

Abstracts

Abstracts of invited Keynote lectures, Keynote and Oral presentations and Poster papers presented at the 17th Congress of the Mediterranean Phytopathological Union, July 6–10, 2025, in Bari, Italy

This Issue of *Phytopathologia Mediterranea* firstly outlines a summary of the Congress, and then publishes abstracts of all the invited Keynote papers, and offered Oral and Poster papers, that were presented at the Congress.

Preface

The 17th International Congress of the Mediterranean Phytopathological Union (MPU) was headlined as addressing “New Phytopathology Frontiers of Research and Education for Plant Health and Food Safety”. The Congress was organized by the Mediterranean Phytopathological Union (MPU), and the Mediterranean intergovernmental organization CIHEAM Bari, International Centre for Advanced Mediterranean Agro-economic Studies.

The Congress Plenary Session was opened with welcoming remarks by Dr Biagio Di Terlizzi, Director of CIHEAM Bari; Prof. Dimitri Tsitsigiannis, President of MPU; and Dr Ugo Della Marta, Director General of the Directorate for Food Hygiene, Safety and Nutrition (DGISAN) of the Italian Ministry of Health.

The Congress was attended by more than 350 participants from 38 countries: 19 countries in the Mediterranean region (Albania, Algeria, Croatia, Egypt, France, Greece, Israel, Italy, Jordan, Lebanon, Montenegro, Morocco, Palestine, Portugal, Slovenia, Spain, Syria, Tunisia, Turkey), eight elsewhere in Europe (Belgium, Germany, Hungary, Latvia, Lettonia, North Macedonia, Serbia, The Netherlands), and 11 other countries (Australia, Chile, Congo, Georgia, New Zealand, Pakistan, Reunion Island, South Africa, United Arab Emirates, United Kingdom, United States of America).

The congress was aimed to promote knowledge exchange and dialogue between science, institutions, industry and civil society, reinforcing a vibrant, connected, and forward-looking scientific community. Among these, beside researchers of the 15 National

Plant Pathology or Plant Protection societies that are MPU members, people also attending included representatives from: IPPC-FAO, FAO-RNE, EUPHRESKO, Ministry of Agriculture, Food Sovereignty and Forests (MASAF), Directorate General for Food Hygiene, Food Safety and Nutrition (DGISAN), EPPO, EFSA, the National Research Council (CNR), the Apulia Region, the European Commission, and numerous universities and research centers.

Papers presented at the Congress included: 14 Keynote lectures, 36 Session Keynote papers, and research papers as 88 oral and 180 poster presentations, in a total of 18 Congress sessions. Beyond its scientific scope, the Congress was a platform to strengthen existing partnerships and forge new collaborations across borders and research disciplines. This included links with the International Society for Plant Pathology, the International Society for Mycotoxicology, EUPHRESKO, and CIHEAM Bari, supporting advancements in plant health sciences as a pillar of the One Health approach recognizing the links between human, animal, plant, and environmental health.

CIHEAM Bari took active steps to make the Congress accessible to young researchers, especially from non-European regions. This included reduced registration and accommodation fees for students (including PhD candidates) and early career scientists.

An all-day field trip on 10 July included observations of ancient olive trees infected by *Xylella fastidiosa* and the characteristic sites of Alberobello, providing a unique opportunity to connect field and territory observations with the latest research and disease mitigation strategies.

To recognize scientific excellence of the early career researchers, MPU presented awards for the oral presentations by Marah Abukhmaish (Palestine), Serafina Amoia (Italy) and Marco Crudele (Italy), and the poster papers of Martini Florian (Belgium), Luiza Sánchez-Pereira (Spain), and Aleksandra Susnjar (Republic of Serbia). The Congress also hosted the award ceremony for the 4th edition of the video competition *Plant Health TV: Promoting the Importance of Plant Health Research*. Organized by EUPHRESKO III, CIHEAM Bari, the Plant Biosecurity Research Initiative (PBRI), IPPC, and CABI, this competition highlights global efforts in plant health research. The award recipient was Spyridoula Dimitropoulou, for the video “Saving Greece’s plane trees: digital technologies for early detection of *Ceratomyces platani*”. The award included a 2-week internship in Australia, sponsored by the PBRI.

In its concluding session, the congress identified key frontiers and pressing challenges in plant pathology that must be addressed, including:

- The integration of Plant Health within the One Health framework.
- The role of climate change in driving the emergence and spread of new pests and diseases.
- The urgent need for predictive tools and early warning systems.
- The integration of digital technologies, genomics, and field ecology into plant health research and practice.
- The necessity of addressing biosecurity vulnerabilities and regulatory gaps.
- The imperative of safeguarding global food security and biosecurity in increasingly complex and interconnected supply chains.

To respond effectively to these developments and challenges, interdisciplinary and collaborative educational models across all professional levels are essential. Such models are crucial for training a new generation of plant pathologists equipped with the necessary skills, technological expertise, and cross-sectoral understanding.

Furthermore, strong support for early career researchers is vital to ensure continuity, innovation, and the advancement of knowledge in the field.

In this context, the Mediterranean Phytopathological Union (MPU), in collaboration with national, regional, and international networks, aims to significantly strengthen interdisciplinary and cross-border cooperation in the field of plant health, while fostering meaningful dialogue among the scientific community, regulatory bodies, and society at large.

Photographs from the Congress are available at: <https://ciheambaricongressmpu2025.org/photo-gallery/>

Editorial disclaimer

The Co-Editors-in-Chief of *Phytopathologia Mediterranea* present these Congress Abstracts with the proviso that all abstracts are published as submitted by the respective paper authors. The abstracts have not been refereed using the normal standard peer review processes applied for research papers in this journal.



Opening Plenary Session

KEYNOTE LECTURES

The past, present, and future for the Mediterranean Phytopathological Union and *Phytopathologia Mediterranea*. L. MUGNAI¹, A. M. D'ONGHIA², R. E. FALLOON³, G. SURICO¹. ¹*Department of Scienze e Tecnologie Agrarie, Alimentari, Ambientali e Forestali, University of Florence, P.le delle Cascine 18, Florence, Italy.* ²*CIHEAM-Bari, Via Ceglie 9, Valenzano, Italy.* ³*154 Hackthorne Road, Christchurch 8022, New Zealand.* E-MAIL: laura.mugnai@unifi.it

This Congress marks the 65th anniversary of foundation of *Phytopathologia Mediterranea*, by the eminent Italian plant pathologists Antonio Ciccarone and Gabriele Goidanich, from Italy. With colleagues from Israel, France and Portugal, these pioneers aimed to create a medium for publishing high quality research papers, enhancing cooperation in meeting the increasing plant health challenges in the ecophysiological areas surrounding the Mediterranean Sea. Two years later, these people also decided to enrich the project by providing opportunity for researchers to meet and collaborate, by organizing congresses, workshops, and joint research planning. These objectives were achieved by establishing the Mediterranean Phytopathological Union (MPU), which became the owner and editor of *Phytopathologia Mediterranea*. Since then, 16 Congresses have been successfully held, and the 17th being held in July 2025 with the theme “New Phytopathology Frontiers of Research and Education for Plant Health and Food Safety.” This Congress aims to highlight education and research as essential tools to address emerging challenges in phytopathology and food safety, while raising public awareness and contributing to the achievement of the Sustainable Development Goals (SDGs). In 2024, the MPU General Assembly restructured the Union by extending membership to researchers who are members of any national society of phytopathology or related fields within the Mediterranean region, provided that these societies have formal agreements with MPU. Now the Union includes 15 National societies from Croatia, Egypt, Greece, Israel, Italy, Palestine, Bosnia and Herzegovina, Serbia, Slovenia, Portugal, Spain, and Türkiye. This reform broadened MPU's vision to include individual researchers from non-Mediterranean countries, as well as international societies and organizations involved in plant health across Mediterranean ecological zones. The revised Statute establishes an Advisory Committee to represent this expanded research community and

actively promotes the global dissemination and coordination of plant health research. It fosters collaboration with other networks, such as International Society of Plant Pathology, Arab Society of Plant Protection, International Society of Mycotoxicology, EUPHRESKO, while encouraging active involvement in European initiatives like EUPHRESKO III. *Phytopathologia Mediterranea* remains a key part of this initiative, with all volumes now available as open access and online, and with an Impact Factor in the first quartiles of relevant subject areas. The Journal is overseen by two Editors-in-Chief, from Italy and New Zealand, and by researchers from 14 different countries. The journal gets value from having a non-profit online editor and publisher, Firenze University Press. Looking ahead, *Phytopathologia Mediterranea* is strengthening its role by expanding international visibility, increasing reviewer engagement, and encouraging submissions on emerging plant diseases and food security, while maintaining non-profit open access status. The journal maintains a rigorous and transparent editorial processes, with artisanal care in manuscript selection and quality control. The Mediterranean Phytopathological Union and *Phytopathologia Mediterranea* have enabled two generations of plant pathologists to communicate research outcomes. The future of the MPU and its journal will be supported by a continuing commitment to knowledge sharing and research collaboration, and to maintaining the objectives and foresight of the people who established the Union and journal.

Plant Pathology is important in today's world. G. SURICO. *Department of Scienze e Tecnologie Agrarie, Alimentari, Ambientali e Forestali, University of Florence, P.le delle Cascine 18, Florence, Italy.* E-MAIL: giuseppe.surico@unifi.it

In December 2021, the American Phytopathological Society (APS) conducted a survey to identify and rank key discoveries in plant pathology that have had broad impacts on science and practical disease management during the past half century. Based on the responses received, the working group has chosen about fifteen of the most important discoveries of all: they ranged from the discovery of the Ti plasmid up to the Type III secretion system; from the deployment and management of host resistance genes up to the utilization of biological controls and suppressive soils. However, despite the acquisition of this and much other knowledge, agricultural production losses due to diseases remain, today, very high. Indeed, losses have increased compared to 50 years ago. Despite this scenario, world public opinion is

pushing for a substantial reduction in the use of pesticides in agriculture. Will we be able to defend plants from diseases with means other than those predominantly used today? Will we be able to practice our profession as Frederick L. Wellman defined it more than 50 years ago?: “*All the academic duties, reading, teaching and research, in a plant pathologist’s life have one central purpose, and that is to control plant diseases*”. Recently a group of our colleagues commissioned by the APS identified 17 major themes among the most important ones to investigate. One of them was the following: “*Early detection, identification, diagnosis, surveillance, and forecasting of emerging, and reemerging plant diseases and economically important plant diseases, including development toolkits for rapid, sensitive, specific, dependable, portable, economic, and accurate diagnostics, development and application of new technologies, and systems for global surveillance of plant diseases*”. Twelve years have passed since the outbreak of the *Xylella* epidemic on olive trees in Italy. The disease probably appeared in Southern Italy in 2008 and only 5 years later the bacterium (*Xylella fastidiosa* subsp. *pauca*) was isolated from infected olive tissues. By that year the disease had already spread over an area of about 8,000 hectares. The event had a huge impact on the population. Considering the particular importance of the affected plant and of the drastic measures taken to try and block the disease, everyone felt obliged to have their say and the scientific element has become almost a secondary object in a scenario that immediately appeared very complex. To date, despite the containment measures adopted, the disease has killed hundreds of thousands of plants, has spread for tens of kilometers beyond the original infection focus, and the presence of two other subspecies, *fastidiosa* and *multiplex*, has been recently diagnosed. Two other topics that need to be remembered are artificial intelligence and combining the human, animal and plant medicine into one. Regarding the first point, it is superfluous to note that artificial intelligence is also coming to agriculture, the slowest sector in welcoming the innovations of the intellect, and it is doing so in an increasingly pervasive way so much so that everything or almost everything could change in a few years for example in the fields of diagnostics, prediction systems, and disease management strategies. The MPU could help to answer some or all of the questions raised earlier, during this Congress or even later, by setting up special study commissions. We just have to want it.

Plant Health research coordination: an international endeavor. B. GIOVANI¹, L. MUGNAI², J. ZORRIL-

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Plant health is a key factor in any strategy to achieve food security, protect the environment and biodiversity and facilitate safe trade and is an essential pillar of the bioeconomy. 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 need to revive the scientific basis of the phytosanitary field has been a priority since the EPPO Madeira Declaration. Upon a request of the EC Council Working Party of the Chief Officers of Plant Health Services (COPHS), 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. In 2019, during the Meeting of the Agricultural Chief Scientists of the G20, the importance ‘to strengthen research collaboration to develop effective measures against transboundary pests and to participate in joint funding networks such as Euphresco’ was noted. In 2020, the article ‘Science diplomacy for plant health’, co-authored by experts from the IPPC, several Regional Plant Protection Organizations (RPPOs), NPPOs, research coordination networks and research organizations, highlighted the need for ‘*a global network for phytosanitary research coordination that can shape research agendas across countries and accelerate the development of science to support regulatory phytosanitary decision makers*’. In 2021, the IPPC Strategic Framework 2020-2030 was adopted at the 15th session of the Commission on Phytosanitary Measures (CPM-15). Global phytosanitary research coordination is one of the development programmes identified, and it is expected that by 2030 ‘*possibilities for establishing an international research col-*

laborative structure have been explored and, if appropriate, the structure has been established. 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. Activities dedicated to the Mediterranean region will be presented and explained.

Plant Health for all: strengthening global phytosanitary capacity through the IPPC Plant Health campus. S. BRUNEL. *IPPC Officer in Charge for day-to-day matters, Implementation and Facilitation Unit Lead.* E-MAIL: sarah.brunel@fao.org

Plant health is the first line of defense against threats to food security, biodiversity, and safe trade. Yet many countries face critical capacity gaps in their ability to manage pests, prevent incursions, and meet international standards. The IPPC Plant Health Campus was created to change that. The IPPC Plant Health Campus is a transformative global learning platform developed by the International Plant Protection Convention (IPPC) in collaboration with the FAO eLearning Academy and funded by the European Union. It is the first initiative of its kind to offer free, structured, and standardized training in phytosanitary practices, accessible both online and offline to users around the world—including those in remote or under-resourced areas. The platform offers multilingual certified e-learning courses, interactive tools, downloadable IPPC guides, and practical training materials designed to strengthen technical and operational capacities across the global plant health community. As public good, the Campus is democratizing access to critical knowledge, reducing disparities, and fostering equitable development. A distinctive feature of the Campus is its tailored learning pathways, built around real-world roles within National Plant Protection Organizations (NPPOs) and other regulatory or technical agencies. These pathways guide learners through structured modules on priority topics including NPPO management, pest risk analysis, market access, import and export procedures, inspection and surveillance, stakeholder engagement, emergency response, wood packaging materials, and resource mobilization. To date, IPPC Campus courses have been completed over 7,000 times by users in more than 100 countries, demonstrating strong global uptake. Learners receive digital badges or certificates upon completion, validating their new competencies and supporting their professional development. These credentials contribute to building a more skilled, recognized, and mobile plant health workforce. Courses

are currently offered in English and French, with selected modules available in Spanish. The IPPC welcomes in-kind and financial contributions to support translation into additional UN languages, expanding accessibility for a truly global audience. Beyond the public sector, the Campus is increasingly seen as a tool for the next generation of plant health professionals. Several universities are exploring the integration of IPPC-certified courses into their agricultural and phytosanitary curricula, embedding international standards and best practices into formal education systems. To amplify its reach, the IPPC has mobilized regional champions who actively promote the Campus in national and international fora, including conferences and media interviews. Their efforts are part of a growing global campaign to raise awareness and drive engagement with the platform. By making expert-developed, standardized training universally available, the IPPC Plant Health Campus strengthens the implementation of International Standards for Phytosanitary Measures (ISPMs), promotes safe international trade, and builds more resilient, knowledge-driven phytosanitary systems. Ultimately, the Campus plays a vital role in advancing the Sustainable Development Goals, particularly in areas such as food security, environmental protection, and inclusive economic development. The IPPC Plant Health Campus is not just a training tool—it is a global investment in a safer, healthier, and more sustainable future.

