results showed that 68 % of the AP-resistant isolates possessed the mutation L412F in *pos5*. Such results indicate the presence of the fungicide resistant populations of *B. cinerea* in Cretan greenhouses and quick detection tools like Tetra primer ARMS PCR could aid in understanding the genetic background of the resistant isolates.

This project is co-funded by the Greece and the European Union. In the frame of the Rural Development Programme 2020-2014, Sub-Measure 16.2 - 16.1 - Establishment and operation of Operational Groups (O.G) of the European Innovation Partnership (EIP) for agricultural productivity and sustainability. Title of the project: Evidence based management of pesticide resistance in vegetable crops. (Project code: M16ΣYN00094-2).

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A SURVEY OF VIRUSES AND VIROIDS OF CANNABIS SATIVA IN SOUTH AFRICA

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Cannabis sativa is currently gaining attention in South Africa and worldwide as a commercial crop for industrial, medicinal and recreational use. In 2019, The South African government released the National Cannabis Master Plan for South Africa that presented an integrated government approach towards unlocking the economic potential of cannabis in the country. However, pest and pathogens, including viruses, negatively affect the cannabinoid content and biomass of medicinal cannabis and industrial hemp, respectively.

The aim of this study was therefore to conduct a survey of viruses of *Cannabis sativa* in South Africa to establish viruses infecting the crop toward management of viral diseases.

Leaf samples were collected from indoor and field grown plants in KwaZulu Natal, Limpopo Gauteng and Western Cape provinces. A metagenomics approach via RNASeq of total RNA extracted from the leaf tissue and *de novo* assembly and analysis of the resultant contigs, was used to detect viruses. Reverse transcriptase PCR was employed to validate the presence of viruses detected via RNASeq.

The results indicated the presence of hop latent viroid, alfalfa mosaic virus, cannabis cryptic virus, grape line pattern virus and cannabis sativa tobamovirus. This is the first report of all these viruses in *Cannabis sativa* in South Africa.

This study will inform virus diagnostics in the growing cannabis industry and enable appropriate virus management measures towards sustainable production of cannabis and hemp in South Africa.

In Maryland the virus was identified in single infections in plants exhibiting yellow lines and oak-leaf patterns and was thus tentatively named blackberry line pattern virus (BLPV). BLPV has a bipartite genome with a negative sense RNA1 and ambisense RNA2. RNA1 encodes the RNA-dependent RNA polymerase (RdRp). RNA2 encodes the movement protein and the nucleocapsid protein. Fifteen genomic sequences were obtained from Arkansas, Maryland, Mississippi, North Carolina, Oregon, and South Carolina. The RdRp of BLPV is closely related to the ortholog from Citrus concave gum-associated virus, Citrus leaf flecking-associated virus, Citrus virus A; and Watermelon crinkle leaf-associated virus 1. All the abovementioned viruses belong to the newly formed genus *Coguvirus* in the family *Phenuiviridae* suggesting that BLPV is also a member of the genus. All BLPV RdRps are very similar showing between 97.8 and 100 % aa identity. BLPV is graft transmissible and grafted plants tested positive for BLPV after natural dormancy, however symptom expression was transient and not evident in all positive plants in greenhouse setting. Limited scope symptomatic plants were screened in a farm setting in Maryland and PCR test results 100 % correlated with line pattern symptoms. Further evaluations are warranted to fully understand the biological significance of BLPV in blackberry.

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DISTRIBUTION AND BIOLOGICAL CONTROL OF THE BLUE GUM CHALCID, LEPTOCYBE INVASA (HYMENOPTERA: EULOPHIDAE) IN URUGUAY

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Eucalypt plantations in Uruguay exceed 80 % of the commercial forested area. *Leptocybe invasa* (Hymenoptera: Eulophidae), native to Australia, is one of the main *Eucalyptus* insect pests that are causing problems. In Uruguay was first detected in 2011. It can induce the formation of galls on the midrib and /or petiole of leaves, young stems, or branches of *Eucalyptus* spp. In addition to apical dominance loss, malformations, super-sprouting, or the death of very susceptible genotypes. Biological control (CB) and plant resistance are considered the best methodologies for their management. In field trips between 2019 to 2021, we found three wasp species associated with *L. invasa* galls. The objective of this work was to establish the *L. invasa* distribution and evaluate the use of biological control in Uruguay. Since 2013, has been carried out in Uruguay samplings to collect *L. invasa* galls. These were georeferenced, processed in the laboratory, and placed in Ziploc bags in a climatic chamber, with a 12:12 photoperiod at 24 °C. A daily review of them was carried out for the collection, identification, and sexing of the emerged wasps. In addition to the record *L. invasa* adults, the parasitoid species *Quadrastichus mendeli* and *Selitrichodes neseri* (Hymenoptera: Eulophidae) were identified. *Leptocybe invasa* was observed in almost all the localities sampled except in the central southeast of the country. *Selitrichodes neseri* were found in the samples from

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A BROAD OCCURRENCE OF A NEW BLACKBERRY VIRUS IN THE U.S.

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A previously uncharacterized virus from blackberries was identified independently by HTS in multiple parts of the U.S.

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Rivera, Cerro Largo, and Tacuarembó. *Quadrastichus mendeli* was also recorded in those departments as well as in Salto and Paysandú. An attempt was made to breed both parasitoids, obtaining offspring. Taking into account that the frequency of registration of *S. nesei* and *Q. mendeli* has increased and that breeding of both parasitoids was possible, it is proposed that the use of biological control for the management of *L. invasa* is feasible in Uruguay.

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PHYLOGENETIC RELATIONSHIPS OF AUCHENORRHYNCHA SPECIES EXISTING ON THE FOLIAGE OF VINES

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Typhlocybae is an abundant and widespread subfamily of Auchenorrhyncha, which contains phytophagous species that feed with the mesophyll of the leaves and can cause stippling on a wide variety of plants, including crops of economic importance. The aim of this research was to study the phylogenetic relationships of Typhlocybae species existing in the foliage of the vines, using the sequencing analysis of COI mitochondrial DNA (mtDNA) gene segment.

Samples for identification were collected fortnightly from the vineyard of the campus of the Agricultural University of Athens, using yellow sticky traps. The samples were stored under freezing conditions. Total DNA extraction was performed using DNeasy Blood & Tissue Kit (Qiagen), and Polymerase Chain Reaction (PCR) using BARCODE primers for the COI mt DNA gene segment. PCR products were purified using the Nucleospin Extract II kit (Macherey-Nagel). Individual sequences were determined via automated sequencing of the above mtDNA gene segment provided by CEMIA S.A. Company (Larissa, Greece). For data processing, the packages Finch TV 1.4.0, BioEdit 7.0.9.0, CLUSTALW2, DnaSP5.10, MEGA 6.0.0, and GenAI EX 6.5 were used.

The results show that the most abundant species collected above grapevines was *Jacobiasca lybica* and genetic variability was detected, while an introduced species, *Anzygina honilola*, was also collected above leaves. Further details with detailed discussion are given in the presentation.

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EXPERIMENTAL DEVELOPMENT AND IMPLEMENTATION OF A MANAGEMENT PROGRAM AGAINST LOCUSTS (ORTHOPTERA: ACRIDIDAE) IN FTHIOTIS AND AGHIOS EFSTRATIOS AREAS IN GREECE

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Locusts (Orthoptera: Acrididae) are potential threat to various crops, since excessive increase of their population can lead to outbreaks causing great crop losses, decreasing farmers' income, damaging native vegetation and reducing biodiversity. Locusts' outbreaks occur in the regions of Kompotades (Lamia) and Agios Efstratios island (North Aegean), since 2022 and 2016, respectively.

The objective of this case study was the management of locusts' population designing an action plan that would also restore the biodiversity of the areas. The hotspots of the locusts' population were detected by scouting teams and surveillance UAV and chemical and non-chemical insecticides were applied in the areas with ULV sprayers. For the assessment of the efficacy of the control programs, the population variation of the locusts and the Simpson biodiversity index of the areas were calculated through samplings.

The results showed that there is more than 80 % decrease in the population of the locusts in the areas even after the first insecticide application. As long as the biodiversity is concerned, the Simpson biodiversity index was extremely high (>0.75) in both areas and the detected species of flora and fauna were also increased. Further details about the results and extensive discussion are given in the presentation.

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ENHANCING THE DEVELOPMENT AND FECUNDITY OF ORIUS STRIGICOLLIS BY USING TWO PROBIOTICS: PANTOEBA SP. AND STAPHYLOCOCCUS SP.

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Orius strigicollis (Poppius) has been commonly released to control minute pests due to its remarkable foraging and predation ability. Improving mass rearing by manipulating the gut microbial composition through providing selected probiotics is a potential approach for enhancing the efficiency of *O. strigicollis* in biological control applications. Here, we used culture-based approaches to isolate the core microbiota characterized through 16S rRNA gene amplicon sequencing. The effect of supplementation with two of these isolates, *Pantoea* sp. and *Staphylococcus* sp., on life history traits and expression levels of *Vg* and AMPs genes were evaluated. Our study shows that supplementing *Staphylococcus* sp. enhances the reproduction of *O. strigicollis* by upregulating the expression level of the *Vg* gene. Additionally, the supplementation with *Pantoea* sp. significantly increased the nymphal survival rates of *O. strigicollis* and boosted the expression level of AMPs gene compared to the control.

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This indicates that supplementation with *Pantoea* sp. has a systemic immune effect on *O. strigicollis*, potentially regulating the microbiota and preparing the immune system against future pathogenic infections. We propose that *Pantoea* and *Staphylococcus* species play a functional role in the successful reproduction and survival of *O. strigicollis* and could potentially serve as a probiotic supplement in rearing systems. This study presents a method for selecting culturable probiotics for *O. strigicollis* to enhance specific life history traits. These findings may inform future efforts to improve other predator rearing through microbiota manipulations.

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POPULATION SIMULATION BASED ON LIFE TABLE LENDS A COMPREHENSIVE PERSPECTIVE TO THE SUSTAINABLE INHIBITION OF FALL ARMYWORM THROUGH NUCLEOPOLYHEDROVIRUS INFECTION

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Spodoptera frugiperda (J.E. Smith) (Lepidopteran: Noctuidae), fall armyworm (FAW), is an edacious pest that can damage numerous host plants with worldwide distribution. With the promotion of sustainable agriculture, the non-pesticide control agent and eco-friendly management strategy are quite necessary to be developed. The biological activity of *S. frugiperda* multiple nucleopolyhedroviruses (SfMNPV) to FAW larvae was determined with a food-contamination bioassay. The result showed absolute pathogenicity to FAW larvae, especially for neonates. The median lethal concentration of SfMNPV to 3rd instars larvae was $10^6 \times 1.46$ OBs/ml for 14 days post-inoculation at 25°C. Besides, we analyzed the effects of SfMNPV at the concentration of 10^6 OBs/ml on the FAW immatures as well as adults based on the age-stage two-sex life table theory. The inhibition of population growth of *S. frugiperda* is caused by SfMNPV due to the lethal mortality in larval, pupal, and adult stages, in addition to the retarded oviposition period and reduced reproduction rate of FAW as well. The intrinsic rate of increase and net reproductive rate in the cohort treated with SfMNPV were lower, i.e., $0.1713 \pm 0.0131 \pm \text{day}^{-1}$ and 18.3 ± 64.6 offspring, than those fed on non-NPV treated maize leaves, $0.1937 \pm 0.0137 \pm \text{day}^{-1}$ and 364.5 ± 64.6 offspring, respectively. Compared to the cohort without NPV treatment the cohort without NPV treatment, the population projection showed that SfMNPV could effectively decrease the population size of *S. frugiperda* into ca. 8.7 % of the non-NPV cohort in 75 days. This study demonstrated an integrated effect of SfMNPV on the demographic characteristics of *S. frugiperda* and the possibility of offspring infected with NPV by vertical transmission. The SfMNPV's pathogenicity was comprehensively evaluated from the perspective of demography, and this information may encourage decision-making on pest management.

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UTILIZING TAGETES MINUTA FOR EFFECTIVE MANAGEMENT OF GRAPEVINE FANLEAF VIRUS (GFLV)

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Grapevine fanleaf virus (GFLV) poses a significant threat to grape cultivation, being transmitted through nematodes of the genus *Xiphinema*. In South Tyrol, Italy, GFLV stands out as a formidable adversary, inflicting substantial economic losses by diminishing yield, compromising fruit quality, and shortening the lifespan of grapevines. This study aimed to mitigate the *Xiphinema* sp. population in the soil, thereby reducing GFLV occurrence in vineyards, employing various soil-improving techniques.

The experiment assessed the effectiveness of sowing *Tagetes minuta* in conjunction with herbicide treatments, one- or two-year fallows, and intensive soil tillage in a commercially managed vineyard featuring the Gewürztraminer cultivar. To gauge treatment efficacy, GFLV-infested plants were identified through visual inspection and enzyme-linked immunosorbent assay (ELISA) tests for GFLV and Arabis Mosaic Virus (ArMV). Additionally, soil samples were scrutinized for the presence of *Xiphinema* sp. nematodes, identified to the genus level based on morphological characteristics.

After four years, the plots treated with *T. minuta* exhibited nearly no GFLV infestation, in stark contrast to the control plot where approximately 30 % of the plants were affected. The conclusive evaluation is scheduled for 2026, promising valuable insights into the long-term effectiveness of the employed strategies.

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"BASIC SUBSTANCES" AS USEFULNESS SUBSTANCES TO LIMIT MAJOR PATHOGENS OF POTATOES IN ORGANIC FARMING

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Basic substances (BS) can represent an opportunity to mitigate the problems associated with chemical method of plant protection (CMPP), they are defined as compounds that are not used as plant protection products (PPPs), but may be useful. They are not placed on the market as PPPs and are not registered. Potential BS could be used in crop protection due to their efficacy in controlling plant pathogens and pests, they have a wide range of applications. BS can act as direct control products for diseases and pests by exerting a fungicidal, bactericidal, and insecticidal activity, or can be employed in indirect control strategies, such as in triggering plant immune response (e.g., elicitors), or as attractants or repellents. European pesticide database (<https://ec.europa.eu/food/plant/pesticides/eu-pesticides-database/start/screen/active-substances>) lists 24 approved BS with usefulness reports.

The combination of basic substances with different PPPs or plant extracts can be an effective alternative to CMPP or used in OF. However, they are not used very often in practice, since their field efficacy depends on various factors, including the

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crop, the pathogen or pest, the origin of the basic substance and the timing and method of application. The composition of the basic substance is highly relevant.

Aims of the study – assessment of the efficacy of different strategies based on BS used to limit of symptoms of potato plant diseases and keep them at a level below economic harmfulness

The experiments were conducted out in the laboratory, the greenhouses and field trials, *in vitro* and *in vivo*. Varied concentration of BS, testing unique strategies involving copper and other BS used alone or combined were made. Potato pathogens were assessed- *Phytophthora infestans*, *Alternaria alternata*, *A.solani*. Different cultivars were used, foliar applications were made from 4-6 times, every 7-10 days. Tests were made in two regions of Poland.

Yield obtained in 2022 from one region of Poland.

Strategies	Mean yield [kg/ha]			
	c. Lilly	c. Vineta	c. Tajfun	Kg/ha
Cu	18333,33	21463,33	16290,00	18 695,56 b
Cu/ sunflower oil	22600,00	23023,33	18636,67	21 420,00 a
Cu/extract onion	18426,67	24366,67	17830,00	20 207,78 ab
Extract onion 25%	19866,67	24113,33	15886,67	19 955,56 ab
Sunflower oil	18793,33	23863,33	18906,67	20 521,11 ab
Cu/ chitozan	22093,33	22500,00	16980,00	20 524,44 ab
Chitozan 1%	22363,33	25456,67	17860,00	21 893,33 a
Untreated	14336,67	16540,00	13350,00	14 742,22 c
Mean	19601,67b	22665,83a	16967,50c	

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SIMULATING CONTROL THRESHOLDS FOR WHITEFLIES ON TOMATOES TO REDUCE THE OCCURRENCE OF TOMATO LEAF CURL THAILAND VIRUS IN TAIWAN

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The ability of monitoring pests is the key factor of successful Integrated Pest Management (IPM) program. Yellow sticky papers are frequently used for monitoring the presence of adult whiteflies and served as an early warning indicator. However, counting the adults of whiteflies on yellow sticky papers is a daunting task for most farmers. To assist farmers in monitor whitefly population density, we introduced the method of turning leaves over instead of using yellow sticky papers to scout adult whiteflies. The number of adult whiteflies was also be analyzed as a control threshold. Consequently, a linear model was developed to estimate the number of whitefly nymphs by incorporating the parameters of non-viruliferous and viruliferous whiteflies in the intrinsic rate of increase, as well as age-specific survival rate. Additionally, the rates of control for insecticides and non-chemical methods were taken into consideration. The results indicated that if without implementing control measures for four consecutive weeks, with an average of two or five adult whiteflies per compound leaf, the number of nymphs per leaflet was 120 and 192 nymphs for non-viruliferous whiteflies. There were more than 700 nymphs per leaflet for viruliferous whiteflies. In contrast, the estimated number of whiteflies is 0.7 and 1 nymphs/ leaflet for non-viruliferous whiteflies, and less than 12 nymphs/leaflet for viruliferous whiteflies, when insecticides were used once a week. Based on the estimation model, it is recommended to set the control threshold at 2 adults per compound leaf or fewer from planting until the beginning of harvest in order to reduce the spread of whitefly-transmitted Tomato Yellow Leaf Curl Thailand Virus (TYLCTHV). According to the control threshold for implementing whitefly IPM, it has the potential to decrease the disease incidences of TYLCTHV by approximately %6-5.

Keywords: whitefly, control threshold, simulation, tomato, Integrated Pest Management, Tomato Yellow Leaf Curl Thailand Virus

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SELECTION TECHNIQUES OF AGROCHEMICAL MATERIALS BY MULTI ARRAY LEAF DIPPING BIOASSAY METHOD FOR THE PRECISION MANAGEMENT OF TWO-SPOTTED SPIDER MITE

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It is necessary to develop an appropriate selection technology for the precise management against two-spotted spider mite (TSSM) in glass houses due to the rapid development of resistance characters against various chemicals. This study focuses on the development of a multi array leaf dipping bioassay method (MLDB) and selection technology from synthetic pesticides and organic agricultural materials. In the suitability evaluation test of MLDB, residual synthetic insecticides of leaf disc were not affected on the mortality of TSSM on adjacent leaf discs, and the preparation time could be reduced about 21.7 % compared to the conventional bioassay methods. As a result of mortality observation at 24h intervals for 4 days from 26 synthetic insecticides and 38 organic agricultural materials, 9 tested materials (P_Test_15, P_Test_19, P_Test_25, O_Test_01, O_Test_06, O_Test_13, O_Test_16, O_Test_23 and O_Test_32) revealed the relatively high efficacy on averages. The low exposure frequency and newly registered synthetic insecticide revealed relatively high acaricidal efficacy. Some organic agricultural materials might be considered as control agent well as prevention. The weighted arithmetic mean mortality rate that can reflect the efficacy expression rate for each agricultural materials was employed, and five levels of efficacy rank index were applied to select appropriate agricultural materials for each field strain. Among of the total 64 candidate materials, 5 to 14 of the test materials revealed a mortality of 60 % or more, and those might be preferentially utilized for the control of TSSM strains in respectively. In summary, the efficacy evaluation by MLDB and efficacy rank index by the weighed arithmetic mean mortality would contribute to the appropriate agricultural materials selection for the precise control of TSSM in glass houses.

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BIOLOGICAL CONTROL AND CROSS INFECTIONS OF THE NEOFUSICOCCUM SPP. CAUSING MANGO POSTHARVEST ROTS IN SPAIN

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Mango and avocado are the main subtropical crops in southern Spain. These fruits are grown and processed in the same geographical area and are transported to the rest of Europe. However, postharvest diseases during transportation and storage could damage the export market. These two

fruits share, on many occasions, farms, processing and even transport, and could act as cross inoculation source. In this work, symptoms of rot in mango fruits were detected, and analyzed in search of the causal agent. Concurrently, asymptomatic avocado fruits were also analyzed. The main fungal genera found, both in mango and avocado, were *Alternaria* sp. and *Neofusicoccum* sp. Of these two genera, only *Neofusicoccum* sp. was able to reproduce the symptoms of rot in mango like previously detected. Likewise, *Neofusicoccum* sp. isolates, but not *Alternaria* sp., could produce rot symptoms in avocado inoculated fruits.

Two microbial biological control agents were tested to study the control of this disease through sustainable strategies. These candidates to biological control were *Pseudomonas chlororaphis* PCL1606. and *Bacillus velezensis* UMAF6639, both antagonists showed antifungal characteristics. The applications of both microorganisms on the fruit showed significant levels of protection during preventive applications, although only UMAF6639 showed greater persistence in the fruit during the preventive applications in the field.

According to our results, it is possible to establish that pathogenic *Neofusicoccum parvum* and *N. mediterraneum* are the main causes of mango postharvest rots in southern Spain. These fungi could also be potential pathogens to avocado fruits, suggesting a risk of cross-infections among crops. Biological strategies to control postharvest rots have shown better efficacy of preventive application strategies. Following the same strategy in open field experiments.

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INSECTICIDAL EFFICACY ASSESSMENT AND MODELING OF THERMAL TREATMENT PENETRATION WITHIN COMMODITY MASS OF CURRANTS

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Currants are among the primary dried fruits cultivated in Mediterranean regions due to their exceptional nutritional value. However, there is a gap regarding data concerning their storage and disinfection processes. The utilization of extreme temperatures offers a promising alternative to chemical methods for effectively disinfecting various stored commodities, including dried fruits and grains. Nevertheless, the lack of data concerning the influence of commodity presence on Thermal Penetration (TP) and the efficacy of such methods creates the potential for misconceptions to arise. This study aims to assess the efficacy of both cold and heat treatments under real-world conditions, taking into account the crucial factor of product presence. An initial thermal

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treatment (TT) model is developed using TP algorithms based on heat transfer equations with the Finite Difference Method and Fourier's law for heat conduction. Predicated on storage facility settings, a simulation model was tested, demonstrating the feasibility of designing an algorithm that is based on the different time-mortality combinations for the different application scenarios and TP. In addition, temperature/humidity sensors were utilized to record temperature values in different settings. Our findings indicate a significant impact of product presence on the effectiveness of the TT method on major pests of currants. In conclusion, the results of our study highlight cold and heat treatment as a viable, more sustainable disinfection strategy for enduring products.

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ASSESSMENT OF THE INSECTICIDAL IMPACT OF PHOSPHINE ON EGGS OF MAJOR INSECT PESTS OF STORED PRODUCTS IN LABORATORY EXPERIMENTS

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Phosphine has been long regarded as the predominant fumigant insecticide in stored product facilities. Despite global reports of phosphine resistance, limited research exists on its impact across different developmental life stages of insects. Our study aimed to assess the effectiveness of phosphine on eggs from different strains of major stored product insects. We investigated egg hatching rates following exposure to phosphine concentrations ranging from 50 to 1000 ppm and exposure durations ranging from 2.5 to 7 days. Our findings uncovered differing egg susceptibility based on species, age, and exposure. Eggs from susceptible strains did not hatch when exposed at 50 ppm, indicating high phosphine efficacy. Additionally, exposure to 200 ppm for more than 5 days resulted in complete mortality in all cases. Contrasting susceptibility between 1-day-old and 2-day-old eggs was evident, with the latter consistently displaying elevated mortality probabilities. In addition, our study underscores the intricate interaction between phosphine concentration and exposure duration in achieving optimal control. Our results significantly enhance comprehension of insect resistance

dynamics and provide valuable insights for refining fumigation practices, enabling more targeted and efficient control of stored product pests.

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EXPLORING THE EFFECT OF ELECTROLYZED WATER AGAINST BROWN ROT DISEASE (MONILINIA SPP.) ON PEACH FRUITS

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Brown rot caused by several species of *Monilinia* genus, stands as one of the most significant diseases affecting stone fruit globally. However, over the past decade, *M. fructicola* has emerged with high prevalence in numerous countries, including Greece. Consequently, the losses incurred due to *Monilinia* spp. remain a primary concern for growers, industry stakeholders, and agricultural researchers given their detrimental impact on both pre- and postharvest phases of peach production. The aim of this study was to evaluate the effectiveness of postharvest application with electrolyzed water (EW) to reduce the losses occurred by *M. fructicola* in peaches. In detail, we evaluated the effect of several types of EW (pH 7.6, pH 8.6, and pH 9.6) and immersing duration, on the development of *M. fructicola* after artificial inoculation. Commercial harvested peaches (cv. Andross) were surface sterilized (2 min, 3 % NaClO), and artificially inoculated with *M. fructicola* conidial suspension (4×10^5 conidia ml⁻¹). Artificially inoculated fruits were immersed in EW solutions for 10 min in different hours post the inoculation (12, 8 and 24 hpi). Subsequently, the fruits were transferred to plastic container (to ensure high humidity levels), and incubated for seven (7) days at room temperature. Results revealed that applications with EW 8 hpi, significantly reduced the disease severity, while no significant differences were observed among the tested pH levels. Similar results were obtained 12 and 24 hpi, however the fruits treated with EW (pH 7.6) presented higher disease severity compared to the control, while application with EW with pH 8.6 and 9.6 considerably reduced the disease development.

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SYNERGISTIC IMPACTS OF PROPARGITE EXPOSURE AND DEFORMED WING VIRUS INFECTION ON THE HEALTH OF WESTERN HONEY BEES

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Every year, unusually high rates of bee colony loss are frequently reported around the world, with possible causes including loss of natural habitats, long-term exposure to agrochemical mixtures, parasite and pathogen infections, and other joint factors. In early 2023, numerous bee die-off events

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occurred in southern Taiwan. Analysis of samples collected from different areas revealed that dead bees contained high amounts of bee viruses, and acaricide residues were also detected in the samples. We speculate that the mass mortality may have been caused by the synergistic effects of pathogens and pesticides.