CIHEAM Bari Educational Programmes to strengthen plant health research in the Mediterranean Region. A.M. D'ONGHIA. *International Centre for Advanced Mediterranean Agronomic Studies, Via Ceglie, 9-70010 Valenzano, Italy.* E-MAIL: donghia@iamb.it

Plant pests pose a major threat to food security and rural livelihoods across the Mediterranean, where agriculture remains a cornerstone of socioeconomic stability. The United Nations designated 2020 as the International Year of Plant Health and 12 May as the International Day of Plant Health, underscoring the vital role of plant protection in global sustainability efforts. CIHEAM Bari has been at the forefront of plant health research and capacity building for over 35 years, fostering scientific innovation, training, and international cooperation across the Mediterranean and neighbouring regions. Its research activities mainly focus on advanced diagnostic methodologies and epidemiological studies of high-impact pests. The institute has also pioneered the integration of smart technologies into pest risk analysis, early warning systems, and predictive modelling—sup-

porting the implementation of precision surveillance and targeted intervention strategies. CIHEAM Bari has made substantial investments in higher education and research, significantly contributing to the development of a regional scientific community in plant health. Since 1985 about 4000 former students were trained in Plant Health through Master, Master of Science, Advanced Specialized Courses & PhD programs—mainly supported by scholarships from the Italian Cooperation—with the aim to prepare young generation towards professional, research and academic careers in plant health. MSc and PhD students conduct original research on regionally relevant phytosanitary challenges, gaining strong technical expertise as well as critical thinking and problem-solving skills. Since 2000, around 60 of 400 MSc graduates have completed PhDs in collaboration with Italian and international universities: 42% from Maghreb, 39% from the Near and Middle East, 7% from the Balkans, and 12% from EU. This sustained academic effort has led to about 300 scientific publications in peer-reviewed journals and conference proceedings. Research thematic areas were mainly in plant pathology, followed by entomology and post-harvest, while research topics were focusing on characterization, diagnosis, surveillance and control. CIHEAM Bari also plays a strategic role in aligning phytosanitary regulations with EU standards, contributing to food safety, facilitating international trade, and promoting agricultural sustainability. With support from international cooperation, modern research facilities and equipment have been provided to enable young scientists to carry out studies in their home countries, helping to limit the so-called “brain drain”. A notable example is the PHYTO BiH project, which supports Bosnia and Herzegovina in aligning its regulatory framework with EU legislation. Similar initiatives in Near Eastern and African countries aim to enhance national systems for managing phytosanitary risks and emergency responses. To advance plant health research in the Mediterranean and beyond, strengthening both national capacities and regional collaboration, is essential. In 2021, CIHEAM Bari and Euphresco launched the first coordinated initiative to establish a shared research agenda: *The Plant Health Research Priorities for the Mediterranean Region*. This laid the foundation for the ongoing EUPHRESCO III project, fostering long-term partnerships and aligning priorities on a global scale. Through its integrated approach—combining research, education, policy support, and international collaboration—CIHEAM Bari continues to serve as a regional leader in building a resilient, cooperative, and forward-looking phytosanitary system for the Mediterranean and beyond.

Transboundary plant pathogens threatening crops and natural landscapes in the Mediterranean region.

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The Mediterranean region, recognized for its rich agricultural heritage and ecological diversity, is becoming increasingly vulnerable to the emergence and spread of transboundary plant pathogens. These pathogens, including fungi, bacteria, and viruses, can cross national borders and rapidly establish in new environments. They pose serious threats to crop productivity, food security, rural livelihoods, ecosystem integrity, and the economic stability of the region. The accelerated pace of globalization, combined with climate change and the intensification of agricultural trade, has significantly increased the transboundary movement and establishment of plant pathogens, thereby overwhelming conventional national phytosanitary systems. The international exchange of planting materials remains a key pathway for the unintentional introduction and spread of invasive and emerging plant diseases. In the Mediterranean context, climate variability, particularly rising temperatures and altered precipitation patterns, is expanding the geographical distribution, seasonality, and persistence of numerous pathogens. This trend is heightening phytosanitary risks across borders. Despite notable initiatives and efforts undertaken by individual countries, responses to transboundary plant pathogens remain constrained by limited technical capacities, fragmented surveillance systems, inadequate quarantine infrastructure, and insufficient risk perception. Many countries encounter difficulties in timely detection, accurate diagnosis, and effective containment of outbreaks. The lack of harmonized phytosanitary protocols and regional data-sharing mechanisms further impedes coordinated responses. Notably, the complexity and cross-border nature of many plant diseases make it difficult for any single country to manage them effectively in isolation. Recent incursions of high-impact pathogens such as *Xylella fastidiosa*, *Fusarium oxysporum* f. sp. *cubense*, and *Candidatus Liberibacter asiaticus* (citrus greening) illustrate the urgent need for collective action, as these diseases threaten critical crops like olives, bananas, and citrus. Addressing the transboundary nature of these threats demands a science-driven, collaborative approach that extends beyond national boundaries. Priority actions include: (i) strengthening early warning and diagnostic systems through advanced molecular tools and remote sensing technologies; (ii) promoting integrated disease manage-

ment practices tailored for Mediterranean agro-ecosystems; (iii) enhancing regional collaboration through joint surveillance platforms and coordinated emergency response frameworks; and (iv) investing in interdisciplinary education, capacity building and training to build resilient and responsive plant health systems. Integrated pest and disease management (IPM), plant breeding for resistance, and nature-based solutions are key to reducing pathogen spread and impact while ensuring environmental sustainability. Regional mechanisms must also support applied research, comprehensive risk assessment, and real-time data exchange to expedite decision-making. This presentation will provide an overview of the current status and emerging trends in transboundary plant pathogens affecting the Mediterranean, drawing on recent case studies and field-based experiences. It will emphasize the importance of regional cooperation, sustainable management practices, and science-based plant health practices. In summary, transboundary plant pathogens represent a complex and evolving threat to agricultural productivity, biodiversity, and natural landscapes across the Mediterranean region. Addressing this challenge aligns with FAO's mandate to promote sustainable plant health systems and requires the integration of cutting-edge research, evidence-based policy, and sustained regional cooperation. Building a resilient Mediterranean plant health system calls for a shared vision, strong political will, sustained investment in innovation, and collaboration.

General overview of the EU policies for Plant health with a focus on *Xylella fastidiosa*. M.B. MARQUEZ GARCIA. *European Commission – DG SANTE, Unit G1 – Plant Health*. E-MAIL: maria.marquez-garcia1@ec.europa.eu

In the EU, Regulation (EU) 2016/2031, known as the Plant Health Regulation, establishes the framework for protecting plant health in the European Union, aiming to prevent the introduction and spread of plant pests and diseases that can have significant impact on agriculture, natural ecosystems and biodiversity. This Regulation defines the different categories of regulated plant pests in the EU, enhances prevention and early detection of Union quarantine pests by establishing the obligation for Member States to create multiannual survey programmes spanning 5-10 years, while requiring annual surveys for those pests whose potential economic, environmental or social impact is the most severe, the priority pests. Moreover, it requires Member States to implement measures to eradicate Union quarantine pests, including the

creation of demarcated areas, composed by infected zone and buffer zone, to prevent the spread of the pest to the rest of the Union territory. *Xylella fastidiosa* is listed as Union quarantine pest in Part B of Annex II of Commission Implementing Regulation (EU) 2019/2072, as a pest known to be present in the EU territory. Due to its economic, environmental and social impact, this pest is also listed as a priority pest in Commission Delegated Regulation (EU) 2019/1702. Since the first finding of *Xylella fastidiosa* in the EU in 2013, the Commission has put in place specific measures to control this pest and has updated them following the scientific developments. Prevention and early detection are key pillars in the fight against this pest. Today, the measures to prevent the introduction into and the spread within the EU are detailed in Commission Implementing Regulation (EU) 2020/1201. The regulation, amended in September 2024, includes the obligation for all Member States to carry out annual surveys and have prepared contingency plans and for those Member States where the pest is present, the regulation details the eradication, and when eradication is no longer possible, the containment measures that have to be taken to prevent the further spread of the pest. Moreover, it also details the rules for the movement of plants known to be host of *Xylella fastidiosa* within the Union and the requirements for the introduction of these plants from third countries. Based on the annual survey campaigns in all Member States, *Xylella fastidiosa* is known as absent in 23 Member States and present in 4 Member States where the affected areas have been demarcated for the control of the pest and they are currently under eradication or containment approach.

Prevention, preparedness and response to plant disease outbreaks in the Mediterranean Basin. A. VICENT. *Centro de Protección Vegetal y Biotecnología, Instituto Valenciano de Investigaciones Agrarias (IVIA), 46113 Moncada, Spain*. E-MAIL: vicent_antciv@gva.es

Plant health should be a priority in the Mediterranean Basin, particularly for the citrus sector, which now faces major threats from pests and diseases. The EU has implemented a risk-based regulatory framework through Regulation (EU) 2016/2031 and Implementing Regulation (EU) 2019/2072. These regulations establish specific requirements for the import, movement of plants, and surveillance of the territory, aiming to prevent the introduction and spread of harmful organisms. Risk analysis plays a central role in this preventive strategy, allowing authorities to evaluate the likelihood and impact of pest and disease incursions and to define appropriate

phytosanitary measures. This framework is supported by risk-based surveillance, epidemiological modeling, contingency planning and social engagement, forming a comprehensive strategy for preparedness and response. Two major threats to citrus crops—*Phyllosticta citricarpa*, the fungus causing citrus black spot (CBS), and ‘*Candidatus Liberibacter spp.*’, associated with huanglongbing (HLB)—are used here as case studies to illustrate this approach. These pathogens are classified as priority ‘pests’ under Implementing Regulation (EU) 2019/1702 due to their potential socioeconomic and environmental impacts, which mandates, reinforced surveillance and contingency plans, among other measures. Risk assessment studies on *P. citricarpa*, have underpinned EU regulations, despite opposition from other studies claiming that Mediterranean climates are unsuitable for CBS development. However, subsequent research and the recent outbreak in Tunisia, have confirmed that CBS epidemics can occur under Mediterranean conditions. Surveillance is another key pillar of the strategy. EU Member States are required to conduct surveys aligned with the phytosanitary status of each region. Tools like the Pest Survey Toolkit developed by EFSA provide methodological support for risk-based and statistically sound survey planning. Epidemiological models enhance outbreak preparedness by simulating disease spread and evaluating control strategies. For HLB, models incorporating spatial variability, vector dynamics, climate conditions, and management actions have shown that, without intervention, the disease could spread rapidly, becoming unmanageable. However, early detection and aggressive vector control could slow its progression, though such strategies may rely on insecticides not currently authorized in the EU. Contingency plans are critical for effective responses to pest and disease introductions. These plans outline procedures, responsibilities, and resources needed to contain or eradicate pests and diseases. For CBS and HLB, the Spanish Ministry of Agriculture has developed dedicated contingency plans. Simulation exercises, such as one held in Valencia in 2021 under the Pre-HLB project, tested these plans with positive feedback from stakeholders and provided opportunities for improvement. Finally, social acceptance plays a decisive role in the success of outbreak management plans. Resistance from producers due to lack of information can undermine control efforts. A survey among citrus sector actors in Spain revealed good awareness of HLB but limited knowledge of the contingency plan. The study highlighted the need to involve stakeholders early in the planning process to ensure better compliance and implementation. In summary, the EU’s integrated, risk-based approach—combining legal regulations, early

detection, epidemiological modeling, contingency planning and stakeholder engagement—is essential for protecting citrus crops from phytosanitary threats. Achieving this goal requires strong coordination among Mediterranean countries.

Plant quarantine challenges under climate change anxiety. S.M. ABDEL-MOMEN. *Former Minister of Agriculture and Land Reclamation of Egypt; MPU Delegate of the Egyptian, Phytopathological Society; Agricultural Research Center, Giza, Egypt.* E-MAIL: salah1993@yahoo.com

Several studies indicated the adverse effects of the climatic changes on most, if not all, elements of the agricultural production. Arable lands, water resources and microorganisms are also subjected to be adversely affected due to their exposure to the changeable weather factors. Plant quarantine as the safe guard of the plant wealth and as the regulator of the exportation and importation of the agricultural commodities is expected to be highly affected and challenged by the climatic Changes. The expected increase in the bio-diversity of the harmful and useful microorganisms will be a challenge facing the regulations of plant quarantine and consequently the trade and movement of commodities among countries. Some secondary pests could be altered to be major ones and new races of a pest could emerge causing new problems that need to be managed in certain methods and new regulations. From a first-hand experience and as being an editor of a scientific journal, several first reports and observations of pests on new hosts. This could be due to the change in virulence of the pathogen or the host due to the exposure to the stress of the climatic change. Also, the quantity and quality of several crops were affected in some seasons and this was with a correlation to the climatic changes. Also, geographic regions were effective in such new observations confirming the role of the climatic changes. The aforementioned challenges and observations are a burden on scientists working in the agricultural field. To face this burden a multidisciplinary team work should cooperate together through integrated and comprehensive work plan. The plan should include elements that mitigate the effects of the climatic changes on all the inputs and outputs of the sustainable agriculture process. Modern approaches for pest detection and identification should be applied. Also, pest dispersal monitoring regional network are needed to put the strategy of facing the current and expected challenges. Moreover, information exchange systems including developing

stress-tolerant crop varieties, reporting new diseases, pest races and bio-types in addition to climate changes information among the Mediterranean countries need to be established. Based on the obtained data, a resilient and accurate strategy will be developed to face the threat of the climatic changes in each country due to its local conditions and in cooperation with whatever progress occurring in neighboring Mediterranean countries. Also, the obtained data will be considered to make the suitable regulations of plant quarantine according to conditions of each country aiming at enhancing exportations and importations of the agricultural commodities among countries. In parallel with, frequent scurvies and monitoring are needed to revise and regulate the plant quarantine policies and regulations.

Fungi and mycotoxins crucial role in food and feed safety. P. BATTILANI. *Department of Sustainable Crop Production, Università Cattolica del Sacro Cuore, Via Emilia Parmense, 84, 29122 Piacenza, Italy.* E-MAIL: paola.battilani@unicatt.it

Mycotoxin contamination is considered the main chronic health risk coming from food and a quite similar impact can be described for feed. Even if three main fungal genera are reported as including mycotoxin producing species, *Fusarium*, *Aspergillus* and *Penicillium*, at least two more should be added because their metabolites are listed among mycotoxins and regulated or candidates for regulation, *Claviceps* and *Alternaria*. This results in a wide range of ecological and host crops requirements, with room for their action worldwide, in many different crops. Most mycotoxin-producing fungi are weak parasites and their main impact on agricultural products is not due, or only partially due, to yield reduction, but rather to cryptic health risks for consumer, due to symptomless contamination, so that grain and fruit are apparently healthy, but containing toxic metabolites. Maize is one of the best crops to consider when discussing mycotoxin/when it comes to mycotoxins. It has a wide use for animal feeding, but also has several food or food industry destinations. It is a perfect host for several fungal species, belonging to at least 3 of the main fungal genera including mycotoxin producers. Therefore, depending on the growing area, at least one fungus can grow and cause mycotoxin contamination. *Aspergillus flavus* merits to be mentioned, being the main producer of aflatoxin B1, the most toxic naturally occurring compound, carcinogenic, teratogenic and immunosuppressor, just to mention some known effects. Also nuts and cotton are among the host crops. Recently, aflatoxin con-

tamination was also reported in fresh fruits, also used as ingredients for baby food, rising further concerns and challenges for the crop chain management. Dairy animals fed with mycotoxin contaminated products, like maize or derived products, excrete in milk a modified form of the toxin, aflatoxin M1, almost equally toxic, contributing to human exposure through food. Tropical and subtropical areas climate optimise *A. flavus* activity, and stress conditions, like heat and water stress, are conducive for aflatoxin production, suggesting climate change as favourable for the increase of contamination, both in terms of the amount quantified in each product and the widening of suitable geographic areas and crops for *A. flavus* occurrence. Maize is not only consumed as grain and related processed product, but also as silage and similar fermented outcomes, giving room to other less studied fungi and metabolites, sometimes with fatal effect on animals. Therefore, it can be surely affirmed that mycotoxin producing fungi play a crucial role in food and feed safety, emphasised by climate change. All these different aspects make it clear that mycotoxin producing fungi challenge food safety and food security with different relevance depending on the geographic area. Consequently, it is more and more urgent to increase the attention towards this hidden challenge to increase resilience and adaptation and optimising mitigation of climate change effects.