Our experiments showed that infections of Varroa mite-borne Deformed Wing Viruses (DWV) alone had a limited impact on bees. However, simultaneous treatment with propargite of different concentrations yielded increased impacts on mortality. Treatment with DWV alone significantly increased the expression of immune response genes, but treatment with propargite alone did not significantly increase the expression of detoxification genes. However, simultaneous treatment using both DWV and propargite significantly increased the expressions of both immune response and detoxification genes, indicating that resistance genes in honey bees were significantly increased following simultaneous treatment. Supplementation of adenosine in sugar water given to bees significantly reduced bee mortality caused by DWV and propargite, a finding which may be consistent with the idea that large amounts of energy are required to combat environmental adversities. Adenosine supplementation may trigger energy transformation in bees and improve their resistance to environmental adversities.

This study reveals a link between viral infections and bee sensitivity to acaricides, which may be caused by impaired physiological or immune mechanisms. Additionally, energy supplementation may help to increase bee resistance to environmental adversities.

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DETERMINING THE POLLINATION ECOLOGY OF MESPILUS CANESCENS (ROSACEAE)

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Pollinators play a vital role in the upkeep of ecosystem services and biodiversity across the world with evidence suggesting that 80 % of flowering plants (including economically important crops) depend on biotic pollination yet, pollinators across all taxa are on a decline due to anthropological and natural phenomena. A unique and critically endangered tree, Stern's Medlar, provides pollinators with much-needed floral resources in May after overwintering, allowing insect populations to easily increase. To understand and conserve Stern's Medlar's ecological importance, it is vital to establish a plant-pollinator interaction network that provides comprehensive information on underlying causes for coevolution, biodiversity indexes, and ecosystem stability for the current and future society to build upon and examine. Aim 1: Establish a plant-pollinator network Stern's Medlar and Aim 2: Determine whether biotic insect pollination is vital to Stern's Medlar's pollination.

For Aim 1 we will study Stern's Medlar situated at the University of Arkansas's (UARK) Rosen Center Horticulture Garden. The tree will be segmented into 4 sections based on the cardinal points. Each segment will receive 10-minute observation periods and will be followed by 10-minute collection periods.

The containers will be placed in a -80°C freezer to ethically euthanize and preserve the insects for later identification. For Aim 3 2 random inflorescence clusters of at least four flowers will be divided into three: 1) the Control Group, 2) The Hand-Pollinated Group, 3) and The Open Group. Each of the 36 inflorescence group branches will be tagged with color-coded tape to revisit post-bloom season. Once Stern's Medlar produces fruit the number of fruits, the fruit's total weight, and after dissection, the seed's total weight will be taken. The results and conclusion will be discussed later. Data acquisition and analysis are still underway.

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DEVELOPMENT OF AN OPTIMAL SEED MIXTURES FOR PERMANENT GROUND COVERS IN VINEYARDS

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The installation of a permanent ground cover in vineyards offers numerous agronomic (soil protection, soil fertility, weed control) and environmental (reduced leaching of nutrients and plant protection products, reduced use of herbicides, promotion of the biodiversity) benefits. Nevertheless, such ground covers are not always free of drawbacks, such as competition for water and nitrogen, increased risks of frost, management. These drawbacks are particularly important with the main seed mixture presently used, composed mainly forage variety of grass species, quite cheap but selected to grow vigorously.

For the growers, the challenge is to manage the green ground covers in such a way as to preserve their advantages while limiting these drawbacks. Among the tools available to the winegrower is the sowing of a seed mixture composed of selected species, potentially less competitive against the crop.

Since 2018, different seed mixtures have been tested in comparative trials as well as on networks of *on farm* plots. On those parcels several aspects have been monitored, such as dynamic of vegetation, biodiversity, biomass production and reduction of the weed pressure. On the crop, plant water statut, yields, as well as quality of the moss and the vine, have been studied.

Today, four different seed mixtures, adapted to diverse agronomic and pedo-climatic conditions, have been developed with plant species such as *Bromus tectorum*, *Lotus corniculatus*, *Medicago lupulina*, *Poa compressa*, *Prunella vulgaris*, *Sanguisorba minor* and *Thymus pulegioides*. Those mixtures offer less biomass production, allowing a reduction of the management intensity (number of cuttings reduced by 3/1) and better biodiversity. Competition for water and nitrogen are still present, probably due to development of underground structures and/or higher rhizodeposition.

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THE WILD EMMER WHEAT GRAIN PROTEIN CONTENT 5B QTL INTROGRESSED INTO BREAD WHEAT IS ASSOCIATED WITH TOLERANCE TO NITROGEN DEFICIENCY

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Plant diseases pose significant threats to global food security. Stripe rust is one of the most wheat-devastating diseases caused by the fungus *Puccinia striiformis* f. sp. *tritici* (*Pst*). Developing disease-resistant crops offers a cost-effective solution for sustainable wheat production. *Yr15* is a broad-spectrum resistance gene derived from wild emmer wheat (*Triticum dicoccoides*) gene that confers resistance against >2000 *Pst* isolates from 47 countries around the globe. *Yr15* encodes a tandem kinase-pseudokinase protein architecture designated wheat tandem kinase 1 (WTK1), which is a new player in plant immunity. WTK1 induces form of immune cell death, called the hypersensitive response (HR), that occurs after fungal haustoria development. To investigate the transcriptional changes activated by WTK1 in response to infection with *Pst*, we conducted transcriptional time-course analyses of a susceptible durum wheat genotype, Kronos, compared with its resistant near-isogenic transgenic line, Kronos+WTK1, after inoculation with *Pst*. To simultaneously identify differentially expressed genes between genotypes over time, we used the R package Moanin. By spline clustering, we identified 20 differentially expressed gene clusters that exhibit various regulation patterns. Thirteen clusters were upregulated in the resistant line and were enriched in genes involved in pathogen recognition, signal transduction, phytohormone production, and HR cell death responses. Pairwise comparisons and weighted gene co-expression network analyses pinpointed 11 modules specific to time points post-infection. We identified a genotype-specific module containing upregulated genes in the resistant Kronos+WTK1 genotype. This module displayed an enrichment of genes involved in basal defense pathways, such as phytoalexin production, callose deposition, and antifungal activity, as well as ROS and HR-cell death immune responses. Thus, the comprehensive transcriptome analysis of wheat NILs revealed intricate genetic networks triggered by WTK1 in response to *Pst* infection. These interactions result in robust defense signaling pathways, ultimately leading to disease resistance.

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DECIPHERING THE MECHANISMS OF DROUGHT STRESS ALLEVIATION BY PSEUDOMONAS N122

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In recent years, drought has emerged as a significant challenge for global food crops, prompting the exploration of innovative solutions. One promising avenue is harnessing the ability of plant-associated microorganisms to enhance drought tolerance, potentially boosting crop yields. In this study, we evaluated 146 bacterial isolates from four main phyla—Proteobacteria (110), Bacteroidota (18), Actinobacteria (11), and Firmicutes (5)—isolated from Terschelling Island (The Netherlands) for their potential to alleviate drought stress in *Arabidopsis thaliana*. Among these isolates, *Pseudomonas* sp. N122 showed notable drought-stress alleviation *in planta* via volatile organic compounds (VOCs) production. Microscopic analysis revealed that these VOCs promoted root hair formation, facilitating the plant's adaptation to drought conditions. To gain deeper insights into the underlying mechanisms, we conducted gas chromatography mass spectrometry (GC-MS) experiments to identify potential compounds involved in this phenotype. Additionally, we are in the process of establishing a comprehensive *Pseudomonas* sp. N122 random mutant library, aimed at elucidating the pivotal genes responsible for mediating drought stress alleviation and root hair formation in planta. Through these combined efforts, we aim to contribute to a better understanding of the intricate interactions between microbes and plants under drought conditions, with potential implications for sustainable agriculture practices.

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RELATIONSHIP OF CLIMATE WITH THE GUATEMALAN MOTH *TECIA SOLANIVORA* (POVOLNÝ, 1973) (LEPIDOPTERA: GELECHIIDAE) IN A POTATO (*SOLANUM TUBEROSUM* L.) CROP

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Climate variability and climate change scenarios influence the behavior of species and natural resources. The potato crop (*Solanum tuberosum* L., Solanaceae) produces a food of high nutritional value that is a staple in the family food basket. In Colombia, the production system is affected by pests, such as the Guatemalan potato moth caused by *Tecia solanivora* (Povolný), with few field studies describing its relationship with climate. The objective of the study was to

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evaluate the effect of climatic elements on the populations of male adults of *T. solanivora* in three potato crop cycles in a locality in Colombia. The experiment was established in the municipality of Mosquera, Cundinamarca, Colombia and baited traps with sex pheromone were used to capture Guatemalan potato moth adults in the three cycles of potato crop var Diacol capiro with simultaneous records of: number of adults, precipitation (mm), relative humidity (%), minimum, average and maximum temperatures (°C), wind speed (km/h), sunshine(h/day), agroclimatic indices of degree days (°Cd), reference evapotranspiration (mm/day) and water index (WI). Pearson and Dickey-Fuller tests, time series and cross-correlation were performed. The results indicate the fluctuation of *T. solanivora* adults with positive correlation with maximum temperature, degree days and sunshine, and negative correlation with precipitation ($p < 0.05$). Maximum temperature ($20 - 23\text{ }^{\circ}\text{C} \pm 0.7\text{ }^{\circ}\text{C}$) presented the highest significance ($p < 0.05$) of all cross-correlations with *Tecia solanivora*, with direct relationship by exceeding the autocorrelation coefficients, which increased the population and movement of *T. solanivora* adults. The autocorrelation with precipitation ($62 - 128\text{ mm} \pm 22\text{ mm}$) showed population decrease ($p < 0.05$). It is concluded that climatic variables were critical limiting factors for the emergence and survival of *T. solanivora*.

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BACTERIA-MEDIATED SALT STRESS ALLEVIATION IN RICE

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Rice (*Oryza sativa* L.) is one of the most important staple foods in the world and a major source of income for millions of people. However, rice cultivation is heavily affected by climate change. Drought and seawater intrusion has led to salinization of rice paddies resulting in significant yield losses across Europe and Asia. Root-associated microbes have been reported to enhance protection against abiotic stresses such as salt stress. We established a large collection of root-associated bacteria from a hypersaline rice paddy located in the Guadalquivir area near Isla Mayor, Spain. We developed and optimized a high-throughput screening for bacteria-mediated salt stress alleviation in rice. Using this high throughput screening we aim to identify novel bacteria which can alleviate salt stress in rice seedlings and increase germination and growth. Preliminary results demonstrate the efficacy of several actinomycete isolates in alleviating salt stress in rice seedlings *in vivo*. This research aims to

elucidate the underlying mechanisms involved in salt stress alleviation by bacteria. Understanding these mechanisms will provide valuable insights into the interactions between root-associated bacteria and rice plants under saline conditions, offering new paths for rice cultivation in salt-affected regions.

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BIOACTIVE COMPOUNDS FROM ALFALFA: EVALUATION OF THEIR POTENTIAL USE AS BIOPESTICIDES AGAINST TOMATO PATHOGENS

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In the frame of AGRITECH project, financed by NextGeneration EU, a task was dedicated to the development of bioactive compounds from plant biomasses as potential new eco-friendly biopesticides. In this work we report the preliminary results on the evaluation of three extracts from *Medicago sativa* towards pathogens infecting *Solanum lycopersicum*. Phenolics and saponins were extracted from alfalfa and purified by open column chromatography. Elution with increasing methanol concentrations allowed to obtain both fractions. Prosapogenins were then prepared by alkaline hydrolysis of saponins. All the extracts were characterized by UPLC.

Saponins and prosapogenins were evaluated for *in vitro* activity (antigerminative and growth inhibition) against the pathogenic fungi *Alternaria alternata* and *Botrytis cinerea*. Antiviral activity *in vivo* against tomato spotted wilt orthotospovirus (TSWV) was assessed in plants inoculated with TSWV and treated with extracts. Plants were weekly monitored for symptom expression and sampled for TSWV titre measurement by real-time RT-PCR until the 31st day *post-inoculum* (d.p.i.).

From UPLC characterization, phenolics showed a high amount of apigenin glycosides, saponins were mainly composed by bidesmosides of medicagenic and zanhic acids, and prosapogenins were rich in monodesmosidic derivatives.

In antifungal assays, prosapogenins were effective in inhibiting mycelial growth and conidia germination. Thus, this extract was further assayed against alternariosis and grey mold of tomato *in vivo*, showing significant control efficacy in a dose-dependent manner.

In the antiviral assay, the best performing extract was the phenolic mixture, which significantly decreased the virus titre compared to the untreated control at 17 and 31 d.p.i.

In the frame of envisaged integrated pest management to

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reduce the use of synthetic pesticides, these treatments represent potential biopesticides for the control of fungi and viruses in tomato crops. Further studies are foreseen to establish the effect of selected substances on the non-target organisms/beneficials *Phytoseiulus persimilis* and *Neoseiulus californicus*.

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HORTICULTURAL SUBSTRATE CONTAINING ACIDIFIED SOFT RUSH FAVOURS GROWTH OF TRICHODERMA ASPERELLUM T34

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European horticulture is facing a change of paradigm. Besides the reduction of the use of pesticides and fertilizers, peat replacement is one of the big challenges. Peat has been widely used as the main material in different horticultural substrates. However, the ecological impact of peat extraction is a matter of concern. Local natural biomass such as soft rush (*Juncus effusus*) clippings can be an alternative material, fulfilling the requirements of sustainability. After chopping and acidification with elemental sulfur, soft rush has been shown to be a promising material to partially replace peat in the production of the calcifuge ornamental *Chamaecyparis lawsoniana* 'Ellwoodii'. To study the capacity of this alternative material to harbor microbial biocontrol agents, we investigated the establishment and persistence of *Trichoderma asperellum* T34, a commercial strain, in a mixture of acidified soft rush and peat (1:2 volume ratio). Rooted cuttings of *C. lawsoniana* 'Ellwoodii' were planted in pots containing either the mixture or peat alone and placed in the greenhouse. Both substrates were treated with *T. asperellum* T34 (Asperello® T34 Biocontrol®) to reach the concentration of 1,0E+04 cfu/mL substrate. Control pots were not treated. Five and eleven weeks after application, the substrate was sampled. Colony forming unit counts on *Trichoderma* semi-selective medium showed higher populations of *T. asperellum* T34 in the mixture than in peat alone. Moreover, plants grown in the mixture had higher shoot fresh weight than those grown in peat alone. Soft rush contains more nutrients than peat and received additional S. This might explain the difference in growth. We found higher values of SO₄, P, K and Ca in the mixture compared to 100 % peat. In conclusion, horticultural substrate containing acidified soft rush favors the growth of *T. asperellum* T34. The results also show that this strain can develop very well in acidic conditions (pH 4).

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THE BEAUVERIA BASSIANA-TRICHODERMA AFROHARZIANUM CONSORTIUM PROTECTS TOMATO PLANTS FROM PATHOGENS AND PESTS

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In their natural habitat, plants interact with several microorganisms, some of which are potentially harmful as pathogens, while others are beneficial as they can promote plant growth and expand the immune response of the plant host. Among them, the fungi *Beauveria bassiana* and *Trichoderma afroharzianum* can colonize plant tissues in an asymptomatic way, triggering a plant growth-promoting effect. They have an extremely broad host spectrum, including tomato (*Solanum lycopersicum* L.), a species of great economic importance extensively cultivated worldwide. Despite the promising role of beneficial microbes in advancing agriculture towards eco-friendly practices and ensuring consumer health, our knowledge of their mechanism of action remains incomplete. The present research activity aimed to identify the molecular basis of the tomato plant response to biotic stress in the presence of the beneficial *B. bassiana*-*T. afroharzianum* consortium. In particular, a large-scale proteomic analysis revealed that *B. bassiana* and *T. afroharzianum*, taken individually, play a role in the activation of different strategies in response to the necrotrophic pathogen *B. cinerea* and the aphid *M. euphorbiae* but when in consortium, the two beneficial microbes seem to cooperate synergistically, strengthening defense responses and enhancing plant growth. Moreover, a hormone analysis revealed increased levels of different hormones, as those belonging to Abscisates, Jasmonates, Auxins and Cytokinins families, highlighting a further contribution of the beneficial consortium in plant growth and defense. Our results pave the way for further studies aimed at understanding the growing importance of these beneficial microbes in building resilient agroecosystems and mitigating the environmental impact of traditional agricultural practices.

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FIELD EVALUATION OF FOUR BACTERIAL BIOCONTROL AGENTS FOR YIELD-INCREASING EFFECT AND CONTROL EFFICACY AGAINST TWO FUSARIUM DISEASES ON WINTER WHEAT

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Fusarium crown rot (FCR) and fusarium head blight (FHB) were economically important diseases in most regions where winter wheat is grown in China. Yield loss to FCR and FHB

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varies from year to year, depending on the severity of the infection and the conditions, but is generally around 10 % and can, under the right circumstances, exceed 30 %. Effective green control measures are urgently needed to be developed for wheat production.

In order to develop green control measures for FCR and FHB, and improve wheat yield, four suspension concentrates (SCs) of Bacterial biological control agent (BBCA), *Tufang1*, *Tufang2*, *Tufang3* and *Tufang4*, were evaluated in winter-wheat fields including one saline-alkali trial site and three high-yield trial sites in 2023. The tested SCs were separately irrigated with a sprinkling irrigation system during the reviving stage of winter wheat at a dosage of 75 L/ha. Each tested SC was implemented in 1 ha plot, and corresponding controls were set up at each trial site.

The results indicate that all tested BBCAs had significant yield-increasing effects and control efficacy against the diseases. On the saline-alkali site, all BBCAs showed substantial yield increases of 41.0%, 81.3%, 62.9% and 46.7%, respectively. Among the three high-yield sites, *Tufang1* and *Tufang2* consistently showed stable yield-increasing effects, with rate ranges of 4.4%-10.4% and 9.4%-27.7%, respectively. At only one high-yield site where FCR and FHB occurred severely, *Tufang1* and *Tufang2* showed significant control efficacy against these diseases at 48.1%/67.4% and 100%/62.8%, respectively. At all four trial sites, *Tufang2* consistently increased the spike number per area and -1000grain weight by 27.7%/4.4%, 10.0%/1.8%, 22.3%/9.4%, and 10.0%/19.4%.

This research demonstrates the effective control of FCR and FHB, coupled with a significant yield increase on wheat through the use of BBCAs. Notably, *Tufang2* exhibits substantial promise for further industrial development.

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FIELD EVALUATION OF FOUR BACTERIAL FLOWABLE CONCENTRATES FOR SEED TREATMENT ON YIELD AND CONTROL EFFICACY AGAINST STALK ROT ON MAIZE

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Stalk rot (FSR) caused by *Fusarium* pathogens are a potential threat to production in most regions where maize is grown in China. Yield loss to FSR varies from year to year, depending on the severity of the infection and the conditions, but is generally ranging from 5 % to 10 % and can, under the right circumstances, be over 30 %. Effective green control measures are urgently needed to be developed for maize production.

Four flowable concentrates for seed treatment (FS) of Bacterial biocontrol agent (BBCA), *Tufang1*, *Tufang2*, *Tufang3* and *Tufang4*, were developed and evaluated for their crop safety with laboratory test, and control efficacy and yield improvement in summer fields including two saline-

alkali sites and two high-yield sites in 2023. The tested FSs subsequently coated with maize seed dressed with chemical pesticide (20 % thiram+carbofuran FS) separately before planting at 1:30 (w/w). Each tested FS was implemented in around 1 ha plot, and the seed coated with chemical pesticide were set as control at each site.

The results indicated that all FSs were safe to maize emergency and growth when the seed were coated at the dosage of 1:15 and below, and showed, under natural-pathogen-infection condition and at the 1:30 dosage, their control efficacy to FSR at 42.20%, 55.40%, 87.51%, and 84.41% separately in field. At the heavy saline-alkali site, all FSs had substantial biomass increases of 39.87%, 43.22%, 61.84%, and 75.65%. At median saline-alkali site, *Tufang1* and *Tufang3* showed grain yield increase by 7.1 % and 29.73 %. Among both high-yield sites, only *Tufang3* had an increase on grain yield up to 6.9 %. This research demonstrates the effective control of FSR, coupled with a significant biomass or yield increase on maize through the use of BBCA FSs. Notably, *Tufang3* exhibits substantial promise for further industrial development.

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ENDOPHYTIC BACTERIA FROM OLIVE DRUPES AS PLANT DEFENSE INDUCERS AGAINST COLLETOTRICHUM ACUTATUM IN OLIVE TREE

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Olive anthracnose (OA) is one of the most important diseases affecting olives worldwide and is caused by fungi belonging to the genus *Colletotrichum*. The predominant strains are associated with *C. acutatum* and *C. gloeosporioides* species complexes. Due to the withdrawal of several fungicides and the risk of pathogen resistance, finding more sustainable control measures of the disease, such as using biological control agents and plant-resistance inducers is considered essential. Some biological agents can act as inducers of plant defense mechanisms, while they can also combine more than one mode of action, which makes them more suitable for their use in agriculture. This study aimed to evaluate antagonistic endophytic bacteria from olive drupes against *C. acutatum* for their ability to induce plant defense mechanisms. The experiments were conducted on young olive trees treated with the bacterial antagonists and the pathogen. The expression of ten defense genes was evaluated by RT-qPCR. All four tested bacterial strains showed increased expression of genes associated with Pathogenesis-related proteins (*PR10*, *MpoI*) compared to controls, while the application of the bacteria isolates K13 (*Bacillus methylotrophicus*), B1 (*B. amyloliquefaciens*) and P8 (*Serratia* sp.) caused increased expression of genes related to biosynthetic pathways of phenylpropanoids and salicylic acid in leaves. Finally, the bacterial strain B1 also induced increased expression of the lipoxygenase (LOX) gene involved in the jasmonic acid biosynthetic pathway proteins. Increased expression was observed in seedlings that were not infected with the pathogen. Therefore, we conclude that

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the earlier establishment of these biological control agents in olive trees is potentially an effective way to control olive anthracnose, as it allows the microorganisms to settle and prepare the plant for the impending pathogen's infection.

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APPLICATION OF MICROBIAL BIOFERTILIZERS TO IMPROVE DROUGHT TOLERANCE, GROWTH PERFORMANCE AND PHYSIOLOGICAL PARAMETERS IN FRUIT SHRUB AGROECOSYSTEMS

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The intensity of climate changes has led to the increased exposure of agricultural land to drought, which leads to the loss of soil fertility and the availability of nutrients, having negative consequences on the morphology, physiology and biochemistry of the plant and finally on the productivity of the crop. Many strains of *Trichoderma* spp. are successful plant beneficial microbial inoculants for plants due to their ability to induce resistance to abiotic stress like drought and high temperatures and as biofertilizers /biostimulants capable of promoting plant growth.

This work investigated the effects of treatments with two selected *Trichoderma* strains and their combination as biofertilizers at blackberry on the plant growth, antioxidant enzymes, proline, assimilatory pigments and leaf nutrient content.

The study was performed to investigate the changes in the content of pigments (chlorophylls, carotenoids), proline, peroxidase and catalase and macroelements in the leaves of blackberry collected from the shrubs of Experimental Station Baneasa after *Trichoderma* treatments. Quantification of these compounds was made using specific analytical methods. A consortium of two mixed *Trichoderma* (*T. viride* and *T. asperellum*) showed the highest growth improvement efficiency. The *Trichoderma* inoculation increased blackberry leaf chlorophyll content, and nutrient content by 15.50%, 13.21% and increased antioxidant enzyme and proline compared with the control untreated.

These findings indicated that the application of *Trichoderma* alone and in combination exerts a positive impact on blackberry plants, demonstrating the efficacy of using microbial biofertilizers in increase resistance to abiotic stress and in promoting plant growth. Also results obtained were important reference for establish assessing the adaptation of blackberry to a high temperatures and drought.

P201

N-ACETYL-CYSTEINE: A POTENTIAL HEALTHY MANAGEMENT STRATEGY FOR PLANT DISEASES

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For several years, we have been exploring the use of a molecule in a sustainable and environmentally friendly way for managing bacterial diseases in citrus. N-acetylcysteine (NAC), with its dual function as an antioxidant and a disulfide bond breaker, emerges as a potential game-changer in defending plants against bacterial pathogens, including *Xylella fastidiosa*, *Xanthomonas citri*, and *Candidatus Liberibacter*. With satisfactory results obtained, we expanded our research to include other crops such as grapes, potatoes, corn, and soybeans. This work illustrates the action of NAC and the benefits derived from its use. We developed fertilizers containing NAC and examined their effectiveness in reducing oxidative stress and improving the plant's antioxidant defense. This, in turn, led to a reduction in fruit drop, an increase in productivity, and, ultimately, an alleviation of HLB symptoms. In citrus orchards afflicted with HLB and a severity of 70 %, disease progression was 7 % less after one year of treatment. The fruit drop in treated plants was 45 % less, and plants treated with NAC produced an additional 10.8 kilograms of fruit per plant. For managing bacterial canker of grapevines (BCG), our tests conducted in a commercial field demonstrated a reduction of the disease on leaves and stems by 60 % and 70 %, respectively, compared to copper treatments, and an increase of 15 % in productivity. These insights have the potential to revolutionize sustainable disease management strategies, offering a promising avenue for healthy plant cultivation and disease control.