Low-risk active substances and plant protection products: a possible opportunity. P. CAVALLARO. *Ministero della Salute, Rome, Italy.* E-MAIL: p.cavallaro@sanita.it

One of the main ways of protecting plants and plant products against harmful organisms (insects, mites, fungi – including those producing mycotoxins toxic to humans - weeds) as well as improving agricultural production, is the use of plant protection products, which are one of the two main categories of “pesticides” (the other being biocides). These products can be based on chemical active substances or microorganisms (bacteria and viruses) that perform the aforementioned protective/healing action against the harmful organisms. Since, in addition to the benefits for agricultural production, they can cause potential damage to human health and the environment, these products are subject to a strict authorization procedure that requires a careful assessment of the risks for humans, animals and the environment. This authorization procedure involves first, at EU level, the active substances that are subjected to a rigorous approval procedure coordinated by EFSA, which leads to their inclusion in a positive list and

afterwards, at National level, the plant protection products that contain such substances. Only plant protection products containing approved substances may be authorised by Member States. The EU evaluation process of active substances, which began in 1993 with Directive 91/414/EEC, has led to the decommissioning of a large part of the active substances previously approved in Member States, many of which have profiles of high acute and/or chronic toxicity for humans and the environment. Regulation (EC) 1107/2009, in order to allow the availability of products useful for supporting agricultural production, introduced the concepts of “low-risk” active substances and biocontrol agents (or biopesticides), and introduced measures to facilitate the approval of said active substances, often based on microorganisms or substances of natural origin, and the authorization of the related plant protection products by the Member States. Given that biopesticides are not currently a defined group at the level of substances and products in EU legislation, these are often so-called “low risk”, although there is no automatic rule of attribution and they must always meet the approval criteria. To be considered low-risk, substances must always meet the criteria defined in Regulation (EU) 1432/2017, which amends Annex II, point 5 of Regulation (EC) 1107/2009. Biopesticides are generally intrinsically less harmful/toxic than usual substances and cause less environmental load or pollution, their control nature is preventive, they work through different modes of action compared to chemical products and can be used up to harvest since they usually do not have residue problems. However, adequate incentives must be developed so that farmers gain confidence and familiarity with the use of these substances. Of the 1,470 substances present in the EU database, only 80 are low-risk substances, most of which are microorganisms while almost 1,000 are substances that have been withdrawn from the market.

Smart digital and artificial intelligence technologies for plant disease detection, territory surveillance and control measures. D. TSITSIGIANNIS. *Department of Crop Science, Iera Odos 75, Agricultural University of Athens, Athens, 11855, Greece.* E-MAIL: dimtsi@aua.gr

The escalating global demand for safe and sustainable food has necessitated the development of smart, digital, and integrated plant protection strategies. These strategies aim to enhance the resilience of agricultural systems through early detection, precise intervention, and data-driven decision-making. Our research focuses on advancing integrated pest management (IPM) by lev-

eraging smart digital technologies, Internet of Things (IoT) sensors, artificial intelligence (AI), and innovative plant protection products to address plant disease management in key crops. To accelerate the prognosis of pest outbreaks, early warning systems are being developed utilizing IoT sensors and predictive models. These systems monitor environmental parameters like humidity and temperature as well as plant disease epidemiological parameters, providing real-time data that, when integrated with forecasting models, can predict disease outbreaks. Such predictive capabilities enable farmers to implement preventive measures proactively, reducing the reliance on reactive treatments. Modern diagnostic tools employ AI, particularly convolutional neural networks (CNNs), to analyze images of plant tissues captured via smartphones, drones, or proximal sensing cameras. These tools can facilitate early identification of plant diseases, enabling timely interventions. Additionally, spectral and hyperspectral imaging technologies, deployed via unmanned aerial vehicles (UAVs)/drones or satellites like European Space Agency's (ESA) Sentinel-2, detect disease symptoms not visible to the human eye, enhancing surveillance capabilities over large agricultural areas. Geographic Information Systems (GIS) further support spatial analysis, mapping disease outbreaks and identifying high-risk zones for targeted management. Our research also explores the discovery and evaluation of novel plant protection products (PPPs) such as biocontrol agents, including beneficial fungal strains, bacteria, and yeasts, to control plant pests at both pre- and post-harvest stages. Innovative prototype sprayers equipped with variable rate control systems have been developed to optimize PPP application. These sprayers adjust the application rate based on real-time data, ensuring that pesticides are applied only where needed, thus minimizing chemical use and environmental impact. Coupled with Decision Support Systems (DSS), which integrate weather, soil, and crop data, these technologies provide farmers with actionable recommendations on crop management, irrigation, pesticide use, and harvest timing, promoting sustainable agricultural practices. Recognizing the importance of knowledge dissemination, our research includes capacity-building activities aimed at training farmers, agronomists, and other stakeholders in the adoption of smart digital and AI agricultural technologies. Furthermore, policy recommendations derived from data collected through early warning, detection, and response systems support European Union goals of reducing pesticide use and managing plant disease outbreaks. The integration of AI, IoT, and biocontrol agents into plant protection strategies offers a transformative approach to agriculture. By enabling early and accurate

disease detection, efficient use of agrochemicals, and informed decision-making, these technologies contribute to reducing crop loss, improving yield, and promoting sustainable agricultural practices. Our research underscores the potential of smart digital technologies in fostering regenerative, resilient, and sustainable agriculture.

The presented research has received funding from the European Union's Horizon Europe research and innovation programmes under grant agreement No 101134750 (<https://stella-pss.eu/>).

Integrating Plant Health into One Health through One Biosecurity. P.E. HULME. *Centre for One Biosecurity Research, Analysis & Synthesis, Lincoln University, New Zealand.* E-MAIL: philip.hulme@lincoln.ac.nz

Invasive alien species are arguably one of the most complex and undervalued emerging biological threat to global health. It is increasingly recognised that human activities have spread pathogens, parasites and their vectors across the world fuelling multiple epidemics and pandemics. However, the considerable impact invasive alien pests, pathogens and weeds have on ecosystem services supporting food and water security and good quality of life remain poorly quantified. As a result, biosecurity interventions essential to prevent, contain or eradicate these biological threats remain chronically under resourced and poorly implemented in most parts of the world. The transboundary nature of biological invasions is facilitated by the international movement of people and goods. Transport networks enable invasive species to rapidly progress from a local problem to a global crisis. Thus, failure to manage a biological threat in one nation can lead to a cascade of economic and environmental consequences across the globe. The current limited regional and global governance of biological invasions undermines any approach to deliver One Health. Embedding biosecurity more strongly within the One Health framework will be the catalyst for a more holistic approach to the management of biological invasions that ensures a common suite of tools, regulatory approaches and policy frameworks are integrated across the human, animal, plant and ecosystem health sectors. Although One Health and biosecurity both aim to protect the health of people, animals, and ecosystems from biological hazards, the two fields remain heavily siloed across distinct policy and research domains. One Health has yet to fully integrate environmental perspectives especially biological invasions, into its workplan while biosecurity lacks an effective inclusion of social and health sciences, further hindering collaboration.

This holistic approach—recently termed “One Biosecurity”—requires a step change in how biological invasions are managed. This includes overcoming and crossing disciplinary barriers to strengthen surveillance systems, extend risk assessments to encompass the breadth of potential impacts, ensure open data sharing, and coordinate effective public engagement. One Biosecurity offers a vital interdisciplinary framework that bridges human, animal, plant, and ecosystem health sectors, fostering a stronger connection between biosecurity and One Health. This comprehensive approach spans the entire biosecurity continuum, from pre-border intelligence scans to border inspections and post-border incursion management, enabling more effective responses to the threats posed by biological invasions. By uniting these efforts, One Biosecurity could engage a broader group of multilateral organizations, bring together diverse stakeholders, and implement balanced strategies that better safeguard human health, agriculture, production systems, and the natural environment. It is in the interest of each individual government to support the biosecurity of its neighbouring countries, of its regional partners, and, indeed, globally to effectively address the threat of invasive alien species. Integrated governance is critical to enable the international community to act collectively to prevent local biological threats from escalating into global emergencies. Such coordination will be critical to underpinning a One Health framework which ultimately can prevent and mitigate health threats that emerge at the interface between humans, animals, plants, and the environment.

Plenary session - *Xylella fastidiosa* in the Euro-Mediterranean countries: Disclosing new genetic diversity and innovations in diagnostics and disease management

SESSION KEYNOTES

Insect vectors of *Xylella fastidiosa* in the EU: an overview. C. LAGO. *IFISC-(CSIC-UIB), Campus Universitat de les Illes Balears Carretera de Valldemossa, km 7,5 Edificio Científico- Técnico, 07122 Palma de Mallorca, Islas Baleares, Balearic Islands, Spain.* E-MAIL: claralago@ifisc.uib-csic.es

Xylella fastidiosa (*Xf*) is a xylem-limited, plant-pathogenic bacterium responsible for severe diseases in a wide range of economically important crops such as olives, grapevines, almonds, and citrus. This bacterium relies on insects as vectors to spread through ecosystems, mak-

ing them a critical component in *Xf* epidemiology and management. Putative vectors of *Xf* feed exclusively on xylem sap, a characteristic shared by insects belonging to the order Hemiptera, suborder Cicadomorpha, including the superfamilies Cercopoidea (spittlebugs), and the family Cicadellidae, subfamily Cicadellinae (sharpshooters) and to less extent Cicadoidea (cicadas). In the Americas, sharpshooters are the primary vectors of *Xf*, while in Europe, the main vectors are spittlebugs (Hemiptera: Aphrophoridae). The meadow spittlebug, *Philaenus spumarius*, has been identified as the most relevant vector of *Xf* in Europe. Additionally, the recent detection of *Draeculacephala robinsoni* (Hemiptera: Cicadellidae: Cicadellinae), a sharpshooter native to the Americas, raises new concerns and underscores the need for continuous monitoring of potential vector introductions. This presentation provides an overview of insect vectors of *Xf* in the EU. Key aspects of vector biology, ecology, and behaviour will be addressed, including *Xf* transmission, life cycle, host plant preferences, dispersal behaviour, and vector control strategies aimed at preventing disease spread. We will specifically highlight studies carried out by our research group over recent years, in combination with the available literature on European vectors. Our studies on the population dynamics and distribution of the main vectors of *X. fastidiosa* in olive groves in the Iberian Peninsula will be summarized. Furthermore, our studies show that the dispersal abilities of spittlebugs have revealed that adult spittlebugs are capable of travelling long distances—far greater than previously assumed. This, combined with the fact that they remain infectious for life, suggests that reducing nymphal populations is essential to manage *Xf* vectors and avoid disease spread. In addition, our research on spittlebug development and its correlation with the temperature enabled us to develop a Growing Degree Day (GDD) model to predict egg hatching and vector phenology under different climatic scenarios. This model has been used to develop a decision-support tool for optimizing the timing and effectiveness of vector control measures across various agroecosystems. Furthermore, experimental insights such as Electrical Penetration Graph (EPG) studies analysing feeding behaviour of *P. spumarius* and its relationship to pathogen transmission will be briefly presented. Finally, our studies on ground cover crops have led to the identification of repellent, trap, and attractive plants. Based on our findings we propose a combined “push-pull” strategy to manage *P. spumarius* populations: repellent plants used to deter vectors from the main crop (push), while trap crops attract vectors to specific areas (pull), facilitating their elimination. We also explored the use of entomopathogenic fungi (EF),

analysing both lethal and sublethal effects against *P. spumarius*, and discuss the potential of EF as a sustainable alternative for *Xf* vector control.

Current status of *Xylella* in the Mediterranean with a focus on the evolution in Apulia, Italy. D. BOSCIA. CNR – Institute for Sustainable Plant Protection, Via Amendola 122/D, 70126 Bari, Italy. E-MAIL: donato.boscia@cnr.it

The discovery of an outbreak of *Xylella fastidiosa* in Puglia in 2013 represented the first case of established bacterial infections in EU. The pathogen is regulated as quarantine organism in EU and in the EPPO region (listed in the EPPO A2 list). Its finding in association with the olive quick decline syndrome, the devastating epidemic disease affecting this major Mediterranean crop, prompted the European Union to issue a strict regulation that, among the prevention measures, obliged all member states to carry out annual monitoring campaigns. Unexpectedly in the framework of this surveillance program several bacterial outbreaks were discovered mainly in southern Countries, affecting different crop species and caused by genetically different strains. Investigations showed they were mostly related to inadvertent introductions from north America, occurred recently or in the past decades. Differently from the situation determined by the spread of isolates harboring the *pauca* ST53 genotype in Puglia, in these outbreaks strains belonging to other subspecies with much limited impact have been characterized (i.e. infections were overlooked until sampling and laboratory tests led to its detection). These include outbreaks reported in Italy, in France, Portugal and Spain, caused by strains belonging to the subspecies *multiplex* and, to a lesser extent, also to the subspecies *fastidiosa*. Therefore, the first detection in Italy in 2013 resulted in a much more complex scenario than expected. Due to the emergence on olives, the Apulian authorities implemented a particularly intense monitoring program with over 1.2 million plants sampled and analyzed in the past 11 years. A program that probably represents an *unicum*, which is not sustainable in the long term, but it is certainly a useful case study to clarify the true status of the presence of *X. fastidiosa* in Europe. From the initial outbreak in Gallipoli (south-west of the Salento peninsula), the spread of the *pauca* isolates occurred at high rate until 2018, then decreased significantly, even so, currently, the demarcated infected area corresponds to the 40% of the region (about 8,000 km²). Moreover, in 2024, while monitoring the *Xf*-free area, in the central part of the region outbreaks associ-

ated to two other subspecies were discovered: *multiplex*, with isolates harboring the genotype ST26, and *fastidiosa*, with isolates harboring the genotype ST1. In the first case, infections affect almost exclusively almond trees, apparently not showing any specific or manifest *Xylella*-symptomatology. The occurrence of ST26-infected trees occurs as spotted infected trees in three areas distant some km each other (up to 50km), currently over 35,000 samples analyzed approx. 600 plants tested positive. The demarcation is not yet definitive as the delimiting survey is still ongoing and, in some area, it overlaps the demarcated areas established for the concomitant occurrence of isolates of the other subspecies. All this, combined with the absence of specific symptoms, leads to the hypothesis that it is due to an old introduction that, having gone unnoticed, has been able to establish itself and spread undisturbed for years. The situation is different for the outbreak of the *fastidiosa*, discovered following a program of monitoring of insect vectors. In this case, the analysis of about 50,000 samples (mainly grapes and almond) has allowed to delimit the outbreak in a relatively small area, about 8 km², surrounded by a 2.5 km wide belt without the presence of the bacterium. In this case, inspections revealed the occurrence of severely affected grapes, suggesting that the management of this outbreak should be carefully designed. Overall, the scenario emerged in Apulia confirms how complex and heterogeneous the occurrence of the bacterium is, suggesting to adopt targeted management measures adapted case by case rather than a unique approach.

BIOVEXO: Can we find solutions to protect plants against *Xylella*? S. COMPANT, THE BIOVEXO CONSORTIUM. Bioresources Unit, AIT Austrian Institute of Technology, Konrad Lorenz Strasse 24, 3430 Tulln, Austria. E-MAIL: stephane.compant@ait.ac.at

Xylella fastidiosa is increasingly causing diseases in olive trees and various 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 35-70% in olive 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 *Philaenus spumarius*. Several chemical insecticides are authorized to control xylem-feeding insects. For products allowed in organic farming, only

temporary authorizations, for limited periods, have been obtained. In BIOVEXO, 11 partners from 5 different countries aim to develop environmentally sustainable and economically viable plant protection solutions to control *Xylella* and its insect vector. After 4 years of the BIOVEXO project, several biopesticides have been upscaled and formulated. They are still currently being assessed and applied via foliar spray or drip irrigation on largescale field trials (olive and almond trees) as preventive and curative approaches. They are also applied in an integrated pest management program in Italy (Apulia). Results obtained so far show that some pesticides reduce the insect vector populations, while some others reduce *Xylella* symptoms on plants such as olive trees, not almonds, and only under certain conditions as assessed after 3.5 years of application. In the case of biopesticides reducing *Xylella* symptoms, only symptoms have been recorded as reduced in curative approaches, while the pathogen population seems not to be affected. A further assessment in the last year of the BIOVEXO project should be carried out to confirm these results obtained so far. However, data from field applications also show biostimulant effects against abiotic stress and an increase in plant vigor, particularly on olive trees treated with some BIOVEXO products. Results from toxicity and genotoxicity tests as well as from life cycle assessment and the shelf life of products showed further that biopesticides can be relevant for the market. Putative mechanisms of action of biopesticides targeting *Xylella* or the insect vector involve direct biocontrol properties, biostimulant effects on olive trees, and induced systemic plant resistance against *Xylella*. Insect and plant microbiomes following biopesticide applications are also currently analyzed. All the results suggest multiple potential future applications of products, i.e., as biostimulants to improve plant growth, protect plants against abiotic stress, or as biopesticides to reduce *Xylella* symptoms and vector populations.

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ORAL PRESENTATIONS

Grapevine recovery from Pierce's disease under Mediterranean conditions. O. BAHAR¹, M. VANUNU^{1,2}, O. DROR¹, E. AMZALLAG^{1,2}, Y. OMER¹, T. ZAHAVI³, N. MAOZ⁴. ¹Department of Plant Pathology and Weed Research, Agricultural Research Organization – Volcani Institute, Rishon LeZion, 7505101, Israel. ²The Robert H.

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Pierce's disease (PD) of grapevine, caused by *Xylella fastidiosa* (Xf), results in systemic colonization of the xylem and often leads to chronic infections. Although recovery from PD has been documented in California vineyards, where some infected vines were shown to clear the infection, the underlying mechanisms remain poorly understood. Notably, despite the growing number of PD reports from other regions over the past decade—including Europe and the Middle East—recovery has neither been systematically reported nor studied outside California. To investigate whether recovery from PD can occur under Mediterranean climate we inoculated Cabernet sauvignon and Chardonnay grapevines at different time points from May to September and examined infection and disease symptoms for two consecutive years. During the dormancy period, grapevines were placed in two locations representing different growing climates in Israel. Infection and symptoms incidence were over 80% when inoculation was done in May or July, while only ~60% infection (with no symptoms) was seen in September inoculations. Over 90% of the plants inoculated in September recovered from the infection, in contrast to ~20% of the plants inoculated in May. Cultivar or dormancy location did not significantly affected recovery. These results demonstrate that PD recovery occurs under Mediterranean climate and confirm previous findings that inoculation timing is a critical determinant of disease outcome. Together with studies evaluating recovery in vineyards, these results will help define critical inoculation windows associated with chronic infection, which will be instrumental in designing effective PD management strategies tailored to Mediterranean climates.

This research was financially supported by the Chief Scientist of the Ministry of Agriculture of Israel, the Israel wine grape board and the BeXyl project (Beyond Xylella, Integrated Management Strategies for Mitigating Xylella fastidiosa Impact in Europe; grant 101060593, from European Union's Horizon Europe "Food, Bioeconomy Natural Resources, Agriculture and Environment" Programme.

Impact of suboptimal and extreme temperatures on *Xylella fastidiosa* survival in the Mediterranean area. M. ROMÁN-ÉCIJA, C. OLIVARES-GARCÍA, B.B. LANDA, J.A. NAVAS-CÓRTES. *Instituto de Agricultura Sos-*

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Temperature plays a key role in the biology and ecology of *Xylella fastidiosa* (Xf). Previous studies have shown that Xf displays a broad optimal temperature range which, combined to its adaptability to diverse environments, increases the risk of establishment and spread in Mediterranean climate- type regions. However, the effects of extreme temperatures on Xf survival remain poorly understood. This study evaluated whether different thermal treatments (TT) (from -20 to 4°C and from 40 to 50°C) at varying exposure times have a lethal or bacteriostatic effect on several Xf strains of different subspecies. To assess cell survival bacterial growth and biofilm formation were monitored, and live and dead cells were quantified using viability-qPCR after TT. Our results showed that Xf was able to survive TT below 4°C, including freezing temperatures, maintaining among 21 and 45% of viable cells, enabling the recovery of growth and biofilm formation once incubated under favourable conditions of 28°C. In contrast, TT at 40°C had a bactericidal effect after 24 to 72 hours of exposure. Treatments above 42°C with shorter exposure times had variable results. Even though cell viability was significantly reduced (survival rate from 0.99 to 7.38% after incubation exposure of 60 minutes), a substantial number of cells remained viable, although with growth reductions of 87– 96% compared to untreated controls. These findings provide valuable information for the development of more accurate risk maps and to support the development and implementation of TT in nurseries as a phytosanitary measure.

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Preliminary transcriptomic analyses reveal *in vitro* and *in planta* overexpression of various bacteriocins in *Xylella fastidiosa*. S.S. AMOIA, M. SAPONARI, P. SALDARELLI, A.M. LIGORIO, C. DEL GROSSO, G. LOCONSOLE, G. D'ATTOMA, D. BOSCIA, A. GIAMPETRUZZI. *National Research Council (CNR), Institute for Sustainable Plant Protection, Via Amendola 165/A Bari, Italy. E- MAIL: serafinaserena.amoia@cnr.it*

Xylella fastidiosa (Xf) is a xylem-limited phyto bacterium of worldwide importance, associated with severe plant diseases, including Olive Quick Decline Syndrome

(OQDS). Despite extensive research on plant responses to *Xf* infection, bacterial gene expression dynamics within the host remain still poorly explored. A pilot application of dual RNA sequencing (dRNA-seq) was conducted to profile bacterial transcripts in xylem-enriched tissues of a naturally infected olive tree. A gene encoding a colicin V-like bacteriocin (*cvaC-1*), a pore-forming toxin, emerged as the most highly regulated, surpassing even housekeeping genes. The overexpression of a Blp family class II bacteriocin was also observed in RNA-seq libraries obtained from *in vitro*-grown *Xf* strains. This result was validated in a time-course experiment by testing total RNA extracts of artificially inoculated olive, citrus, and periwinkle plants using the newly developed probe-based RT-qPCR assay targeting *cvaC-1*. Notably, the test showed to be more sensitive and accurate than the conventional qPCR, widely employed for bacterial DNA detection, also being able to distinguish between growing and non-growing bacterial cells in tissues, particularly for those plant matrices from which it is laborious to retrieve *Xf* colonies on artificial media. These data support the use of *cvaC-1* as a novel biomarker to be applied in the research context for an earlier *Xf* detection, to track the bacterial progression within plants, thus predicting the pathogen-host compatibility, and to assess the impact of antimicrobial compounds. This work contributes to a better understanding of *Xf* interaction mechanisms with host plants and endophytic microbial communities.

This research was partially supported by the Regione Puglia (DD no. 495 del 14/10/2015 and no. 279 del 9/8/2016, Progetto di ricerca Linea B-STIPXYT and by Ministero dell'agricoltura, della sovranità alimentare e delle foreste in the framework of the Research Project 1LIVEXYLELLA - ID01- Trasmissione D.M. n. 664519 del 28/12/2022 (Tecnologie portatili e protocolli innovativi per la diagnosi ultrasensibile di Xylella fastidiosa direttamente in piante e vettori).

Sustainable management of *Xylella fastidiosa* through innovative delivery of plant extracts. D. SCHIAVI¹, M.A. MUAWIYA¹, L. FELICI¹, C. MICCOLI¹, E. GHERBALI¹, D. RONGAI², M.G. DI SERIO², R. RONCHETTI³, P. WOJTYLO³, S. GIOVAGNOLI³, E. CAMAIONI³, V. TAGLIAVENTO⁴, G.M. BALESTRA¹⁻⁴. ¹Dipartimento di Scienze Agrarie e Forestali, Università degli Studi della Tuscia, Viterbo, Italy. ²CREA-IT PE Centro di ricerca Ingegneria e Trasformazioni agroalimentari, Pescara, Italy. ³Dipartimento di Scienze Farmaceutiche, Università degli Studi di Perugia, Italy. ⁴Phy.Dia. srl, Viterbo, Italy. E-MAIL: balestra@unitus.it

Bioactive compounds from agricultural waste could represent a valuable tool in the management of plant health issues if paired with targeted delivery systems which enhance antimicrobial properties, plant absorption, and chemical stability. This research has been evaluating a plant protection strategy against *Xylella fastidiosa* subsp. *pauca* (*Xfp*) that combines natural molecules with nanoscale delivery technologies. In particular, pomegranate peel extract (PGPE) demonstrated the capability to reduce the growth of *Xfp in vitro* starting from 0.5% w/v. PGPE was evaluated through endotherapy and root application, exploring its biostimulant and eliciting activity on olive plants. Experiments were conducted both on 2-year-old olive plants and on cuttings of 'Leccino' and 'Ogliarola'. Parameters evaluated at different time points included Nitrogen Balance Index (NBI), root-to-total plant weight ratio, and soil pH, while the expression of genes related to photosynthetic, plant defence and polyphenol pathways were analyzed in leaves. Sap from the cuttings was also extracted, and the main metabolites were identified through UHPLC/HRMS. Furthermore, the expression of genes involved in oxidative stress response and hormone biosynthesis was evaluated in root tissues. Results revealed both biostimulant and resistance activation in plants treated with our formulations, especially at 30 days post-treatment. Interestingly, significant differences were observed among the two tested cvs, suggesting specific responses to the treatments. Preliminary experiments on artificial *Xfp*-pathosystems, using tobacco plants, are under evaluation to determine the control efficacy of the proposed approaches on the disease progression as well as open field trials in *Xfp* naturally infected areas.

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Integration of membrane-based DNA extraction with LAMP for rapid detection of *Xylella fastidiosa* in olive trees. W. MELLIKECHE¹, R. CARACCILO², M. COPPOLA², G. ARCOLEO², M. GALLO¹, A.M. D'ONGHIA¹, F. VALENTINI¹. ¹International Centre for Advanced Mediterranean Agronomic Studies, Via Ceglie, 9-70010, Valenzano (BA), Italy. ²Enbiotech SRL, Via Masuccio Salernitano, 28-84012, Angri (SA), Italy. E-MAIL: mellikeche@iamb.it

Early and accurate detection of plant pathogens is critical for managing outbreaks of destructive diseases such as Olive Quick Decline Syndrome (OQDS) in olive trees,

caused by *Xylella fastidiosa*. This study evaluates a novel diagnostic protocol that integrates a membrane-based DNA extraction method with real-time loop-mediated isothermal amplification (LAMP) to enhance sample handling and enable rapid pathogen detection. Naturally infected olive stem imprints on nitrocellulose were stored at room temperature and tested after their extraction by real-time LAMP over five months. These samples were compared with extracts obtained from conventional plant material stored at +4 °C and -20 °C. Validation of the LAMP assay combined with the membrane-based extraction method was performed in parallel with a reference qPCR method. Results showed that the membrane-based extraction method maintained high detection accuracy, while enhancing biosafety, facilitating sample handling, and reducing processing time. Furthermore, the real-time LAMP method achieved 100% diagnostic specificity and high sensitivity with a detection limit of 10^2 cfu/mL. This low-resource, storage-friendly protocol enhances LAMP's potential for routine detection of *X. fastidiosa* in field surveys, nurseries and control checkpoints, particularly within surveillance.

POSTER PRESENTATIONS

Early detection of *Xylella fastidiosa*-induced symptoms through hyperspectral imaging using a phenotyping platform. V. EGEE-COBRERO, J.A. JIMÉNEZ-BERNI, R. CALDERÓN, P.J. ZARCO-TEJADA, M. ROMÁN-ÉCIJA, G. LEÓN-ROPERO, A. HORNERO, J.A. NAVAS-CORTÉS, B.B. LANDA. IAS-CSIC, Institute for Sustainable Agriculture - Spanish National Research Council, Av. Menéndez Pidal s/n, 14004 Córdoba, Spain. E-MAIL: vegea@ias.csic.es

Xylella fastidiosa (Xf) detection is hindered by asymptomatic infections, long incubation periods and non-specific symptoms, allowing infected plants to spread the pathogen before visible symptoms appear. A phenotyping platform that integrates a motorized vertical sliding mechanism, an illumination device, and a high-resolution hyperspectral imaging camera was designed to monitor and quantify physiological and biochemical responses in plants after Xf infection under controlled conditions. Xf subsp. *fastidiosa* (IVIA5770) was inoculated in two commercial grapevine cultivars and incubated under full and deficit irrigation and three temperature-CO₂ combinations simulating current and future climate change scenarios. Hyperspectral images spanning 272 bands in the VNIR range (400 to 1000 nm) with 6 nm spectral and 3.5 mm spatial resolutions were acquired

at 2- and 4- months post-inoculation. Additionally, Xf-infection was monitored via qPCR analysis of leaf petioles. An image processing pipeline was developed, including smoothing and segmentation techniques to extract pure vegetation pixels using thresholding within regions of interest in supervised classification algorithms. Furthermore, 70 vegetation spectral indices (VIs) were calculated and integrated into machine learning classification models to distinguish between Xf-inoculated and non-inoculated plants. Preliminary results show that VIs captured treatment-related differences, with tree-based and gaussian-based models achieving over 70% validation accuracy in detecting early Xf-infections. This phenotyping platform holds great potential for early detection of Xf at asymptomatic stages and can facilitate the selection of Xf-resistant genotypes in breeding programs, providing at the same time valuable information on grapevine physiological responses under various environmental stress conditions including future climate change scenarios.

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Integrating commercial fertilizers into IPDM strategies mitigates Olive Quick Decline Syndrome in Southern Italy. C. DEL GROSSO^{1,2}, M. SAPONARI², P. SILDARELLI², D. PALMIERI¹, G. ALTAMURA³, R. ABOU KUBAA², A. PASTORINI¹, F. DE CURTIS¹, G. LIMA¹. ¹Department of Agricultural, Environmental and Food Sciences, University of Molise, 86100, Campobasso, Italy. ²Institute for Sustainable Plant Protection, National Research Council (CNR), 70126 Bari, Italy. ³CRSFA- Centro Ricerca, Sperimentazione e Formazione in Agricoltura Basile Caramia, 70010 Locorotondo, Italy. E-MAIL: lima@unimol.it, carmine.delgrosso@cnr.it

The management of *Xylella fastidiosa* subsp. *pauca* (Xfp) remains a major challenge due to the limited availability of effective antimicrobial, few chemical control options, and increasing restrictions on the use of traditional bactericides, particularly high dose of copper-based compounds, in the European Union. Therefore, alternative approaches, such as the evaluation of certain systemic fertilizers/biostimulants improving plant disease resistance and/or displaying bactericidal side effects, are increasingly being explored. This study investigates the antibacterial side effects of certain commercial fertilizers containing low concentration of copper and/or zinc complexed with phosphites and/or bioavailable silicon. These compounds were initially selected through in vitro assays for

their activity against *Xfp*. Greenhouse trials confirmed the effectiveness of these products in reducing disease severity and pathogen populations. In open field trials, when included into an Integrated Pest and Disease Management (IPDM) strategy, including agronomic practices and insect vector control measures, these treatments effectively reduced symptoms and improved fruit yield. Beyond their antimicrobial properties, they may also enhance plant immunity, as similar compounds are known to trigger defense mechanisms in other pathosystems. The results suggest that selected systemic fertilizers with antimicrobial properties offer a promising and sustainable approach to *Xfp* control, reducing the reliance on traditional bactericides, and improving crop resilience. This strategy is particularly important to safeguarding olive production in *Xfp*-affected areas such as Apulia, where centuries-old olive groves represent not only an agricultural resource but also an invaluable cultural heritage. Further research is needed to optimize and confirm their long-term effectiveness under broader field conditions.