P202

EVALUATING THE POTENTIAL ROLE OF DEBARYOMYCES HANSENII ENHANCING PLANT NUTRITION AND NATURAL DEFENSE IN CUCUMBER PLANTS

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Soil yeasts alongside plant-growth-promoting rhizobacteria (PGPR) or fungi (PGPF) are recognized as valuable contributors to plant growth promotion, plant-enhanced nutrition and defenses, and soil conditioning. In this context, iron (Fe) emerges as a primary nutritional limitation for crops, particularly in soils with high calcium carbonate content,

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where its solubility is notably reduced. Ongoing research has uncovered that two yeast species, *Debaryomyces hansenii* and *Hansenula polymorpha*, exhibit the ability to upregulate genes associated with Fe acquisition and induce morphological adaptations in plants grown under hydroponic conditions. Thus, this study aimed (i) to delve deeper into the potential of *D. hansenii* as Fe biofertilizer, and (ii) to explore the potential induction of systemic resistance in cucumber plants under natural sterile or non-sterile calcareous soil conditions. To this end, cucumber (*Cucumis sativus* L. cv Ashley) plants growing in pots were inoculated by irrigating with a cell suspension of *D. hansenii* to reach a final concentration of 10^8 cells g^{-1} of soil at transplant and at 20 days after transplant. Two sampling times were considered (20 and 28 days after transplant) where the total uptake of Fe and other elements like copper (Cu), manganese (Mn), zinc (Zn), and phosphorus (P), dry weight, number of flowers, and the expression of ethylene (ET) and salicylic acid (SA) related genes in the plants, were determined. The inoculation with *D. hansenii* significantly enhanced Fe uptake in non-sterile soil, whereas Cu, Mn, Zn, and P uptake was significantly enhanced under both soil conditions. Moreover, inoculated plants displayed increased dry weight, and upregulated expression of ET- and SA-related genes, as indicators of the induction of plant-induced defense mechanisms (ISR and SAR). These findings highlight the potential contribution of *D. hansenii* to sustainable and environmentally friendly integrated crop management by stimulating plant mineral uptake and by possibly activating ISR and/or SAR.

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P203 GENETIC DETERMINANTS OF COUMARIN RESISTANCE IN PLANT-ASSOCIATED PSEUDOMONADS

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Plants co-inhabit the soil with millions of microbes, the composition of which determines plant health. To communicate with and shape this community, plants produce and secrete a multitude of metabolites via the roots. Among these root-secreted metabolites, coumarins have been studied in recent years for their role in alleviating Fe-deficiency in dicots. Current research has demonstrated that these compounds can direct the rhizosphere microbiome composition, affecting plant health. More specifically some plant beneficial rhizobacteria such as *Pseudomonas simiae* WCS417 (WCS417) are coumarin resistant, while certain plant pathogens are not. Nevertheless, the mechanism by which coumarins shape the microbiome is unknown. We

hypothesize that bacterial resistance to coumarins is a strong determinant of coumarin-induced changes in microbiome composition, allowing bacteria to competitively colonize coumarin-secreting plants. To study this, we screened a genome-sequenced *Pseudomonas* library for their resistance to scopoletin and fraxetin, the main secreted coumarins during Fe deficiency. Comparing the WCS417 genome with other *Pseudomonads* might thus lead us to identify genes involved in this resistance. We found that fraxetin affected growth of *Pseudomonads* variably, while this genus seems generally resistant to high concentrations of scopoletin. We identified orthologous genes between the *Pseudomonads* using Orthofinder and investigated the presence of certain orthogroups in relation to coumarin resistance phenotypes with Scoary2. We identified candidate genes for fraxetin resistance, but not for scopoletin resistance. To establish the relevance of these genes in a microbial context we will cross reference them with publicly available microbiome data, to assess whether their abundance increases in microbial communities exposed to plant-secreted coumarins. This will allow us to identify plant and genetic bacterial factors that determine plant microbiome selection, opening the door for new ways of screening microbes and constructing plant-beneficial bacterial inocula.

P204 BIOLOGICAL CONTROL OF COLLETOTRICHUM ACUTATUM CAUSING OLIVE ANTHRACNOSE, WITH THE USE OF ESSENTIAL OILS

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A serious threat to olive tree cultivation (*Olea europaea* L.) is the fungus *Colletotrichum acutatum*, considered the primary causal agent of anthracnose disease and responsible for epidemics in olive growing countries. The pathogen mainly infects olive drupes causing fruit rot and subsequently fruit drop and mummification. This leads to significant yield losses and degradation of olive oil quality. Several fungicide treatments are used to control the disease, which underlines the need for alternative environmentally friendly managing methods of olive anthracnose. Aim of this study was the *in vitro* evaluation of essential oils (EO) of geranium (*Pelargonium graveolens*), thyme (*Thymus vulgaris*), cinnamon (*Cinnamomum zeylanicum*), and peppermint (*Mentha piperita*), regarding their ability to inhibit mycelial growth and conidial production of the fungus *C. acutatum* in amended PDA medium with five concentrations of each EO. Furthermore, the effectiveness of these EOs in reducing disease severity and conidial production was also evaluated on detached olive drupes of cv. Kalamon, after their immersion in different concentrated EOs solutions and their infection with the pathogen. Disease severity was assessed using a fruit rot percentage scale to create a disease progression curve and calculate AUDPC for each application. Cinnamon oil was found to be more effective, reaching inhibition percentages as high as 100 %, with the rest of the EOs also showing high efficiencies. Finally, it was investigated by RT-qPCR whether treating olive trees with a

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solution of cinnamon EO modifies the expression of the *PR-1O*, *Bglu*, *Mpol*, and *LOX* genes involved in the plant's defense mechanism. Results showed that *PR-1O* and *Mpol* genes were overexpressed on the cinnamon EO treated plants.

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EVALUATION OF BIOLOGICAL CONTROL EFFICACY OF A *BACILLUS HALOTOLERANS* STRAIN AGAINST *VERTICILLIUM* WILT OF EGGPLANT UNDER GREENHOUSE CONDITIONS

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Verticillium wilt, caused by the soilborne fungus *Verticillium dahliae*, is a devastating disease affecting a wide range of crops, including eggplant (*Solanum melongena*), leading to significant yield losses worldwide. Due to the ineffective chemical treatments to control this pathogen, biological control agents offer a sustainable and environmentally friendly alternative. *Bacillus halotolerans* Cal.I.30 has been isolated endophytically from the roots of *Calendula officinalis* with proven antagonistic activity against phytopathogenic fungi *in vitro* and *in planta*. In this study, we evaluated the efficacy of this *B. halotolerans* strain as a biological control agent against Verticillium wilt of eggplant under greenhouse conditions. The experiment was conducted in a randomized complete block design with four treatments: i. *B. halotolerans* Cal.I.30-amended soil, ii. *Verticillium dahliae*-infested soil, iii. *Bacillus halotolerans* Cal.I.30 + *Verticillium dahliae*-infested soil, and iv. untreated control. Eggplant seedlings were transplanted into pots with the respective soil treatments and maintained under greenhouse conditions. Disease severity and plant growth parameters were evaluated at regular intervals over the course of the experiment. Our results demonstrated that the application of Cal.I.30 significantly reduced the severity of Verticillium wilt symptoms in eggplants compared to the *Verticillium dahliae*-infested soil treatment. Plants treated with *B. halotolerans* exhibited improved growth parameters, including plant height, fresh weight, stem diameter, leaf area index, wilting, yellowing and defoliation during plant growth, indicating enhanced plant vigor and resilience against pathogen infection. Our findings suggest that Cal.I.30 is a promising biological control agent for managing Verticillium wilt of eggplant under greenhouse conditions. Further research is underway to explore the mechanisms underlying the antagonistic activity of *Bacillus halotolerans* Cal.I.30 against *Verticillium dahliae* and its potential for field application in integrated disease management strategies.

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EFFECT OF BIOCONTROL AGENTS AND RESISTANT INDUCERS AGAINST *FUSARIUM OXYSPORUM* F.SP. *LACTUCAE* ON LETTUCE IN A SIMULATED CLIMATE CHANGE SCENARIO

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Climate change may affect the incidence and severity of plant diseases as well as the efficacy of disease management measures; this aspect is less investigated, especially considering the combined impact of increased temperature and elevated CO₂. Trials were carried out under phytotron conditions on the lettuce-*Fusarium oxysporum* f. sp. *lactucae* (race 1) pathosystem to test the impact of temperature (-20 24°C, 28-24°C, 32-28°C) combined with increased CO₂ levels (850-800 ppm), compared to standard CO₂ air concentrations (450-400 ppm), on resistant inducers (calcium oxide and potassium phosphite) and commercially available BCAs, that is, *Streptomyces griseoviridis*, *Trichoderma asperellum* and *Beauveria bassiana*. The disease reduction provided by the tested products was found to be influenced by the disease pressure, with temperature being the main factor that influenced Fusarium wilt severity in the inoculated and untreated controls. Potassium phosphite generally provided consistent disease control under all the tested conditions, and resulted to be more effective at 24-20°Cx 450-400 ppm of CO₂ (76 % efficacy) than at 800 ppm of CO₂ (47 % efficacy), while calcium oxide provided the greatest lettuce wilt severity reduction (43% - 46.2% efficacy) at 20-24°C, with the tested CO₂ concentrations having no significant effect. Among the considered BCAs, *B. bassiana* provided consistent disease control at 24-20°C (53-56 % efficacy) for both CO₂ concentrations, while increases in CO₂ significantly improved the efficacy of *S. griseoviridis* across this temperature range. *T. asperellum* was inconsistent in the biocontrol of the pathogen under all tested environmental conditions. Potassium phosphite and *B. bassiana* provided the best Fusarium wilt reduction, that is, of 23-35 % and 28-32 %, respectively, for the 28-24°C temperature range and for both the tested CO₂ concentrations, while calcium oxide was only effective for the standard CO₂ levels (30 % efficacy). None of the tested BCAs were effective at 28-32 °C with 400-450 ppm of CO₂. More attention should be paid to evaluating the control efficacy of resistant inducers and biocontrol agents considering the predicted climate changes.

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ASSESSING THE POTENTIAL IMPACT OF NO FUNGICIDE SEED TREATMENTS ON COMMON WHEAT AND MAIZE YIELD QUANTITY AND QUALITY IN FRANCE

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Most, if not all, arable crops grown in the conventional system undergo fungicide seed treatments (FSTs) before planting, worldwide. In the EU, the planting of fungicide-treated seeds is considered in the calculation of treatment frequency index (i.e. TFI = 1). As an instrument of the EU's common agricultural policy, the EU offers financial incentives for farmers who are able to decrease the TFI in their cropping systems. Therefore, reducing the TFI related to FSTs will provide direct (lower seed costs in the absence of treatments) and indirect (financial incentives in return for adopting low-input farming practices) benefits for farmers. Decreasing availability of conventional fungicides previously used for FSTs due to their ban (e.g. recent ban on Metalaxyl-M for outdoor use) calls for alternatives to conventional crop protection. However, there are knowledge gaps as to whether the absence of FSTs will lead to yield losses, which may jeopardize the economic viability of arable farms. The key objective of this study was to evaluate economic and environmental impact of arable cropping systems without FSTs compared to the conventional systems using FSTs, with a particular emphasis on yield quantity and quality. A -6year on-farm research was conducted across 26 experimental sites in north-eastern France using key arable cropping sequences (wheat, maize, barley, oilseed rape etc.). For most of the experimental sites, we found no yield quantity and quality (wheat bunt and mycotoxin contamination) differences between the treatments with and without FSTs. Our results provide a key reference for arable farmers in France who are willing to significantly reduce or not use FSTs at all. No yield penalty in the absence of FSTs represents a win-win solution for farmers in terms of reducing production costs while also ensuring access to financial incentives due to reduction or elimination of FSTs in their cropping systems.

Samples were analyzed using the double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) with a commercial kit from BIOREBA AG (Switzerland) for following viral infections:

1. Apple mosaic virus (ApMV)
2. Apple chlorotic leaf spot virus (ACLSV)
3. Apple stem pitting virus (ASPV)
4. Apple stem grooving virus (ASGV)
5. Plum pox virus (PPV)
6. Prunus necrotic ringspot virus (PNRSV)
7. Prune dwarf virus (PDV)
8. Strawberry latent ringspot virus (SLRSV)

A total of 690 samples were collected from 8 leading local and introduced apple cultivars, 2 plum and 7 peach cultivars in the orchard of SRCA Jighaura village in Mtskheta Municipality. Research results showed that ASGV are predominant in local apple cultivars 'Kekhura' and 'Achabetura' (10.6 %) following ASPV virus in the cultivar 'Georgian Sinapi' (8.4 %). While, ACLSV showed minor infection (3.3 %) in the 'Winter banana' cultivar.

In stone fruits, the PDV was the primary infection source for a total of 6.6 % among tested cultivars. The prevalence of PNRSV, SLRSV, PPV and SLRSV viruses was recorded with equal intensity (total 3.3 %). Only PVD virus was detected within of plums, more specifically in "Guldedava" (8.3 %).

Interestingly that, the ApMV virus, which is basically is more common in apple orchards, was not detected in sampled apple trees, but its presence was observed in collection of hazelnut cultivars (5.2 %). Based on the research findings, a proper scheme for virus-free mother stock fruit trees was established.

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ASSESSMENT OF VIRAL PATHOGENS IN THE POME AND STONE FRUIT CROPS IN EAST GEORGIA

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Fruit trees are constantly exposed to the attack of viruses, this risk is present both in the field and during their vegetative propagations. Testing viral infection is a unique possibilities to reveal all the present pathogens and prevent their widespread and yield loss.

A comprehensive study was conducted to investigate viral diseases in peach, apple, and plum trees at nursery and ex-situ collection orchards in East Georgia.

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STUDY OF HAZELNUT VIRAL INFECTIONS IN GEORGIA

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Hazelnut culture is one of the important crops in Georgia. There is a high risk of spreading hazelnut diseases due to introduced varieties. In recent years, symptoms of a viral disease have appeared. Viral infection requires strict monitoring and laboratory testing to avoid further distribution of viral infections.