This research was financially supported by INTEGROLIV project "Integrated Eco-friendly Approach for the Containment of Xylella fastidiosa and for the Regeneration of Olive Growing and the Environment (Project Code H33C22000860001) - Minister of Agriculture, Food Sovereignty and Forests Italy (MASAF), D.M. n. 664829 del 29/12/2022 – Research Topic 2: Investigations and tests to identify methods of control of Xylella fastidiosa. The findings presented are based on a study accepted for publication in Plant Disease DOI: <https://doi.org/10.1094/PDIS-08-24-1770-RE>.

Management of *Xylella fastidiosa* disease in almond plants using a peptide that interferes with twitching motility. L. MOLL¹, M. PLANAS², L. FELIU², L. DE LA FUENTE³, B.B. LANDA¹, E. MONTESINOS⁴, E. BADOSA⁴, A. BONATERRA⁴. ¹Institute for Sustainable Agriculture (IAS) of the Spanish National Research Council (CSIC), Spain. ²LIPPSO, Department of Chemistry, Campus Montilivi, University of Girona, 17003 Girona, Spain. ³Department of Entomology and Plant Pathology, Auburn University, AL 36849 Auburn, Alabama, United States of America. ⁴Laboratory of Plant Pathology, Institute of Food and Agricultural Technology-CIDSAV, Campus Montilivi, University of Girona, 17003 Girona, Spain. E-MAIL: ljmoll@ias.csic.es

Xylella fastidiosa is a xylem-limited bacterial pathogen that poses a major threat to global agriculture, severely affecting economically important crops such as almond. Within the plant xylem, the bacterium displays a dual lifestyle, alternating between sessile microbial aggregates

and planktonic cells that move by twitching motility. Movement is mediated by type IV pili which is crucial for systemic colonization of the host. Although previous studies have demonstrated that peptides can modulate key biological processes in *X. fastidiosa*, their impact on motility had not yet been investigated. In this work, we evaluated the effect of both previously identified peptides and newly designed analogs on *X. fastidiosa* motility *in vitro*, and explored their protective potential against almond leaf scorch disease. By measuring the twitching fringe width and through microfluidic chamber observations, we demonstrated that BP100 significantly inhibits twitching motility. Interestingly, transmission electron microscopy observations showed that BP100-treated cells exhibited type IV pili that were similar in frequency, length, and morphology to those of non-treated controls, suggesting that the inhibition was not due to structural defects. Furthermore, endotherapy application of BP100 to almond plants inoculated with *X. fastidiosa* under greenhouse conditions resulted in a significant reduction of bacterial populations and xylem vessels were less severely affected, which correlated with a decrease in disease symptoms. Overall, our findings reveal that BP100 not only disrupts the motility of *X. fastidiosa*, but also contributes to disease mitigation *in planta*, highlighting its potential as a promising tool for the management of almond leaf scorch.

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Exploring xylem-Inhabiting bacteria in synthetic communities for the control of vascular diseases in olive trees. L. ALONSO-VILLAR¹, N.J. FLORES-DUARTE², M. ROMÁN-ÉCIJA¹, M.P. VELASCO-AMO¹, C. OLIVARES-GARCÍA¹, J.A. NAVAS-CORTÉS¹, C. HARO¹, L. MOLL¹, B.B. LANDA¹. ¹Department of Crop Protection, Institute for Sustainable Agriculture, Spanish National Research Council, Córdoba, Spain. ²Department of Microbiology and Parasitology, University of Seville, Seville, Spain. E-MAIL: lalonso@ias.csic.es

Infections caused by the fungus *Verticillium dahliae* (Vd) and the bacterium *Xylella fastidiosa* (Xf) pose major threats to economically important crops such as olive (*Olea europaea* L.). In the context of the European Green Deal and its emphasis on sustainable agriculture, endophytic microorganisms are emerging as a promis-

ing strategy for the biological control of plant pathogens. In particular, synthetic microbial communities (SynComs) composed of xylem-inhabiting bacteria from olive trees can offer a novel approach for the control of these vascular plant pathogens. In this work, over 100 bacterial strains isolated from olive xylem were characterized for phenotypic and genetic traits to assess their potential as biocontrol agents and/or plant growth promoters. The analyzed traits included: (i) carbon source assimilation and tolerance to abiotic stresses (antibiotics, low pH, salinity); (ii) plant growth- promoting features (siderophore production, indole-3-acetic acid synthesis, nitrogen fixation); (iii) antimicrobial enzyme production (amylase, chitinase, protease, etc.); and (iv) in vitro inhibition of *Vd* and *Xf*. Compatibility assays among some of the most promising strains enabled the design of two SynComs, each consisting of three compatible strains. SynCom biocontrol activity is currently being tested against *Vd* and *Xf* in vitro using dual culture plates and microfluidic chambers simulating xylem vessels. Their *in planta* efficacy is being assessed in various pathosystems: *Xf* in *Nicotiana benthamiana* (model plant), *Xf* in olive, and *Vd* in olive. Additionally, the capacity of the SynComs bacterial members as well as both pathogens to systemically colonize xylem vessels after endotherapy application will be monitored via species-specific qPCR.

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Genomic characterization and phylogenetic analysis of *Xylella fastidiosa* subsp. *fastidiosa* strains from Cova da Beira region, Portugal. E. GARCIA^{1,2}, A.K. KAHN³, C. RODRIGUES², A. CAMELO^{1,4}, C. ESPÍRITO SANTO^{1,4}, H.D. COLETTA-FILHO⁵, R.P.P. ALMEIDA³, J. COSTA^{1,2}. ¹University of Coimbra, Centre for Functional Ecology, Associate Laboratory TERRA, Department of Life Sciences, Coimbra, Portugal. ²Instituto Pedro Nunes, Laboratory for Phytopathology, Coimbra, Portugal. ³Department of Environmental Science, Policy, and Management, University of California, Berkeley, Berkeley, CA 94720, USA. ⁴CATAA - Centro de Apoio Tecnológico Agro-Alimentar, Castelo Branco, Portugal. ⁵Centro APTA Citros Sylvio Moreira, Instituto Agronômico de Campinas, Cordeirópolis, SP, Brazil. E-MAIL: jcosta@uc.pt

The emergence of *Xylella fastidiosa*, a xylem-limited bacterial pathogen with a broad host range, represents a significant threat to plant health and agricultural sustainability in Mediterranean ecosystems. In this study, six *X. fastidiosa* subsp. *fastidiosa* strains isolated from the Beira Interior region of Portugal, an important area for fruit production, were analysed. Whole-genome sequencing and phylogenetic analyses were performed to genetically characterize these strains and determine their origin. All strains belong to sequence type ST1, known to infect economically important crops such as cherry, plum, almond, and grapevine. The isolates were obtained from both cultivated (*V. vinifera*) and native (*Cytisus* sp.) plant species, indicating the potential role of wild vegetation as a reservoir for the pathogen. Phylogenetic reconstruction suggests a single introduction event linked to California, USA, with molecular clock analysis estimating the introduction between 2002 and 2016, with the strains forming a clade that diverged circa 2020. The increasing number of demarcated areas where *X. fastidiosa* has been detected in Portugal, along with the co-occurrence of subsp. *fastidiosa* and *multiplex* highlight the need for sustained surveillance efforts in both agricultural and unmanaged ecosystems. Moreover, the extent of *X. fastidiosa*'s establishment and its long-term ecological and economic impact on Portuguese landscapes remain largely unknown, emphasizing the necessity of intensified research efforts. The findings of this study provide insights into the pathogen's introduction in Portugal and reinforce the importance of proactive measures to protect Mediterranean agroecosystems.

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Screening the response of Greek olive cultivars to *Xylella fastidiosa* infections. M.M. MATHIOUDAKIS¹, N. KAVROULAKIS¹, G. KOUBOURIS¹, A.M. LIGORIO², V. CAVALIERI², R. SPANÒ², P. SILDARELLI², M. SAPONARI². ¹Institute of Olive tree, Subtropical Crops and Viticulture, Plant Pathology Laboratory, Karamanlis Ave. 167, GR-73134, Chania, Crete, Greece. ²National Research Council -Institute for Sustainable Plant Protection, Via Amendola, 122/D Bari, 70126, BA, Italy. E-MAIL: maria.saponari@cnr.it

In this work we evaluated the response of the major Greek olive cultivars to the infections caused by *Xylella fastidiosa* subsp. *pauca* (*Xfp*). Isolates of this subspecies have been found to severely impact olives, with severe desiccations on the canopies leading to tree decline in highly susceptible cultivars. Huge research efforts are devoted to screen the response to the infections of a large number of cultivars representing the vast Mediterranean olive germplasm. In the framework of the projects CERTIFIED NURSERIES^a and XYLEVA^b a collaboration was established between the Greek and Italian research centers to assess the vulnerability of Greek cultivars to *Xfp*. Briefly, plants of the cultivars Adramitini, Kolovi, Throubolia N. Aegean, Amfissis, Chalkidikis, Gaidourelia, Throuba Kritis, Throuba Thassou and Tsounati, are currently under testing. For the first three cultivars, phenotypic assessment and diagnostic tests have been completed at 2 years post-inoculation, while they are still ongoing for the remaining cultivars. For these three cultivars systemic infections were detected in 80- 100% of the replicates, similar to that of the susceptible control, the cultivar Ogliarola salentina, which also showed defoliation, shoot dieback and complete desiccation of the plants. Although, none of the infected plants of the three N. Aegean cultivars showed severe symptoms as those reported in the control, defoliation and shoot dieback were recorded, with slightly higher impact on Throubolia N. Aegean. It should be remarked that in the cv Adramitini a slower rate of plant colonization and symptoms progression were recorded. Assessments are ongoing for the remaining cultivars.

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Evaluation of molecular tests for the determination of the *Xylella fastidiosa* subspecies: results from a European test performance study and a proficiency test. T.M. RAAJMAKERS, R.A.M. VREEBURG, A.A.L.A.M. VAN DUIJNHOFEN, M.A.W. VOGELAAR, M. BERGSMAN-VLAMI. Netherlands Institute for Vectors, Invasive plants and Plant Health (NIVIP), National Plant Protection Organization (NPPO), Netherlands Food and Consumer Product Safety Authority (NVWA), Geertjesweg

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Xylella fastidiosa (*Xf*) is an European Union (EU) priority pest with a broad host range. Plants can be infected with *Xf* for prolonged periods before symptoms develop. Symptomless plants are a source for introduction and spread of the bacteria. The detection of *Xf* has been harmonized among the EU national reference laboratories (NRLs). Until recently, *Xf* subspecies determination required a laborious multilocus sequence typing (MLST) test. Over the past few years, two real-time PCR tests have been published that focus on the *Xf* subspecies designation. These real-time PCRs offer several advantages when compared with the MLST test: firstly, they should increase the sensitivity of detection, secondly, they decrease the analysis time and costs for subspecies designation. Furthermore, subspecies determination is more easily achieved in plant extracts. Available validation data was limited, an international test performance study was therefore organized by the Netherlands Institute for Vectors, Invasive plants and Plant health (NIVIP) in its role as EU Reference Laboratory for pests of plants on bacteria (EURL) to evaluate the novel real-time PCRs. The results showed that these tests are fit for purpose and addition of both real-time PCR tests to EU legislation (Annex IV B of Commission Implementing Regulation (EU) 2020/1201) was recommended and implemented. Finally, a proficiency test on the determination of *Xf* subspecies was organized by NIVIP to assess the diagnostic competence of EU NRLs. The results revealed a high level of accuracy among participating laboratories.

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In vitro screening of antimicrobial potential of *Myrtus communis* extract against *Xylella fastidiosa* subsp. *pauca*, *multiplex* and *fastidiosa*. G. INCAMPO*, M. MOUROU*, D. CORNACCHIA, V. MONTILON, D. GERIN, F. FARETRA, F. NIGRO, S. POLLASTRO. Department of Soil, Plant and Food Sciences-University of Bari Aldo Moro, via Amendola 165/A 70126 Bari, Italy E-MAIL: francesco.faretra@uniba.it. *These two authors contributed equally to this work

Xylella fastidiosa is a Gram-negative bacterium responsible for severe damage to several economically important crops worldwide. The absence of effective and sustainable management strategies highlights the urgent need for innovative antibacterial solutions. Screening

plant-derived extracts for antimicrobial activity may offer promising alternatives for the control of xylem-colonizing pathogens, including *X. fastidiosa*. In this study, *in vitro* well diffusion and broth dilution assays were performed to evaluate the antimicrobial activity of *Myrtus communis* extract against *X. fastidiosa* subspecies *pauca*, *multiplex*, and *fastidiosa*, all reported in Apulia. Clear inhibition zones (ranging from 4 to 20 mm) were observed for all tested subspecies compared to the control. Moreover, *Myrtus* extract significantly reduced the growth of *X. fastidiosa* subsp. *pauca* by 45% after 336 hours in liquid cultures. Similar inhibitory effects were recorded for the other subspecies: a remarkable 75% growth reduction for subsp. *multiplex* and a 40% reduction for subsp. *fastidiosa*, relative to their respective controls. These findings suggest that *Myrtus communis* extract exhibits notable antimicrobial potential against different *X. fastidiosa* subspecies. However, further investigations under *in vivo* conditions are necessary to validate its efficacy and assess the feasibility of incorporating such natural products into integrated management strategies for *Xylella*-associated diseases.

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***In vitro* high-throughput screening of the antimicrobial activity of different compounds against Xylella fastidiosa subsp. pauca.** C. DEL GROSSO¹, L. GRANDI², T. LOMBARDI³, G. D'ATTOMA¹, N. SCHMITT², V. R. DE MICHELE², M. SAPONARI¹. ¹*Institute for Sustainable Plant Protection, National Research Council (CNR), Bari, Italy.* ²*Invaio Sciences International GmbH, Basel, Switzerland.* ³*Invaio Sciences Italy SRL, Milan, Italy.* E-MAIL: carmine.delgrosso@cnr.it

In recent years, the distribution and host range of *Xylella fastidiosa* (Xf) have expanded significantly, with severe outbreaks reported in Europe. The subspecies *pauca* (Xfp) is considered the most dangerous, as it causes olive quick decline syndrome (OQDS) devastating olive trees in southern Italy. Despite the severe impact of this pathogen, there are no active substances officially approved to directly cure Xf infections in plants. Management strategies are limited to preventive actions, vec-

tor control, and eradication measures, underlining the urgent need for new therapeutic options. In this context, our research aimed to identify and evaluate new compounds with antimicrobial and antibiofilm activities against Xfp. To address the challenges posed by the slow growth of Xfp, we optimized high-throughput screening protocols to efficiently assess bactericidal and biofilm-inhibiting effects of a broad panel of products, including metal ions, micronutrients, antibiotics, and phenolic compounds. Most products showed a dose-response effect. Among micronutrients and phenolic compounds, CuSO₄·5H₂O, Dentamet®, pyrocatechol, and 4-methylcatechol displayed the highest bactericidal and antibiofilm activity. Antibiotics demonstrated strong bacteriostatic effects and effectively inhibited biofilm formation. Some metal ions, such as CoCl₂, K₂B₄O₇·4H₂O, and MnSO₄·H₂O, significantly impacted bacterial cell viability but did not completely kill Xfp. Regarding biofilm formation, some ions inhibited biofilm, while others promoted it, such as Ca(NO₃)₂·4H₂O and Na₂MoO₄, which enhanced bacterial growth. This study provides valuable insights into the potential use of these compounds for managing Xf infections. The methodologies employed offer a reliable platform for profiling antimicrobial activity and identifying effective compounds for future therapeutic strategies.

This research is based on material licensed under a Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License (CC BY-NC-ND 4.0) already published DOI: <https://doi.org/10.1186/s40538-025-00734-w>. This research received the UNASA 2025 Award, conferred by the National Union of Academies for Sciences Applied to Agriculture and Environmental Protection (UNASA), for its innovative contribution to mitigating the environmental impact of Xylella fastidiosa.

Preliminary screening of new olive genotypes for resistance to Xylella fastidiosa. A. CARLUCCI¹, M.L. RAIMONDO¹, G. RICCIARDI¹, M.G. MOREA, F. INGROSSO², G. VERGARI², F. PENNETTA², F. LOPS¹. ¹*Department of Agricultural Sciences, Food, Natural Resources and Engineering, University of Foggia, Italy.* ²*Centro Studi Olea, Lecce, Italy.* E-MAIL: antonia.carlucci@unifg.it

In the last decade, olive groves in Apulia have faced a significant threat from the quarantine bacterium *Xylella fastidiosa* subsp. *pauca* (XF). It is feared that the spread of this bacterium will expand from Salento to northern Apulia, mainly through insect vectors. Despite intense control efforts by the SSF of the Apulia Region, the XF

infection has also spread to the province of Bari. Among various control approaches being tested, genetic resistance shows promise for significant results. The present work aims to evaluate 608 new olive genotypes collected over 30 years through the crossing of numerous olive varieties. In October 2016, these genotypes were transplanted to a private farm in the Monteroni (LE), a major area infected by XF. No chemical control and fertilization treatments were carried out except to light pruning when it has been necessary. After almost eight years, 522 plants survived, from which samples were collected and analysed for the presence of the bacterium using the EPPO protocol. From the initial pool of 163 genotypes (31.23%) found not to be infected by XF at the first sampling, 45 (8.62%) were identified as XF-resistant. The non-infected olive trees were carefully observed for symptoms of other common olive diseases such as peacock spot, *Cercospora* leaf spot, and knot disease. Preliminary findings suggest that genetic resistance could effectively limit the spread of XF in Apulian olive groves. This study aims to offer new potentially resistant olive genotypes to olive growers of Mediterranean basin, raising their hopes for the future.

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Spectral signatures to reveal *Xylella fastidiosa* infection in the resistant olive cultivar ‘Leccino’. E. CHIAROMONTE¹, M. MAGARELLI¹, V. MONTILON¹, A. AGNUSDEI¹, P. LUCCHESI¹, S. TANGARO^{1,2}, S. POLLASTRO¹, F. NIGRO¹. ¹Department of Soil, Plant and Food Sciences, (DiSSPA), University of Bari Aldo Moro, Bari, via Amendola 165/A, 70126- Bari, Italy. ²National Institute for Nuclear Physics (INFN), Bari Section, Via A. Orabona 4, Bari, 70125, Italy. E-MAIL: franco.nigro@uniba.it

The containment of *Xylella fastidiosa*, the causal agent of Olive Quick Decline Syndrome (OQDS), depends largely on the early detection of infected plants, particularly asymptomatic ones, including recently infected individuals or resistant cultivars such as *Olea europaea* cv. Leccino. Given the xylem-limited lifestyle of the pathogen and its ability to impair water flow through biofilm formation, this study applied hyperspectral imaging to identify infection-specific spectral signatures. Spectral data were acquired using an ASD FieldSpec® 4 spectroradiometer. To standardize the acquisition procedure,

initial comparisons were made between spectra collected in the field and under controlled laboratory conditions. A preliminary analysis of 1350 spectra from 150 leaves revealed no significant statistical differences between environments, supporting the use of laboratory settings for calibration. Subsequently, spectral profiles were obtained from two sets of 70 leaves each-originating from infected and healthy branches of olive trees growing in the same field within an infected area. Infection status was confirmed via qPCR using the Harper protocol, with DNA extracted from individual leaves. For each leaf, three spectra were recorded from the basal, central, and apical regions to determine the most responsive area to infection-induced spectral variation. Data were analyzed using a Random Forest classifier to identify key wavelengths associated with infection. The portability and resolution of the spectroradiometer, combined with the results obtained under controlled conditions, suggest that this non-destructive approach can be adapted for rapid field screening of trees in quarantine zones.

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Field-deployable molecular diagnostic platform for *Xylella fastidiosa* detection. A. PUCA, A.L. PEPORI, F. PECORI, S. TORRE, F. SEBASTIANI, A. SANTINI, N. LUCHI. National Research Council, Institute for Sustainable Plant Protection (CNR-IPSP), Via Madonna del Piano 10, 50127 Sesto Fiorentino, FI, Italy. E-MAIL: alesandropuca@cnr.it

Xylella fastidiosa (Xf) is a quarantine pathogen threatening several plants of economic and social importance in Europe. Accurate surveillance of Xf is crucial for planning phytosanitary measures to eradicate or contain outbreaks. Given the numerous potential hosts and the possible lack of visible symptoms, effective surveillance requires Point-of-Care (POC) evaluations using fast, sensitive, and specific tests. To this aim, we tested a new pipeline using the portable bCUBE device (Hyris, London, UK) for real-time PCR (qPCR) and isothermal amplification (LAMP) to detect Xf in naturally infected plants. We validated the qPCR and LAMP bCUBE systems using *in vitro* assays with DNA extracted from pure cultures, and compared them to standard labora-

tory instruments. No statistical differences were found in qPCR Ct values. For LAMP assays, the bCUBE was slightly faster and more effective in detecting low DNA concentrations of the target *Xf*. Our workflow could enhance POC monitoring, leading to better and faster data collection and aiding in rapid response for *Xf* containment.

This research was financially supported by the Ministero dell'agricoltura, della sovranità alimentare e delle foreste through the Research Project 1LIVEXYLELLA - ID01 - Trasmissione D.M. n. 664519 del 28/12/2022 (Tecnologie portatili e protocolli innovativi per la diagnosi ultrasensibile di Xylella fastidiosa direttamente in piante e vettori).

Evaluation of olive genotypes response to quick decline syndrome caused by *Xylella fastidiosa* subsp. *pauca* in the Apulian outbreak zone. A.M. SAVOIA¹, M. CARLUCCI¹, P.G. LUCCHESI¹, V. FANELLI¹, I. MAS-CIO¹, F.L. AURELIO¹, M.M. MIAZZI¹, A. PACIFICO¹, C. MONTEMURRO^{1,2}, F. NIGRO¹. ¹Department of Soil, Plant and Food Sciences (DiSSPA), Via Giovanni Amendola 165/A, University of Bari Aldo Moro, Bari, 70126, Italy. ²Spin Off Sinagri s.r.l., Via Giovanni Amendola 165/A, University of Bari Aldo Moro, Bari, 70126, Italy. E-MAIL: michele.savoia@uniba.it

Xylella fastidiosa subsp. *pauca* ST53 (*Xfp*) is a Gram-negative bacterium responsible for olive quick decline syndrome (OQDS). *Xfp* is a highly destructive pathogen transmitted by xylem-sap feeding insects. Currently, no effective curative treatments are available. One of the most significant advances in combating OQDS has been the identification of olive cultivars exhibiting resistance to *Xfp* and the subsequent replanting with resistant genotypes. In this context, the aim of this study was to identify new resistant/tolerant genotypes (RTG) to *Xfp* and to investigate their genetic relationships with a broad panel of 244 cultivars from the Mediterranean region. Sixteen RTG olive genotypes were monitored over six years to assess symptom severity and bacterial load. A set of nine SSR markers was used to analyze genetic relationships between the RTG genotypes and widely cultivated Mediterranean varieties, enabling synonym identification (via LRM analysis) and phylogenetic clustering (using Unweighted Neighbor-Joining analysis). Additionally, paternity tests were performed to determine the parentage of unknown genotypes. Data on bacterial population dynamics and symptom expression revealed considerable variability among genotypes, indicating different levels of susceptibility and potential resilience. Phylogenetic analy-

sis grouped most RTG genotypes near the resistant cultivars 'Leccino' and 'FS17', suggesting significant genetic affinity. Some RTG genotypes also showed close genetic relationships with foreign cultivars such as the Albanian 'Kalinjot' and the Greek 'Leucocarpa', as well as with local Apulian cultivars. These findings underscore the potential of both local and broader Mediterranean olive germplasm as valuable sources of resistance to *Xfp*.

This research was financially supported by RIGENERA (Approcci IntegRati per il mIglioramento GENetico, la selezione e l'ottenimento di materiali vegetali Resistenti a Xylella fastidiosa) (CUP: H93C22000750001); NOVIXGEN (Nuove prospettive di sviluppo per l'Olivicoltura italiana attraverso la valorizzazione della biodiversità e la selezione di materiale GENetico d'olivo tollerante/resistente a Xylella fastidiosa e azioni mirate a prevenire il possibile impatto sulla Viticoltura (CUP: C83C22001280006).

INTEGROLIV: an integrated and eco-friendly approach for containing *Xylella fastidiosa* and regenerating the olive growing systems. G. LIMA¹, R. CASTORIA¹, F. DE CURTIS¹, D. PALMIERI¹, G. IANIRI¹, S. MARINO¹, A. PASTORINI¹, C. ALTOMARE², P. SALDARELLI³, C. DEL GROSSO³, M. MORELLI³, F. CARAGLIA³, A.M. MINCUZZI³, P. DE BELLIS², G. BLEVE², A. GALLO², M. CIUFFO³, F. LIMONGELLI², A.M. TARANTINI², A.M. SALZANO⁴, C. D'AMBROSIO⁴, G. RENZONE⁴, F. NIGRO⁵, F. FARETRA⁵, A. IPPOLITO⁵, S. POLLASTRO⁵, R.M. DE MICCOLIS⁵, O. POTERE⁵, M. MOUROU⁵, R. MARRA⁶, S.L. WOO⁷, V. LANZOTTI⁶, N. LOMBARDI, M. SINNO⁷, C. DONGIOVANNI⁸, M. DI CAROLO⁸, M. TAGLIENTE⁸, M. CARRIERI⁸, D. PERRELLI⁸, M. CASETTA⁸, R. ARGENTIERI⁸. ¹Department of Agricultural, Environmental and Food Sciences, University of Molise, 86100 Campobasso, Italy, ²Institute of Food Production Sciences, National Research Council (CNR), 70126 Bari, Italy, ³Institute for Sustainable Plant Protection, National Research Council (CNR), 70126 Bari, Italy, ⁴Institute for Animal Production System in Mediterranean Environment, National Research Council (CNR), 80055 Portici (NA), Italy, ⁵Department of Soil, Plant and Food Sciences, University of Bari "Aldo Moro", 70126 Bari, Italy, ⁶Department of Agricultural Sciences, University of Naples "Federico II", 80055 Portici (NA), Italy, ⁷Department of Pharmacy, University of Naples "Federico II", 80131 Napoli, Italy, ⁸Research, Experimentation and Training Center in Agriculture "Basile Caramia", 70010 Locorotondo (BA), Italy, E-MAIL: lima@unimol.it

The spread of *Xylella fastidiosa* subsp. *pauca* ST53, responsible for Olive Quick Decline Syndrome (OQDS),

has severely damaged olive cultivation in Salento (Southern Italy), causing dramatic economic and environmental losses. Although the implementation of containment measures reduced the rate of spread of the pathogen, the request of a control tools for infected plants is still urgent, particularly to preserve centuries-old native olive trees, which shape the Apulian landscape. To date, replanting with resistant cultivars remains the most effective strategy to promote the recovery in affected areas. The INTEGROLIV project (D.M. MASAF No. 664829 of 29/12/2022) addresses this crucial challenge by promoting a multidisciplinary and eco-friendly strategy. The project aims to identify, evaluate, and transfer innovative solutions to mitigate the impact of *Xylella fastidiosa* in infected areas. Selected antibacterial products are being tested *in vitro*, in greenhouse, and in the field to be successively included into new sustainable management protocols, targeting both the pathogen and its main vector, *Philaeus spumarius*. Advanced biochemical and molecular-genetic techniques are also employed to investigate the direct and indirect modes of action of the most promising products, with the goal of optimizing their effectiveness and ensuring environmental and economic sustainability. By selecting and validating novel strategies, INTEGROLIV aims to support olive grove regeneration, improve the resilience of traditional cultivars, ensure a sustainable future for olive growing in areas affected by *Xylella fastidiosa* and contribute to the long-term conservation of the Mediterranean olive agroecosystem.

This research was financially supported by INTEGROLIV project "Integrated Eco-friendly Approach for the Containment of Xylella fastidiosa and for the Regeneration of Olive Growing and the Environment (Project Code H33C22000860001) - Minister of Agriculture, Food Sovereignty and Forests Italy (MASAF), D.M. n. 664829 del 29/12/2022 – Research Topic 2: Investigations and tests to identify methods of control of Xylella fastidiosa.

From Risk to Response: The BeXyl Project's Integrated Approach to *Xylella fastidiosa* in Europe. B.B. LANDA, THE BEXYL CONSORTIUM. *Instituto de Agricultura Sostenible (IAS), Consejo Superior de Investigaciones Científicas (CSIC), 14004 Córdoba, Spain.* E-MAIL: blanca.landa@csic.es

The BeXyl project is a Horizon Europe-funded multi-actor initiative aimed at strengthening research and innovation around the priority quarantine pathogen *Xylella fastidiosa* (Xf). By offering interdisciplinary, stakeholder-driven solutions, BeXyl addresses three main

objectives: (i) enhancing capacities for prevention, monitoring, and response to Xf; (ii) improving control measures, especially through biological approaches; and (iii) providing scientific support for EU and associated countries' plant health policies. Aligned within the EU Plant Health Law, BeXyl focuses on reinforcing the two fundamental pillars of plant protection: prevention and control. The project assesses the risk of future Xf outbreaks under current and climate change scenarios (WP1), improves early detection through enhanced surveillance and border inspection (WP2), and ensures phytosanitary standards in plant trade via thermal treatments (WP3). Restoration of affected areas is supported by the development of resistant host plant varieties (WP4). In parallel, the project advances innovative biological control strategies targeting insect vectors (WP5) and *X. fastidiosa* itself (WP6). These efforts are integrated into decision-support tools and protocols tailored for both conventional and organic production systems (WP7). Furthermore, BeXyl integrates ecological and socioeconomic factors (WP8) to support the effective adoption of management strategies. Through dedicated efforts in research, innovation, and knowledge transfer (WP9), the project contributes to the EU Green Deal by supporting the transition toward sustainable, resilient agriculture and forestry. BeXyl ultimately aims to improve crisis preparedness, help prevent new Xf outbreaks, and mitigate the spread and impact of current infestations across Europe.

BeXyl Project is financed by HORIZON-CL6-2021-FARM-2FORK-01 (Grant 101060593).