The study aimed to investigate two viruses that mainly infect the hazelnut: Prunes necrotic ringspot virus (PNRSV) and Apple mosaic virus (ApMV) using serological double-antibody sandwich enzyme-linked immunosorbent (DAS- ELISA) assay for the samples collected in the east and west part of Georgia. This investigation was carried out in 2019-2023 years.

Absorbance values as the results obtained from the DAS-ELISA were determined at 405/450 nm using ELX800 microplate reader (Bio-Tek Instruments, Winooski, VT). The samples were considered positive if their ODs were three times higher than the negative control.

The visual observation was carried out for 1000 hazelnuts in several Municipalities of west (Khobi, vill. Nojikhevi; Zugdidi, vill. Darcheli) and east (Mtskheta, vill. Jigaura) part of Georgia. 377 symptomatic plants were collected for laboratory testing. 139 samples from Darcheli, 52 from Nojikhevi, and 186 from Jigaura.

It was revealed that the most infected region is Mthkheta vill. Jigaura (18.2 %), following Zugdidi municipality, vill. Darcheli (7.9 %) and the minor infection rate was shown in the Khobi municipality (4.24 %).

PNRSV infection was characterized by relatively high distribution in Mthkheta (8.6 %), the infection rate in Zugdidi achieved only 2.2 %. Nevertheless, no infection rate was relived in Khobi.

A total 11.2 % hazelnut samples tested positive for ApMV in Mthkheta and only this viral infection was shown in the Nojikhevi region of Khobi (4.24 %) as well.

Thus, the distribution of PNRSV and ApMV viral infections in hazelnut was shown for the first time in several regions of Georgia.

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EPIDEMIOLOGY OF CERATOCYSTIS WILT OF EUCALYPTUS GRANDIS IN URUGUAY

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Eucalyptus grandis and its hybrids provides a mainstay for the forestry industry of Uruguay. These trees, established in plantations, are affected by various diseases, the most serious of which are caused by introduced pathogens. In 2003, a serious disease problem arose on trees shortly after they had been pruned. Isolations from the discolored wood associated with the pruning wounds consistently yielded cultures of a *Ceratocystis* species that was determined as the causal agent of the disease. The aim of this study was to determine whether pruning in different seasons was related to infection. The association of *Ceratocystis* infections with physical wounds was considered by artificially wounding trees, monthly for 22 consecutive months in three different northern Uruguay plantations. The wounds (five per plantation) were made by exposing a 100 cm² area of cambium and then making 5mm deep slits at the centre of wounds to expose xylem tissue. These wounds were inspected for discoloration after 30 days and isolations were made from infected tissue. In addition, surveys were made for wilting trees in 29 *E. grandis* plantations in northern Uruguay and trees displaying symptoms were sampled for *Ceratocystis* infections. No infections were found on the artificially induced wounds during winter months or when there was less than 150 mm of rainfall. *Ceratocystis* wilt was observed on trees in 25 % of the plantations surveyed and never on trees pruned during the winter. Overall, the results of the study suggest that *Ceratocystis* wilt can be avoided by pruning trees in winter months.

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GENETIC AND PATHOGENICITY VARIABILITY OF *Erwinia amylovora* A RE-EMERGING BACTERIUM CAUSING FIRE BLIGHT ON APPLE

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Erwinia amylovora is a well-known bacterium responsible for causing fire blight on apples and pears globally. Although this disease has been reported as sporadic in the north of the country, an outbreak has been stated recently affecting apples in the main producing regions of Mexico. This work was aimed at determining whether genetic or pathogenicity variability occurs in the *E. amylovora* population. Thirty-one isolates were recovered from leaves and fruit exhibiting fire blight; they were assessed for pathogenicity on apple fruit var. Gala by injecting 10⁵ CFU mL⁻¹ and maintaining under greenhouse conditions. Results indicated that 23 isolates were pathogenic after 6 days post-inoculation. All pathogenic isolates were

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identified by multilocus phylogenetic analysis of the 16S rDNA, *gyrB*, and *rpoB* through phylogenetic reconstruction using Bayesian inference. Results indicated that 23 isolates were grouped with *Erwinia amylovora* type strain LMG 2024. The phylogenetic tree shows five subgroups. Additionally, all pathogenic isolates were analyzed for biofilm production; they produced different amounts of biofilm, but no correlation was found between pathogenicity and biofilm production. These results make it important to establish disease management measures before the beginning of flowering in order to reduce the damage caused by the bacterium during fruit formation. Also, to reduce the spread of more virulent strains to other plantations.

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MANAGING HULL ROT IN AUSTRALIAN ALMOND ORCHARDS: INSIGHTS AND STRATEGIES

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Hull rot poses a significant challenge to almond cultivation in Australia, leading to direct yield losses and quality degradation. This disease impacts nut production through infected nuts, while twig dieback and death of fruiting spurs further exacerbate yield reductions in subsequent years. This disease affects all production regions of Australia except Western Australia, with onset occurring as early as January during the vulnerable stage of early hull split. *Rhizopus sp.* is the key pathogen causing hull rot in Australian almond orchards. Recent trends indicate that an increase in rainfall during hull split, potentially attributed to climate change, further amplifies the susceptibility of almonds to hull rot. Our research aims to address this challenge by focusing on effective management strategies. Our findings show some promising insights. Evaluation of Australian breeding material, encompassing 19 advanced breeding lines, revealed lower hull rot incidence compared to the industry standard Nonpareil. Assessment of an established rootstock trial demonstrated that Nonpareil grafted onto Peach Almond (*P. persica* x *P. dulcis*) rootstocks, particularly Monegro, Garnem, Hansen, and Felinem, exhibited increased susceptibility to hull rot. In our investigation into water and nitrogen management we observed that maintaining a sustained deficit water schedule can reduce disease severity, while nitrogen levels showed no significant impact on hull rot development. Management trials demonstrated that while current chemical treatments can mitigate hull rot, they do not completely eradicate the disease. Timely application of protectant treatments during hull split emerges as a key strategy, particularly in high-risk areas. Through this research, we aim to provide almond growers with effective tools and strategies to manage hull rot, ensuring the sustainability and productivity of Australian almond orchards.

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THE COMPREHENSIVE EVALUATION OF LITHUANIAN FARMER'S KNOWLEDGE OF STRAWBERRY GROWING SYSTEMS

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Strawberry diseases cause a significant threat to crop yield, resulting in substantial losses. The inefficient use of pesticides and fertilisers escalates costs and contributes to environmental concerns. Therefore, it's important to evaluate farmer's understanding of harmful organisms and the use of alternative plant protection methods. Reducing the usage of fertilizers and pesticides is one of the EU Green Deal's objectives, highlighting the need for targeted plant protection. We aimed to evaluate the knowledge level among Lithuanian farmers regarding plant diseases and propose recommendations based on their insights. The survey data revealed that all participating farmers were familiar with various plant diseases. Approximately 61.90 % of farmers were aware of plant diseases and knew where to gather further information. Some farmers use disease forecasting models (5.13 %) and consultant adviser services (15.38 %), while the majority rely on their personal experience (64.10 %) to evaluate plant diseases. Only 9.52 % of farmers had agricultural education, with many acquiring knowledge through specialised courses (33.33 %) or consultant services (21.43 %). The 11.76 % of farmers use biological plant protection products. Alternative methods mentioned by farmers were such as mechanical weed removal (29.49 %), elimination of infected plants (23.53 %), and the use of mulches (21.57 %). However, 59.09 % of respondents think biological plant protection products are efficient but not as chemicals. The most popular strawberry cultivars are Malwina (20.83 %), secondary Rumba (18.75 %), and then Asia (16.67 %). This data comprehensively evaluates farmers' knowledge levels and shows areas that may require further improvement and education. Acknowledgement. This project has received funding from the Research Council of Lithuania (LMTLT), agreement No [S-NORDFORSK6-23-].

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IMPACT OF SOYBEAN RED CROWN ROT ON NODULE HEALTH

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Soybean (*Glycine max*) is one of the most important crops, and soybean seeds are rich in vegetative oil and proteins to support food sustainability worldwide. With a high demand of N input for producing protein in seeds, soybean forms nodules with N₂-fixing bacteria such as *Bradyrhizobia* to acquire extra N from biological N₂ fixation. In addition to N₂ fixation, *Bradyrhizobia* in nodules may also perform denitrification to reduce NO₃⁻ to N₂, with an intermediate compound N₂O, which is a greenhouse gas with a great concern. Red crown rot (RCR) is an emerging soilborne disease caused by *Calonectria ilicicola*, threatening the global soybean production, and limited

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study has focused on how RCR may affect the soybean nodules and the N_2O content in soybean nodules. Therefore, this study investigates the impact of RCR on soybean and nodule health. The results indicate that the infection of *C. ilicicola* reduces the nodule number and size, meanwhile increases the concern of N_2O production. Advanced studies are ongoing to further elucidate the mechanistic insight of RCR influencing not only the soybean nodule health, but also the ecological net-zero ambition.

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TWO4TANGO: A NOVEL TOOL FOR EXPLORING BIOSYNTHETIC GENE CLUSTER CO-OCCURRENCE ACROSS BACTERIAL GENOMES

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Many natural products (NPs) used in agriculture, medicine or biotechnology are derived from microorganisms. The genes encoding the enzymes that produce these NPs usually lie clustered on genomes in so-called Biosynthetic Gene Clusters (BGCs). Classical genome mining strategies for discovering novel NPs mostly assume that they are produced from a single BGC. However, some NPs require the cooperation of multiple genomic loci for their biosynthesis. Here, we developed an innovative computational strategy for the identification of such co-occurring BGCs across bacterial genomes, which may represent novel multi-locus biosynthetic pathways. BGCs are first identified using antiSMASH and then clustered into Gene Cluster Families (GCFs) by using BiG-SCAPE. Then, the two4tango algorithm utilizes a combination of statistical metrics, including co-occurrence patterns and phylogenetic distance, to identify significant associations between GCFs. Associations with minimal lineage dependency are of particular interest, as they may suggest conserved co-occurrence patterns with potential functional significance. As a proof of concept, the tool has been tested on all available genomes in NCBI belonging to the genera *Photobacterium*, *Xenorhabdus* and *Pseudomonas*. In this analysis, a total of 37 pairs of co-occurring GCFs were identified by two4tango. Currently, site-directed mutagenesis and metabolomic analysis are ongoing to further validate these predictions. Unraveling co-occurring BGCs using two4tango may provide key insights into microbial NP biosynthesis, guiding future

endeavors towards experimental validation. This approach could elucidate their functional significance, ultimately advancing our understanding of complex biosynthetic pathways and facilitating the discovery of novel bioactive compounds with potential applications in agriculture, medicine, and biotechnology.

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BACLIFE: A USER-FRIENDLY COMPUTATIONAL WORKFLOW FOR GENOME ANALYSIS AND PREDICTION OF LIFESTYLE-ASSOCIATED GENES IN BACTERIA

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Bacteria have an extensive adaptive ability to live in close association with eukaryotic hosts, exhibiting detrimental, neutral or beneficial effects on host growth and health. However, the genes involved in niche adaptation are mostly unknown and their functions poorly characterized. Here, we present bacLIFE (<https://github.com/Carrión-lab/bacLIFE>) a streamlined computational workflow for genome annotation, large-scale comparative genomics, and prediction of lifestyle-associated genes (LAGs). As a proof of concept, we analyzed 16,846 genomes from the (*Para*) *Burkholderia* and *Pseudomonas* genera, which led to the identification of hundreds of genes potentially associated with a plant pathogenic lifestyle. Site-directed mutagenesis of 14 of these predicted LAGs of unknown function, followed by plant bioassays, showed that 6 predicted LAGs are indeed involved in the phytopathogenic lifestyle of *Burkholderia plantarii* and *Pseudomonas syringae* pv. phaseolicola. These 6 LAGs encompassed a glycosyltransferase, extracellular binding proteins, homoserine dehydrogenases and hypothetical proteins. Collectively, our results highlight bacLIFE as an effective computational tool for prediction of LAGs and the generation of hypotheses for a better understanding of bacteria-host interactions.

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SYNTHESIS OF ENVIRONMENTALLY SAFE PLANT RESISTANCE INDUCERS AS AN ALTERNATIVE TO PESTICIDES FOR PROTECTING APPLE TREES

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For many years, Poland has been a leader in apple production in the EU (4 million tons in 2021) and invariably holds third place in the world apple production after the USA and China. The diseases that cause severe damage to apple cultivation are (1) powdery mildew caused by the fungus *Podosphaera leucotricha*; (2) apple scab caused by the fungus *Venturia inaequalis* and (3) fire blight caused by the bacteria *Erwinia amylovora*. Plant protection products containing active substances with different modes of action are approved to control the aforementioned diseases. However, these active substances have certain limitations due to their mechanism of action and negative environmental impact. Moreover, the European Union, aware of the dangers resulting from the excessive use of pesticides, introduces new legal regulations that eliminate or limit certain chemical substances.