Bacterial Communities of *Philaeus tessellatus*. S. BOUHACHEM^{1,2}, C. NACCACH³, R. ABOU KUBAA⁴, M. MEZGHANI³. ¹*Institut National de la Recherche Agronomique de Tunisie, Rue Hedi Karray, University of Carthage, Tunis, Tunisia.* ²*GVRF Laboratoy INRGREF,* ³*Faculté des Sciences de Tunis, Université El Manar, Tunisia,* ⁴*Department of Plant Pathology, University of California, Davis, CA, USA.* E-MAIL: bouhachem.sonia@inrat.ucar.tn/bouhachems@gmail.com

Philaeus tessellatus is a potential vector of *Xylella fastidiosa*, a highly dangerous bacterium that poses a serious threat to olive trees and other fruit crops in the Mediterranean region. Given the growing interest in microbiota-mediated vector control strategies, microorganisms associated with insects are being explored as alternative biocontrol agents due to their ability to influence host-parasite interactions. To investigate the composition and structure of the symbiotic microbiota in *P. tessellatus*, we collected ten adult specimens from each of five

host plants—*Rumex crispus*, *Cirsium arvense*, *Glebionis segetum*, *Smyrniolus olusatrum*, and *Daucus carota*—in the Cap Bon region of Tunisia. The microbial community was then analyzed using next-generation sequencing (NGS) of 16S rRNA gene amplicons. Amplicon Sequence Variants (ASVs) were classified into a total of 17 phyla, 34 classes, 82 orders, 114 families, 195 genera, and 74 species. Among these, 44 ASVs were consistently detected across all samples and considered as core microbiota, representing a substantial fraction of the total microbial community. The bacteriome of *P. tessellatus* comprised 20 distinct bacterial phyla, with *Proteobacteria*, *Cyanobacteria*, *Bacteroidota*, *Actinobacteriota*, *Bdellovibrionota*, and *Firmicutes* being the most abundant. The most well-represented bacterial orders include *Pseudomonadales*, Chloroplast (derived from symbiotic plant-associated bacteria), *Rhizobiales*, *Burkholderiales*, *Rickettsiales*, *Sphingomonadales*, *Sphingobacteriales*, *Flavobacteriales*, and *Enterobacteriales*. Notably, key symbionts were found primarily within the orders *Rickettsiales* (*Wolbachia*, *Rickettsia*, *Candidatus Megaira*), *Sphingobacteriales*, *Enterobacteriales* (*Rosenbergiella*), *Flavobacteriales*, and *Burkholderiales* (*Duganella*), with corresponding genera/species identified. Significant variation in endosymbiotic bacterial communities was observed across specimens collected from different host plant, highlighting the crucial role of host plant species and environmental conditions on microbial community composition. These findings provide valuable insights into the ecological of *P. tessellatus*, suggesting that host-associated microbial diversity may play a key role in the insect's biology and potential as a disease vector.

CONCURRENT SESSION B1— Plant viruses and phytoplasma diseases in a changing climate

SESSION KEYNOTES

Success and challenges: the new diagnostic era using High Throughput Sequencing. M. AL RWAHNIH. Department of Plant Pathology, Foundation Plant Services, University of California, Davis, USA. E-MAIL: malrwahnih@ucdavis.edu

Foundation Plant Services (FPS), located in Davis, California, USA, is a source of elite propagation materials of grape, Prunus, pistachio, olive, strawberry, rose and sweetpotato cultivars as a member of the US National Clean Plant Network (NCPN). NCPN clean plant centers such as FPS are responsible for conducting diagnos-

tic testing, therapeutics, and propagation, and distribution of certain vegetatively propagated speciality crops. FPS also facilitates the introduction, quarantine, testing and release of material under a Controlled Import Permit from the United States Department of Agriculture, Animal Plant Health Inspection Service, Plant Protection and Quarantine (USDA- APHIS-PPQ). When FPS was established in 1958, conventional techniques for detection of plant viruses relied on biological indexing using indicator plants. Diagnostic methods advanced to immunodetection (like ELISA) and then polymerase-chain reaction (PCR) testing, though indexing was still considered the “gold standard”. FPS has pioneered the use of high throughput sequencing (HTS) as a diagnostic method, which is now approved by USDA-APHIS-PPQ for release of plant material from quarantine and by California Department of Food and Agriculture for use in grape and fruit tree certification programs. HTS for virus detection and characterization in plants was first used at FPS in 2007 and compared to biological indexing and other conventional methods beginning in 2012. After demonstrating that HTS was more effective at detecting virus infection than biological indexing, FPS validated its protocols and took part in an interlaboratory validation study to demonstrate reproducibility across labs. After this rigorous validation of HTS protocols, FPS received approval to utilize this technology on regulated grape and fruit tree plant materials in 2021. To date, the USA is one of only a few countries who have adopted HTS in quarantine and certification programs, although other countries have been evaluating the technology and the academic reference literature continues to grow. HTS adoption has been slow, primarily due to challenges in standardizing preparation and evaluation methods. Human error caused by fatigue due to repetitive processes can be alleviated by using automated sample processing equipment, which allows for high sample throughput and consistent results. A laboratory utilizing HTS must have a skilled bioinformatician to align and analyze the complex sequence data that is generated. A trained virologist must review the data provided by the bioinformatician and consider whether viruses detected are evidence of real infections, or if they are artifacts of cross-contamination or non-host contribution to the sample. Using HTS, new viruses are being discovered at an unprecedented rate. It is important that when a new virus is detected when testing plant material using HTS, detection is confirmed with a second testing method such as PCR, graft transmissibility is demonstrated, and scientists remain in regular and clear communication with regulatory agencies to assess the threat to plant health.

ORAL PRESENTATIONS

Comparative analysis of Stolbur Phytoplasma genomes highlights key genomic features of the novel 16SrXII-P Stolbur pathogen.

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Stolbur phytoplasmas (Mollicutes) are a heterogeneous group of insect-transmitted phytopathogenic bacteria that cause several diseases in crops including grapevine, potato, tomato, corn, and sugar beet. Since the 1990s, it has been known that stolbur phytoplasmas are also present in sugar beets diseased by syndrome “basses richesses”, caused by the ‘*Candidatus Arsenophonus phytopathogen*’ (γ-Proteobacteria). Both pathogens are transmitted to sugar beet and other crops by the cixiid *Pentastiridius leporinus* L. The ongoing epidemic in Germany is driven by a novel, poorly characterised phytoplasma of the 16SrXII-P subgroup. We determined and analysed the complete genome of the 16SrXII-P phytoplasma strain GOE from a metagenomic high-molecular-weight DNA template obtained from infected *P. leporinus* individuals using single-molecule real-time sequencing (PacBio). The metagenomic dataset was filtered for phytoplasma reads via taxonomic binning. *De novo* assembly was performed on the selected reads. The circular GOE chromosome was compared with other stolbur genomes regarding phylogeny, genome organization, encoded metabolism and virulence factors. The results suggest that 16SrXII-P phytoplasmas represent a new phytoplasma species. GOE is characterised by the smallest size and lowest G+C content compared to other complete stolbur genomes. While stolbur phytoplasmas share conserved membrane proteins and minimal metabolism of phytoplasmas, they differ in encoding a riboflavin kinase, suggesting a lost pathway outside the 16SrXII and 16SrXIII group. In addition, virulence factor analysis showed that GOE encodes a complete *tra5*-transposon forming a phytoplasma-pathogenicity island since it harbours important effector proteins. These findings emphasise the diversity within the 16SrXII group and improve the understanding of phytoplasma evolution.

This research is part of the project “Differentiation of pathogens and course of infection in SBR-associated bacterioses in sugar

beet to derive resistance testing methods to ensure yield stability” which is coordinated by the “Gemeinschaft zur Förderung von Pflanzeninnovation e. V.” (GFPI) and founded by the Federal Ministry of Economic Affairs and Climate Protection (BMWK) (funding no. 22943 N).

‘Candidatus Phytoplasma phoenicium’, a causal agent of stone fruits diseases and a target for surveillance.

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A disease affecting almond trees, characterized by symptoms such as small yellowish leaves, shoot proliferation, and dieback, was first observed in Lebanon during the 1990s. In 2001 it was reported as being associated with phytoplasmas belonging to the pigeon pea witches'-broom (PPWB) group (16SrIX). In 2002 other authors carried out a survey in Lebanon showing the association of Almond Witches'-broom disease (AlmWB) with a phytoplasma from a newly subgroup 16SrIX-B, later classified as ‘*Candidatus Phytoplasma phoenicium*’ and detected in both Lebanon and Iran. From 2010 to 2015, a scientific cooperation project between Lebanese and Italian institutions was funded with the aim to investigate the host range of the pathogen, the geographic distribution of the disease, and the identification of potential insect vectors. These studies revealed that the epidemiological cycle of AlmWB involves *Asymmetrasca decedens* (Hemiptera: Cicadellidae), possibly responsible for almond-to-almond transmission of ‘*Ca. P. phoenicium*’. In addition, two species of *Tachycixius*, *T. viperinus* and *T. cf. cypricus* (Hemiptera, Cixiidae), were identified as potential vectors responsible for transmitting the phytoplasma from weed hosts to almond trees. Further studies, including draft genome sequencing, demonstrated that ‘*Ca. P. phoenicium*’ (i) infects almond, peach, nectarine, and apricot in both Lebanon and Iran; (ii) the genome encodes membrane proteins and effector-like

proteins that may play a role in host-pathogen interactions and pathogenicity. In 2017, the European and Mediterranean Plant Protection Organization (EPPO) published a pest risk analysis of 'Ca. P. phoenicium', and the EUPHRESKO network supported the project "DIP-CAPP" (Development of reliable protocols for the detection and identification of 'Ca. P. phoenicium').

Insights into Wisteria vein mosaic virus: an evolutionary framework for understanding the emergence of new viral threats. M. MORELLI, G. D'ATTOMA, P. SILDARELLI, A. MINAFRA. CNR, *Institute for Sustainable Plant Protection (IPSP), Via Amendola 122/D, 70126 Bari, Italy.* E-MAIL: massimiliano.morelli@cnr.it

Wisteria vein mosaic virus (WVMV, *Potyvirus wisteriae*), a member of the *Potyvirus* genus, is the causative agent of Wisteria vein mosaic disease (WMD), a severe infection threatening wisteria plants, ornamentals highly valued worldwide. Likely originating in East Asia, WVMV was first officially documented in the United States, and subsequently reported in multiple countries. In Italy, the virus was first identified as the WVMV Bari isolate, and its complete genome was sequenced using NGS barcoding technology. Phylogenetic analysis via PhyML, corroborated by clustering algorithms, revealed a distinct division into two phylogenetic groups. The dominant clade included isolates from *Wisteria* spp., while the minor clade comprised three genetically divergent strains, with sequence identity close to the species boundary, infecting herbaceous non-wisteria hosts. Molecular clock estimates, based on the Relative Time Dated Tips (RTDT) approach, placed the divergence of these lineages around the 17th century. Network inference further validated the genetic separation between the two host-related groups and indicated the potential existence of intermediate variants occurring in the host jump transition. Population genetic analyses demonstrated a pronounced genetic divergence, reinforced by significant permutation tests. The fixation index (*FST*) and migrant number (*Nm*) indicators pointed to limited gene flow and well-structured populations. Negative neutrality tests and a low dN/dS ratio suggested that purifying selection is actively removing deleterious mutations. This study proposes an evolution-based framework approach, using the intriguing evolutionary dynamics of WVMV as a case study to explore how other overlooked potyviruses or newly emerging viruses could spread and infect economically important crops.

RNA-Seq transcriptomic analysis reveals genetic mechanisms enhancing virus tolerance in grafted *Cucumis melo* onto *C. melo* cv. Barattiere. M. CRUDELE, M. MARASHI, R.M. DE MICCOLIS ANGE-LINI, M. MASTROCHIRICO, T. MASCIA. *Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, Bari, Italy.* E-MAIL: marco.crudele@uniba.it

Tomato leaf curl New Delhi virus is an emerging begomovirus listed in the EPPO Alert-list 2, first reported in 2015 in Southern Italy on Sicilian courgette, and particularly harmful to cucurbit crops, often leading to complete production losses. RNA-Seq analyses of *C. melo* cv. Retato Standard F1 (RSF1) (susceptible), not grafted and grafted onto *C. melo* cv. Barattiere (B) (tolerant), was performed to study mechanisms conferring tolerance in grafted plants. A total of 189,419,643 Illumina short reads (2 x 150 bp) were generated and mapped to the *C. melo* reference genome. By comparing grafted vs ungrafted and inoculated vs mock-inoculated plants, a total of 7,962 differentially expressed genes (DEGs) ($FC \geq |2|$, $FDR \leq 0.05$) were identified. ToLCNDV infection in grafted plants strongly induced the expression of genes involved in defense mechanisms (e.g., pathogen-induced protein CuPi1 and acidic endochitinase precursor) and the activation of 798 genes involved in signal reception and transduction, response to pathogen (e.g., Major Latex Protein 28 and 149-like isoform X1) and stress factors (e.g., protein RADIALIS-like 1 and 4). DEGs activated by grafting included genes exclusively or more strongly induced after virus infection. Among them, several genes were involved in stress response and oxidative stress (e.g., protein RADIALIS-like 1 and glutaredoxin C11 and C13) and in the biosynthesis of volatile terpenoids and aromatic compounds (e.g., nerolidol synthase 1-like and benzyl alcohol O-benzoyltransferase), respectively. This study provides insights genetic mechanisms underlying virus tolerance in RSF1 plants grafted onto B plants and into molecular interactions between ToLCNDV and *C. melo*.

This research was financially supported by National Recovery and Resilience Plan, Mission 4, Component 2: "From Research to Business" – Investment 3.3. This study was carried out within the Regione Puglia Administration under Rural Development Program 2014–2020, Project 'Biodiversity of Apulian fruit vegetables (BiodiverSO Karpos)', Measure 10, Sub measure 10.2, Operation 1 "Program for the conservation and the valorization of the genetic resources in agriculture" (DDS n. 04250178565) and Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032

17/06/2022, CN00000022).

Biological characterization of beet western yellows virus-pepper isolate. N. BUZKAN¹, B.B. ARPACI², C. VILLEROY³, V. ZIEGLER-GRAFF⁴, V. BRAULT³. ¹Plant Protection Department, Agriculture Faculty, Kahramanmaraş Sütçü Imam University, Kahramanmaraş, Türkiye. ²Horticulture Department, Agriculture Faculty, Kilis Yedi Aralık University, Kilis, Türkiye. ³INRAE, Colmar, France. ⁴CNRS-IBMP, Strasbourg, France. E-MAIL: nbuzkan@ksu.edu.tr; nbuzkan@gmail.com

Beet western yellows virus (BWYV) infecting over 150 plant species from 23 dicotyledonous families is the most well-known member of the genus *Polerovirus* in the family *Solemoviridae*. An isolate of this virus was first described in Turkey, which represents the first polerovirus reported to infect cultivated pepper naturally and to cause a severe disease. This polerovirus displays sequence similarities with BWYV and is distantly related to the Chinese and American isolates from sugar beet and lettuce. This study investigated the host range of BWYV- pepper and -sugar beet isolates. Whole peppers collected from symptomatic plants were used as virus source in aphid (*Myzus persicae* Sulzer) transmission assays onto healthy peppers. Virus purification was performed from peppers to use as inoculum source in aphid transmission tests while the purified BWYV-sugar beet was already available in stock. Non viruliferous aphids (ca. 200 individuals) were fed on an artificial medium containing the purified virus preparations through stretched parafilm for 24 hours. Then, groups of 10 aphids were transferred onto healthy sugar beet (*Beta vulgaris*), pepper (*Capsicum annuum*), *Claytonia* spp., spinach (*Spinacia oleracea*) and pea (*Pisum sativum*) plants at 2- to 4-true leaf stage for 6 days. Virus presence was checked by ELISA using a turnip yellows virus (polerovirus) antiserum combined with molecular detection. Results showed that BWYV-pepper and -sugar beet isolates could not infect sugar beet and pepper respectively, although both isolates were successfully transmitted onto other hosts in the experiments.

This research was granted by TUBITAK- FRANCE PHC BOSPHEUS (221 N 050).