An alternative to protection provided by pesticides, which act directly towards pathogens, is the use of systemic acquired resistance (SAR) inducers that act on plant metabolism instead of pathogens. Our goal was to synthesize new plant resistance inducers based on naturally occurring salicylic acid, in ionic form, where counterion was choline, to control the diseases in apple tree cultivation. This method can be used in the future as an environmentally friendly and effective method to protect apples.

The "Searching for new chemical compounds inducing resistance of apple to diseases and determination of the molecular mechanism of their action" project is carried out within the Sonata (UMO47/2022-D/NZO2327/9) programme of the National Science Center, Poland

wherein the plant undergoes a preconditioning period to positive-low temperatures—to bolster its hardiness. Age, health, and tissue condition influence this tolerance, with olive leaves often proving more susceptible than shoots. We aimed to assess frost tolerance in 18 principal olive cultivars by combining acclimation dynamics through visual and fluorometric analysis. A predefined acclimation protocol was implemented, subjecting 10 one-year-old plants per cultivar during 6 weeks at 8-10°C. Visual assessments were conducted following a standardized frost protocol, wherein 20 detached leaves per cultivar were exposed to -10°C for 30 min. Damage incidence (%) and leaf mortality (%) were visually quantified after 72 h. Concurrently, a parallel experiment was conducted using non-acclimated plants under greenhouse conditions (Figure 1).

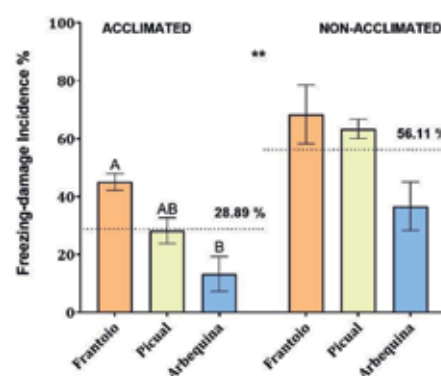


Figure 1. Visual evaluations of the freezing damage incidence of three main olive cultivars after following a frost protocol on detached leaves.

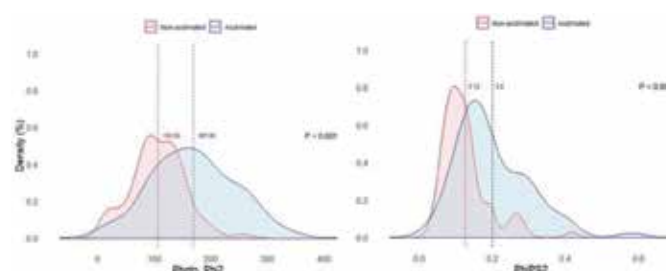


Figure 2. Olive cultivars density plots comparing chlorophyll fluorescence values measured in-field using PhotosynQ (Photo_Phi2) and LICOR LI-600 (PhiPS2).

Subsequently, plants were transferred to an olive plot prone to light frost events during the winter. Two months later, chlorophyll fluorescence was measured in-field using PhotosynQ (MultispeQ V2.0) and LICOR LI600- (LICOR, Lincoln, USA) equipment to detect variability in freezing damage among cultivars and acclimation status (Figure 2). The culmination of our current findings, alongside data on in-field freezing damage incidence and resprouting capacity in the following spring, promises to provide valuable insights for the accurate classification of major olive cultivars in terms of frost tolerance. Thus, paving the way for improved cultivar choice and management strategies in frost-prone environments.

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EVALUATION OF FROST TOLERANCE IN OLIVE (OLEA EUROPAEA L.) CULTIVARS: INTEGRATING ACCLIMATION DYNAMICS THROUGH VISUAL AND FLUOROMETRIC ANALYSIS

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The olive (*Olea europaea* L.) is an evergreen woody crop with notable cultivar variability to frost tolerance, yet poorly characterized. Extended sub-zero exposure can damage plants, emphasizing the vital role of acclimation—a process



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DISSECTING THE WALNUT MOLD AND ITS MANAGEMENT IN CALIFORNIA

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The first description of walnut mold dates back in the 1970s. Early research has shown that delaying harvest and relative humidity of 100-85 % increase the risk of mold development. Those days, the predominant cause of mold was identified as *Penicillium* spp. In 2004 and 2005, after dissecting 4,000 walnuts, we determined that nuts have more mold when: a) are infested by the navel orangeworm, b) when are sunburned, c) when have shriveled husks, d) when show hull rot, e) when have large stem opening, f) when they stay on the ground (and/or on the trees) for more than 24 hours after tree shaking. During 2018 to 2022 high levels (10 to 30 %) of mold have been reported. This study was initiated to find out the causes of mold, factors affecting mold development, and perform trials to manage it. The following fungal species were determined: *Fusarium proliferatum*, *F. verticillioides*, *Fusarium solani*, *Alternaria alternata*, *A. tenuissima*, *A. solani*, *Aspergillus niger*, *Botryosphaeria dothidea*, *Neofusicoccum mediterraneum*, *Lasiodiplodia theobromae*, *Neoscytalidium dimittatum*, *Phomopsis theicola*, *P. rhushicola*, and *Nigrospora* spp. The fact that *Alternaria* and *Fusarium* were isolated from the stigma of female flowers, the incidence of the origin of the fungal colony causing mold being greater at the stylar end and the higher incidence of mold after inoculating flowers of walnut with *Alternaria* and *Fusarium* suggest that at least some infection can initiate at bloom. The strong correlation of fungal infections of hulls with the fungi causing mold also suggests that some mold develops during fruit maturation. A fungicide spray at bloom and/or at pre- and during early-hull split stage reduced significantly walnut mold. Especially, spray(s) at the pre-hull-split and during hull-split stages reduced mold by 30 to 70 %. Fungicides used include, trifloxystrobin+tebuconazole (Luna Experience), frutriafol (Rhyme), and pyraclostrobin+fluxapyroxad (Merivon), and tebuconazole (Tebucon).

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BIOPROTECTION OF OLIVE TREES AGAINST VERTICILLIUM WILT BY POMEGRANATE AND CAROB EXTRACTS, AND NATURAL COMMERCIAL PRODUCTS

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Verticillium wilt of olive (*Olea europaea*; VWO), caused by the soil-borne pathogen *Verticillium dahliae*, is one of the major diseases of this crop. Its wide host range, high survival capacity and the ineffectiveness of chemicals makes the control of VWO a phytopathological challenge. Search for environmentally friendly alternatives such as bioprotectors based on plant extracts and natural products that can act as host resistance inducers is an urgent demand. The goal of this study was to evaluate the effect of pomegranate and carob extracts obtained in the laboratory and ten commercial natural products against VWO. Extracts based on pomegranate peels and carob pods and leaves were obtained by methanol, ethanol, and ethyl acetate in the laboratory. Commercial products based on seaweed, horsetail, mimosa and oak, olive, willow, and chitosan extracts, aluminum lignosulfonate, and a mixture of two products were selected. A copper phosphonate salt was used as reference control. The effect of all extracts and products on mycelial growth, conidia germination and microsclerotia viability of *V. dahliae* was evaluated *in vitro*, and their effect on disease progress in olive plants of cv. Picual grown on a soil artificially infested with *V. dahliae*. Pomegranate and carob extracts from methanol or ethanol significantly reduced the viability of *V. dahliae* structures as well as the disease severity in inoculated olive plants when they were applied by root irrigation, always at 3 g/l. Most of the products inhibited *V. dahliae* conidia germination up to 90%, but only seaweed and willow extracts, and the copper phosphonate salt inhibited *V. dahliae* mycelial growth up to 50% at the high commercial dose. Root treatments with willow extract significantly reduced the disease severity in inoculated olive plants. Pomegranate, carob and willow extracts may be considered as effective bioprotectors against VWO and should be further studied as potential inducers of host resistance.

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P02IVS

RESISTANCE TO VERTICILLIUM DAHLIAE OF DIFFERENT PISTACIA ROOTSTOCKS IN A HEAVILY INFESTED SOIL

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Verticillium wilt of pistachio (*Pistacia vera* L.), caused by the soil-borne fungus *Verticillium dahliae*, is one of the most important diseases in this crop. To date, the use of resistant rootstocks is the most effective and economical control measure. The aim of this work was to evaluate the resistance of pistachio trees grafted on different Pistacia rootstocks against Verticillium wilt under field conditions. An experimental field of pistachio plants of cv. Kerman grafted on *P. atlantica*, *P. integerrima*, *P. terebinthus* and 'Platinum' and 'UCBI' hybrids was established in Granada (Southern Spain) in January 2019 in a soil heavily infested with *V. dahliae* non-defoliating pathotype (14.9 microsclerotia per gram of soil). The identity of the scion and all the rootstocks was confirmed by DNA analysis. The disease was monitored during 53 months after planting using a severity rating scale and the disease incidence and tree mortality were estimated. Pistachio trees grafted on *P. terebinthus* showed the highest severity values, followed by those grafted on *P. integerrima* four years after planting. Normally, *P. atlantica* is considered to be more susceptible to the disease than *P. integerrima*. However, in our study *P. atlantica* was found to be significantly more resistant to the disease than *P. integerrima*, demonstrating the high intraspecific variability within these species. Pistachio trees grafted on *P. atlantica*, 'Platinum' and 'UCBI' hybrids showed statistically equal disease severity. No pistachio trees grafted on the 'UCBI' hybrid died from the disease throughout the experiment. On the contrary, at the end of the trial, no plant grafted on *P. terebinthus* remained symptomless. This study highlights the importance of the rootstock selection prior to the establishment of pistachio orchards, as well as the intraspecific variability within Pistacia species.

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P03IVS

DEVELOPMENT OF A POWDER FORMULATION BASED ON *BACILLUS* SPP. FOR BIOLOGICAL CONTROL OF POTATO VERTICILLIUM WILT

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Potato Verticillium wilt (PVW) was an economically significant soil-borne disease in the main planting areas of China. Yield losses to PVW vary from 20% to 50%, significantly impeding the healthy development of the potato growing. There is an urgent need to develop safe and effective microbial agents for potato production. To develop *Bacillus* wettable powder (BWP) for controlling PVW, spores of *Bacillus subtilis* HMB26553 and *B. amyloliquefaciens* PHODG36 were used as the active ingredients. The types and dosages of adjuvants were screened through a toxin medium. The impacts of BWP on potato growth and its efficacy in controlling PVW were evaluated through pot and field tests, employing seed-dressing at different dosages of 15, 30 and 45 kg/ha. The results were demonstrated in various potato-growing regions using a dosage of 30 kg/ha. The optimal formulation of 3 billion CFU/g BWP was established as follows: 10% HMB26553 technical concentrate (TK), 10% PHODG36 TK, 1% wetting agent LT-569, 2% dispersant MF, 1% UV protective agent ascorbic acid, and talc powder 76%. Pot and field experiments revealed that seed-dressing with different tested dosages of BWP were safe for potato seedling emergence and growth. The control efficacy on PVW of all treatments ranged from 50.3% to 68.0%, and the yield increase rate was 4.7% to 11.6%. Notably, the treatment with 30 kg/ha exhibited the highest control efficacy and yield increase rate. Field trial demonstrations in different regions showed control efficacy on PVW ranging from 61.8% to 62.1%, accompanied by potato yield increases ranging from 8.7% to 33.4%. The results indicated that BWP at a concentration of 3 billion CFU/g could effectively control PVW and significantly increase potato yield, showcasing considerable potential for further industrial development.

P04IVS

CONTROL EFFICIENCY AND MECHANISM OF SPINETORAM SEED-PELLETING AGAINST THE STRIPED FLEA BEETLE *PHYLLOTRETA STRIOLATA*

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The striped flea beetle (SFB, *Phyllotreta striolata*) is an important pest of the cruciferous crops in Asia. SFB is regarded as the most destructive pest of cruciferous crops in China due to the severe crop loss and frequent infestation incidents. As no SFB resistant cultivar is available at present, therefore, application of insecticides is the primary method of SFB control. On the contrary, the exploitation of chemical

insecticides causes severe environmental issues and is not cost-effective. The use of a seed-pelletized coating of spinetoram effectively reduced SFB feedings on the flowering cabbage seedlings, whereas in combination with the insect-proof net, it controlled the SFB infestation throughout the cabbage growth period. The analysis of the pesticide residues in soil and different cabbage parts indicated the degradation dynamics of spinetoram. Furthermore, estimation of the half-life of spinetoram revealed that via seed-palletized application spinetoram half-life was found to be 2.82 days in soil, 4.21 days in the root, 5.77 days in the stem, and 3.57 days in the leaf, respectively. Both the lower pesticide residues and the half-life of spinetoram in soil and cabbage parts suggested it to be a promising environment and food-safe pesticide in controlling SFB. Moreover, the seed-pelletized coating ensured a sustainable release of spinetoram that can reduce the pesticide application frequency and be cost-effective and pocket-friendly for the farmers.

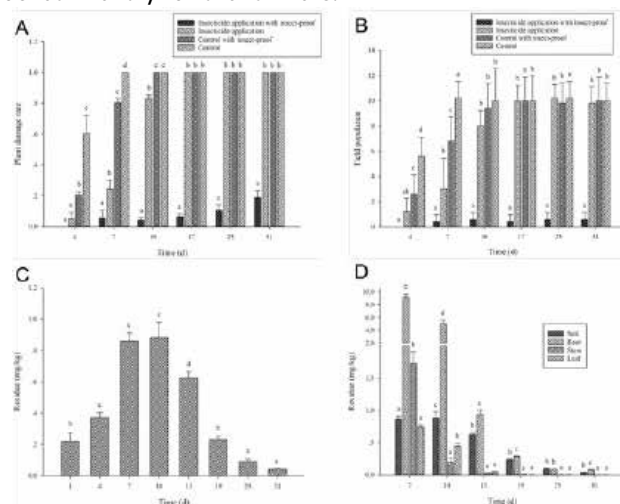


Fig. 1. The damage rate on the flowering cabbage plants under control environment (A) and field environment (B), the degradation dynamics of spinetoram in the soil (C) and in different parts of the flowering cabbage (D). Alphabets (a-d) indicate the statistical significance at $P \leq 0.05$.

P05IVS

DEVELOPMENT OF BIOCONTROL MEASURES OF COTTON VERTICILLIUM WILT BASED ON *BACILLUS* SPP. SPORES AND BROCCOLI RESIDUE

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Cotton verticillium wilt (CVW), caused by *Verticillium dahliae*, represents a typical soil-borne disease that leads to widespread economic losses in cotton production. In China, annual losses of approximately 250-310 million US dollars have been reported due to CVW. Therefore, it is imperative to develop novel control strategies to combat this devastating disease. A preparation of powder for dry seed treatment (DS) for controlling CVW was developed using *Bacillus*

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amyloliquefaciens PHODG36 as the active ingredient, and dry broccoli residues powder as the carrier. The seed dressing treatment with PHODG36 DS at a dosage of 10% seed weight was evaluated for the control efficacy on CVW through greenhouse and field plot tests. Field demonstrations were conducted in 7 different regions in Hebei and Shandong provinces. The optimal formula of the preparation - 10 billion CFU/g *B. amyloliquefaciens* PHODG36 DS was developed as follows: 20% PHODG36 technical concentrate (TK), 77% dry broccoli residues powder, 1% dispersant NNO, and 2% wetting agent SDBS. Meanwhile, this preparation had a light-yellow appearance and no agglomeration, with an average particle size of 37.65 μm , no caking or sticking after storage. The results from the evaluating tests showed that the treatment had a promoting effect on cotton plant height and biomass, and the control efficacy on CVW reached 73.03% in greenhouse tests, 61.60% in field plot tests, and ranged from 61.08% to 74.8% in field demonstrations, while the boll number per plant increased from 3.38% to 27.42%. Comprehensive analysis shows that 10 billion CFU/g PHODG36 DS can effectively reduce the incidence of CVW and significantly increase yield. It holds broad application prospects for health cotton production in the Yellow River Basin cotton region of China.

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PREPARATION OF BACILLUS AMYLOLIQUEFACIENS PHODG36 POWDER FOR DRY SEED TREATMENT AND ITS APPLICATION ON COTTON

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biomass, and the control efficacy on CVW reached 73.03% in greenhouse tests, 61.60% in field plot tests, and ranged from 61.08% to 74.8% in field demonstrations, while the boll number per plant increased from 3.38% to 27.42%. Comprehensive analysis shows that 10 billion CFU/g PHODG36 DS can effectively reduce the incidence of CVW and significantly increase yield. It holds broad application prospects for health cotton production in the Yellow River Basin cotton region of China.

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VERTICILLIUM WILT IN PEPPER: WATER RELATIONS AND EMBOLISM FORMATION

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Despite the fact that phloem is responsible for transporting sugars, which can be a valuable nutrient resource, *Verticillium dahliae* exclusively colonizes the xylem, the conduit responsible for long-distance transport of water and minerals. Because wilting is one of the main symptoms caused by xylem-invading pathogens, this study aims to elucidate the impact of *V. dahliae* VD1 infection on the hydraulic function of pepper. To this end, plant infection experiments were conducted in which sap flow and water potential, serving as an indicator of the plant's water status, were monitored with custom-built ExoBeat sap flow sensors and the Scholander pressure chamber, respectively, alongside quantitative PCR analysis to assess fungal colonization. At 14 days post inoculation (dpi), a transition in fungal colonization from roots to shoots was observed, together with a significant decrease in sap flow and water potential and the occurrence of wilting symptoms. Due to the significant impact of infection on the plant's water transport dynamics, the second objective of this study was to evaluate the contribution of vessel occlusions (e.g. tyloses, pectin-rich gels, and/or fungal structures) through microscopic analysis of stem cross-sections. In addition, presence of non-conducting air-filled conduits, called 'embolized conduits' was investigated with X-ray micro-computed tomography (μCT) and hydraulic conductance experiments. In conclusion, our data suggest a minor role for occluding factors such as tyloses, pectin-rich gels, and embolisms from 14 dpi onwards while physical obstruction by the pathogen itself seems to contribute most towards the compromised water transport. Further research will be conducted to investigate the potential involvement of embolism formation in the *V. dahliae*-pepper interaction, with a specific emphasis on plants inoculated at a later, more mature stage.

P08IVS

ASSESEMENT OF THE CYTOTOXIC ACTIVITY OF VDNEP, A VERTICILLIUM DAHLIAE ELICITOR, VIA A VIRUS-BASED HOST INDUCED GENE SILENCING APPROACH

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Verticillium dahliae is a widely distributed plant pathogen, which can cause severe damage in economically important crops. This fungus possesses an arsenal of protein effectors to manipulate and overcome plant immunity systems, and to promote pathogenicity. VdNEP, a necrosis and ethylene inducing protein in *V. dahliae* has been reported to induce cell death, production of reactive oxygen species, while activates plant's defense responses. The aim of this study was to investigate the cytotoxic activity and the subcellular localization of VdNEP in two *V. dahliae* hosts, *Arabidopsis thaliana* and cotton. The full-length cDNA of the VdNEP gene, fused with the green fluorescent protein encoding gene, was initially cloned into the binary vector SK1727. Subsequently, the construct was subcloned into the modified Tobacco Rattle Virus (TRV) plasmid vector and introduced into *Agrobacterium tumefaciens* AGL-1 cells. Agroinfiltration of cotton and *A. thaliana* leaves was carried out using a 1:1 ratio of *A. tumefaciens* cells harboring the TRV:RNA1 and TRV:RNA2:VdNEP-gfp plasmids. Seven days post infiltration, all agroinfiltrated cotton and *A. thaliana* plants developed the wilting phenotype, indicating that VdNEP, is associated with the severe wilting phenotype in addition to the induction of defense responses activation, confirming its double-acting role. UV light monitoring showed that TRV is translocated in the whole plant expressing the VdNEP-gfp fusion protein, while subcellular localization of the tagged protein using laser scanning confocal microscopy revealed that VdNEP is localized in the cell membrane, and specifically around dead cells of *A. thaliana*. To the best of our knowledge, this is the first report of the ectopic, yet transient expression of a fungal gfp-tagged gene in plants using a TRV-based HIGS technology in lack of the pathogen's presence. Elucidating the role of this gene could provide valuable insights for targeted gene silencing in fungi, facilitating the development of alternative control strategies.

P09IVS

RAPID BARE EYE DETECTION OF THE SOILBORNE PATHOGEN VERTICILLIUM DAHLIAE IN TOMATO PLANTS USING LOOP MEDIATED ISOTHERMAL AMPLIFICATION

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Verticillium dahliae is a vascular wilt pathogen causing serious economic losses in a wide range of hosts. Early detection of the pathogen is important for disease management. LAMP (Loop-Mediated Isothermal Amplification) is a rapid and simple isothermal DNA amplification technique employed for on-field detection of pathogens. In the present study a LAMP assay targeting the ITS (Internal Transcribed Spacer) region was developed for accurate detection of *V. dahliae* in plant

tissue and soil. Therefore, the LAMP primers were specifically designed to avoid amplification of the less virulent pathogen *Verticillium tricorpus*, which is prevalent in both soil and plant tissues, leading to mild symptoms. LAMP detection relied on a visible color change discernible to the naked eye, based on phenol red reacting to a decrease in pH. The detection limit of the LAMP assay was 5pg of *V. dahliae* DNA / μ l reaction (100 pg / reaction) while *V. tricorpus* DNA was not amplified at 50 pg of *V. tricorpus* DNA / μ l reaction (1 ng / reaction). To evaluate *V. dahliae* detection in crude samples, tomato plants of a sensitive and resistant variety were separately inoculated with *V. dahliae* and *V. tricorpus*. Results showed that the new LAMP assay detected *V. dahliae* in crude samples (homogenized in water) of tomato stems, only in the sensitive variety. No cross reaction with non-inoculated plants or plants inoculated with *V. tricorpus* was observed. Quantification of fungal DNA in plant tissue is underway to assess the detection limit of the developed LAMP assay in plant tissue. The LAMP assay developed in this study proves to be a highly valuable tool, significantly contributing to the early diagnosis of this destructive vascular wilt pathogen.

P10IVS

STUDYING THE TRANS-KINGDOM RNAi PHENOMENON IN VERTICILLIUM NONALFALFAE-HOP PATHOSYSTEM

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RNA interference (RNAi) is a conserved eukaryotic mechanism mediated by small RNA molecules (sRNA), crucial for regulating gene expression in fungi and other organisms. The exchange of sRNAs between organisms, known as trans-kingdom RNAi, has shown promise in innovative approaches to pest control and disease management. In recent years, it has become clear that the main pathway for exchanging signals in the trans-kingdom RNAi pathways is the transport of biological molecules captured in extracellular vesicles (EV). In this way, RNAi signals are transmitted from the pathogen to the host plant, where they modulate defence mechanisms of the host, contributing significantly to fungal pathogenicity. The study aims to elucidate RNAi interactions between *Verticillium nonalfalfae* and hop, with a specific emphasis on extracellular vesicles (EVs) as potential carriers of RNAi signals. The primary objective involves the isolation and characterization of fungal EVs from culture supernatants, employing ultracentrifugation, scanning electron microscopy, and nanoparticle tracking analysis (NTA). Next-generation RNA sequencing and bioinformatics analysis will be used to determine whether *Verticillium nonalfalfae* synthesizes and transports small RNAs enclosed in EVs. From fungal culture supernatants, cultivated for 6 days, EVs were successfully purified using differential centrifugation with final ultracentrifugation at 118 000 x g. The NTA analysis revealed that the isolated vesicles range in size from 150 to 300 nm. Using the SEM microscopy, the particles were visualized and characterized based on their round shape and size. The size and shape of the isolated EVs of *Verticillium nonalfalfae* is in accordance with the characteristics of fungal EVs. Next, the RNA will be isolated from the EVs and small and total RNAs sequenced to determine the extracellular transcriptome of the *Verticillium nonalfalfae*. This study provides insights into the role of EV-mediated RNAi in fungal pathogenicity.

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P11IVS

IN VITRO ACTIVITY OF ESSENTIAL OILS AGAINST VERTICILLIUM DAHLIAE

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The soilborne pathogen *Verticillium dahliae* poses a significant threat to various economically important crops and is challenging to manage. Recently, biological, and sustainable control approaches have shown considerable potential to effectively manage the disease. This study explored the antifungal activities of essential oils (EOs) from thyme, oregano, rosemary, and sage against *V. dahliae*. EOs were assessed for their capacity to inhibit mycelial growth and diminish the viability and germination of conidia and microsclerotia (MS) through *in vitro* tests. The activity of EOs was evaluated at 3%, 0.6% (1:5), and 0.3% (1:10) concentration. Inhibition of mycelial growth was observed after 5 and 7 days incubation by measuring the inhibition halo formed around the EOs in two perpendicular directions. Conidial germination was monitored at 4, 8, and 48 hours of incubation. The germination of microsclerotia (MS) was assessed at 4, 10, 24, and 72 hours post-treatment (hpt) with EOs. Ten µL of conidia and MS suspension were loaded in a Thoma's chamber, and germination was examined under a microscope at 40x magnification. Conidia and MS were deemed to have germinated if the germ tube was twice the diameter of conidia or MS. The results indicate that 3% concentrations of thyme and oregano EOs were highly effective (100%) in arresting the mycelial growth of *V. dahliae* after five and seven days of incubation. Similarly, thyme and oregano EOs at 3% concentrations were also successful in reducing the conidial germination of *V. dahliae* by 77.8% and 74.5%, respectively. At 4 hpt, a 3% concentration of rosemary and sage EOs reduced MS germination by 41.47% and 49.03%, respectively. However, at 24 hpt, thyme (1:10) and oregano (1:5) showed a decreasing trend in MS germination by 42.74% and 33.52%, respectively. Thyme and oregano EOs indeed demonstrated significant fungicidal and fungistatic activities. Subsequent studies on plants may validate these effects, and further research is necessary to gain a better understanding of their impact on microsclerotia. EOs are considered a good alternative to chemical products in terms of safety, environmental impact, and user-friendliness. High variability in EOs composition and the presence of different chemotypes may impose challenging problems in product formulation and efficiency. Nevertheless, plant EOs represent a potential natural reservoir of antifungal compounds, offering a valuable resource for the future development of novel fungicides and to improve food safety and develop sustainable strategies for protecting plants.