Biological characterization of tomato fruit blotch virus (genus *Blunervirus*, family *Kitaviridae*) a “HTS-borne” plant virus. A. TIBERINI¹, A. SYBILSKA², M. LUIGI¹, F. TARCHI³, A. TAGLIENTI¹, D. LUISON¹, F. FAGGIOLI¹, S. SIMONI³, M. LEWANDOWSKI², S.

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High-throughput sequencing (HTS) is a powerful tool for detecting and identifying viruses in plant hosts, regardless of their genome nature or structure. HTS also provides insights into virus population structure, ecology, and evolution. Despite the exponential growth of HTS data, many newly identified viruses remain biologically uncharacterized, and their epidemiological significance is unassessed. The Euphresco project “Valorisation of HTS output data in view of a timely risk assessment of regulated or emerging plant viruses (VALORHIGHTS)”, recently started, aims to provide biological information to support the evaluation of phytosanitary risks posed by novel or poorly characterized viruses. Tomato fruit blotch virus (ToFBV - *Blunervirus solani*; genus *Blunervirus*, family *Kitaviridae*) is a recently identified virus causing uneven ripening and dark spots on tomato fruits. First identified by HTS in 2018 in Italy, ToFBV shows a recurrent emergence trend and is included in the EPPO Alert list. Recent experiments demonstrated the virus’s replication and translocation within the host plant and identified potential weed hosts. Additionally, HTS data mining of existing RNAseq datasets from ToFBV-infected tomato samples suggested a mite as a potential vector. The sequences retrieved were taxonomically assigned to the tomato russet mite (TRM) *Aculops lycopersici* (Tyron). The presence of this mite was confirmed at virus outbreak sites. Two independent transmission trials unequivocally identified the mite as the vector of ToFBV. These findings provide new insights into the potential spread of ToFBV and inform the development of effective control and surveillance strategies.

Witches’ broom disease of lime in the Arabian Peninsula: a multiscale study of pathogen dynamics, climate influence, and trade-driven spread. A.M. AL-SADI^{1,2}, A.M. AL-SUBHI², A.G. AL-GHAITHI², R.A. AL-YAHYAI². ¹College of Agriculture, University of Al Dhaid, Sharjah, P.O. Box 27272, United Arab Emirates. ²Department of Plant Sciences, Sultan Qaboos University, P.O. Box 34, Al Khod 123, Sultanate of Oman. E-MAIL: alsadi@uodh.ac.ae

Witches' Broom Disease of Lime (WBDL), caused by '*Candidatus* Phytoplasma aurantifolia', has led to severe losses in lime production across the Middle East. In our study, we investigated the molecular, ecological, and environmental factors influencing the spread and severity of WBDL, with a particular focus on the Arabian Peninsula. Through field surveys conducted across 18 districts in Oman and the UAE, we documented widespread phytoplasma infection, with symptom severity varying by climate. Witches' broom symptoms were most pronounced in subtropical areas, while trees in arid and semi-tropical zones showed milder symptoms despite harboring the pathogen. Molecular analysis revealed significantly higher phytoplasma titers in symptomatic leaves compared to asymptomatic ones. We detected elevated expression of the phytoplasma effector gene *SAP11WBDL*, which correlates with the down-regulation of plant defense-related pathways, particularly those involving jasmonate and salicylic acid signaling. Our findings confirm that *SAP11* interferes with TCP transcription factors, promoting excessive shoot proliferation, which in turn increases phytoplasma accumulation and enhances transmission by the leafhopper vector *Hishimonus phycitis*. In parallel, we examined historical trade patterns and phytosanitary practices in the region. The results indicate that the introduction and spread of WBDL were facilitated by increased international trade and the movement of infected plant material and insect vectors. Our study highlights the urgent need for enhanced biosecurity measures, including stricter quarantine protocols and coordinated regional surveillance, to mitigate the further spread of WBDL and protect citrus production systems in the Middle East.

Development of turn-back loop-mediated isothermal amplification assay for the detection of Tomato fruit blotch virus. S. Y. ATEŞ¹, B. ÖZGÖREN². ¹Ministry of Agriculture and Forestry, Izmir Agricultural Quarantine Directorate 35230 Izmir, Türkiye. ²DOSA Bilgi ve Bilim Teknolojileri Ltd.Şti., Ege Üniversitesi Teknopark 35100 Izmir, Türkiye. E-MAIL: songulyalcinates@outlook.com

Tomato fruit blotch virus (ToFBV-*Blunervirus solani*) is a new emerging virus in tomato and was added to the EPPO's alert list as of 2024-01. As it is a new pathogen, its epidemiology and rate of spread are still unknown. Currently, ToFBV is only detected by RT-PCR which is time consuming. In this study, a novel turn-back loop primer-accelerated LAMP (TLAMP), for reliable and sensitive detection of the virus directly from field samples

at quarantine laboratories, was developed. Basic Local Alignment Search Tool (BLAST), Clustal Omega and PrimerExplorer v5 were used to design primers targeting conserved regions of RNA3 and RNA4, enabling the detection of a wide range of ToFBV isolates. They were successfully used for colorimetric TLAMP and fluorometric TLAMP methods in several laboratory assays. Additionally, BLAST and laboratory studies showed that there was no cross-reaction with other viruses such as tomato brown rugose fruit virus and tomato spotted wilt virus, commonly infecting tomato plants.

POSTER PRESENTATIONS

Detection of grapevine virus A in grapevine in Tekirdağ province of Türkiye. S.D. NOGAYLAR, H. İLBAGI. Department of Plant Protection, Faculty of Agriculture, Tekirdağ Namık Kemal University, 59030, Tekirdağ, Türkiye. E-MAIL: hilbagi@nku.edu.tr

Grapevine virus A (GVA) is a type of member of the genus *Vitivirus*, family *Betaflexiviridae*. It is associated with the Kober stem grooving disease of the Rugose wood complex (RW) and is transmitted by grafting, mealybugs, and scale insects. The virus is among the most frequently identified viruses in vineyards worldwide; however, no comprehensive study has been associated with GVA in Tekirdağ province to determine its prevalence. In this study, extensive surveys were conducted to investigate the presence and prevalence of GVA at the end of summer 2024 in the vineyards of Tekirdağ province, Trakya, which has the largest germplasm and collection vineyard in Turkey. The surveys involved the collection of a total of 139 symptomatic and asymptomatic leaf samples from autochthonous and foreign grapevine cultivars. Leaf tissues from all samples were screened using DAS-ELISA, and then subjected to amplify a 429 bp using a specific primer of GVA-coat protein (CP) gene by RT-PCR assay. None of the autochthonous and foreign grapevine cultivars, such as 'Semillion', 'Merlot', 'Gamay', 'Hamburg Misketi', 'Alphonso Lavalle', 'Cinsaut', 'Adakarası', 'İlkeren', 'Cabernet Sauvignon' were found to be infected with GVA. However, both DAS-ELISA and RT-PCR assays revealed that only 'Yapıncak' autochthonous cultivar showed a positive reaction in 9 out of 139 leaf samples. As a result of this study, the infection rate of GVA was found to be 6.47% in the commercial vineyards of Tekirdağ province of Trakya, Turkey.

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Investigation of maize dwarf mosaic virus in maize and common reed in Türkiye. H. İLBAĞI, N. ÇETİN. *Department of Plant Protection, Faculty of Agriculture, Tekirdağ Namık Kemal University, 59030, Tekirdağ, Türkiye.* E-MAIL: hilbagi@nku.edu.tr

Maize (*Zea mays* L.) is one of the most important cereals for human and animal consumption and is grown for grain, forage, and silage in the Trakya region of Türkiye. Viral diseases negatively affect maize production yearly by causing significant yield loss in maize fields worldwide. Among maize-infecting viruses, maize dwarf mosaic virus (MDMV) is one of the economically important viruses that cause yield loss in maize. Our study aimed to determine the presence of maize dwarf mosaic virus in both maize and common reed. By this aim, 34 maize and 19 reed samples were collected from Tekirdağ province of Trakya in 2023. Leaf tissues from symptomatic and asymptomatic plants were first screened for the presence of maize dwarf mosaic virus (MDMV) by the DAS-ELISA test. Subsequently, all leaf samples were tested to amplify a 720 bp using the primer pair designated from the Nib gene region of the MDMV genome. The ELISA and RT-PCR results revealed that 7 out of 34 maize and 10 out of 19 reeds were found to be infected with MDMV. PCR amplicons were subsequently sequenced and subjected to a BLASTn search to identify MDMV sequences. Multiple nucleotide sequence alignments of Turkish maize and reed MDMV isolates revealed that the highest level of identity was 85.37–76.77% and 86.36–76.77% with Golestan and SP isolates from Iran and Spain, respectively. Amino acid multiple sequence alignments of Turkish maize and reed isolates with other published MDMV isolates revealed that the highest level of identity was 95.93–66.66% with TR34, TekirdagE41, TekirdagK43, and TekirdagT42 isolates from Türkiye. The nucleotide-based phylogenetic tree clustered with MDMV isolates from Türkiye. The amino acid-based phylogenetic tree clustered with MDMV isolates from Türkiye, Germany, Hungary, Bulgaria, and Iran.

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Heat shock proteins 70 in viruses: diversity and evolution. A. MAACHI. *Regional Center of Agricultural Research of Agadir, National Institute of Agricultural Research (INRA), Inezgane, 86350, Agadir, Morocco.*

The heat shock proteins 70 (Hsp70) are a family of conserved ubiquitously proteins, reported from all living organisms. They play a key role in performing chaperoning functions, helping to protect cells from adverse effects of physiological stress. Hsp70 consists of two high conserved domains, a nucleotide binding domain at the N-terminal, and a substrate binding domain at the C-terminal. In viruses, hsp70 were previously reported from closteroviruses, however, little is known on their abundance, diversity, and evolution. In this work, we searched for hsp70s from virus protein database from the National Center for Biotechnology Information. Hsp70s retrieved belonged mainly to viruses from two different classes: Alsurivicetes (viruses reported to infect plants) and Megaviricetes (Giant viruses reported to infect protists). In the megavirids, multiple copies of hsp70 were recovered from theHir genomes suggesting either gene duplication or multiple acquisition events. A correspondence analysis based on the similarity matrix among these proteins showed two different clusters, one grouped hsp70s from giant viruses, and the other, grouping the hsp70 from plant viruses, suggesting different evolutionary origins of these proteins. The relationship among virus hsp70s were uncovered based on the phylogenetic trees, showing that the hsp70 from plant viruses exhibited more diversification, suggesting higher evolutionary rates, and higher genetic diversity in comparison to the ones from megaviricids. Our results show that the hsp70 presence is restricted in viruses, suggesting the occurrence of few gene transfer events of these proteins between viruses and their hosts.

Sanitary status of onion (*Allium cepa*) in Tunisia. S. NAHDI^{1,2}, W. ABIDI¹, B. M'RABET SAMAAALI³, M. BRAHAM³, S. MATIC^{4,5}. ¹*Ecole Supérieure d'Agriculture du Kef, ESAK, rte de Dahmani Boulifa 7100 Kef - KEF, Le Kef, Université de Jendouba, Jendouba, Tunisia.* ²*Laboratoire de Protection des Végétaux (LPV), INRAT, Université de Carthage, Rue Hédi Karray, 1004 El Menzah, Ariana, Tunisia.* ³*Laboratoire des Analyses Virologiques, Direction Générale de la Santé Végétale et du Contrôle des Intrants Agricoles, 30 Rue Alain Savary, Le belvédère, Tunis 1002, Tunisia.* ⁴*Institute for Sustainable Plant Protection, National Research Council of Italy (IPSP-CNR), Strada delle Cacce 73, 10135 Turin, Italy.* ⁵*Department of Agricultural, Food and Forest Sciences (SAAF), University of Palermo, Viale delle Scienze, Ed. 5, 90128 Palermo, Italy.* E-MAIL: nehdimah@yahoo.fr

Similar to other vegetatively propagated crops, onion

(*Allium cepa* L.) is highly vulnerable to viral infections that can accumulate over successive cycles, leading to yield and quality losses. In Tunisia, despite the crop's economic importance, little information is available about the diversity of viruses affecting onion. Until recently, only onion yellow dwarf virus (OYDV) had been officially reported, although field observations often show virus-like symptoms. To investigate this gap, a survey was conducted in spring 2023 to assess the presence of other viruses in symptomatic onion plants. A total of 111 leaf samples were collected from five major onion-producing regions (Sousse, Mahdia, Gabes, Kebili, and Cap Bon). Samples were tested using DAS-ELISA for Leek yellow stripe virus (LYSV), garlic common latent virus (GarCLV), and three allieviruses (garlic Virus A (GarV-A), garlic Virus B (GarV-B), garlic virus C (GarV-C)). Results showed that 75 out of 111 samples (67.6%) were infected with at least one virus. GarCLV was mostly prevalent (72.05%), followed by GarV-C (27%), GarV-A (21.61%), LYSV (4.5%), and GarV-B (9%). Mixed infections were more common than single infections. These findings reveal a broader viral diversity in Tunisian onions than previously reported. Further investigations involving high-throughput sequencing, vector identification, and epidemiological studies are necessary to enhance our understanding of the onion virome and to support the development of integrated and sustainable disease management strategies.

Response of Local Tomato Ecotypes from the Campania Region (Italy) to Infections by Potato virus Y (C-to strain) and Potato Spindle Tuber Viroid. L. VACCARO¹, R. MARZIALE², R. SPANÒ², D. ALIOTO³, B. NAVARRO², F. DI SERIO², T. MASCIA¹. ¹Department of Soil, Plant and Food Sciences, University of Bari "Aldo Moro", 70126 Bari, Italy. ²Institute for the Sustainable Protection of Plants, National Research Council (CNR) Bari, Italy. ³Department of Agriculture, University of Naples Federico II, 80055 Portici, Naples, Italy. E-MAIL: lorenza.vaccaro@uniba.it

Tomato (*Solanum lycopersicum*) is one of the most important solanaceous crops due to its economic and nutritional relevance. However, it is susceptible to a wide range of plant pathogens, including a recombinant strain of potato virus Y (PVY^{C-to}) and potato spindle tuber viroid (PSTVd), both of which are considered major threats to tomato cultivation. PVY^{C-to}, a single-stranded RNA virus (family *Potyviridae*), induces symptoms such as mosaic, chlorosis, and vein necrosis in horticultural crops, leading to significant production losses.

PSTVd, a nuclear-replicating viroid (family *Pospiviroidae*), elicits stunting and leaf curling in tomato plants. This study aimed to assess the susceptibility of five local tomato ecotypes from the Campania region (Corbarino, Vesuviano, Giallo determinato da Serbo, Pizzutello, and San Marzano) to PVY^{C-to} and PSTVd infections. Visual symptom assessment at 14 and 28 days post-inoculation (dpi) and quantification of virus and viroid accumulation using qDot-Blot or RT-qPCR showed that all ecotypes were susceptible to PVY^{C-to} and PSTVd infections, although the severity of symptoms and the accumulation level of the virus/viroid in the infected tissues was differed depending on the ecotype. Notably, Corbarino and San Marzano showed mild symptoms and reduced pathogen accumulation upon PVY^{C-to} and PSTVd infection, respectively, revealing a certain level of tolerance. In contrast, the ecotypes Giallo determinato da serbo and San Marzano were particularly susceptible to PSTVd and PVY^{C-to}, respectively. The local tomato ecotypes identified in this study may serve as valuable resources to further explore the molecular pathways underlying tomato defence responses against PVY^{C-to} and PSTVd infections.

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First Report of Citrus Leaf Blotch Virus on Nagami Kumquat in Turkey. B. FIDANCI-AVCI¹, N. ÖNELGE¹, A. ESKALEN². ¹Department of Plant Protection, Cukurova University, Adana, Turkey. ²Department of Plant Pathology, University of California, Davis, CA, USA. E-MAIL: aeskale@ucdavis.edu

Nagami kumquat (*Fortunella margarita*) production has expanded in Turkey due to its agronomic potential and growing market demand. In May 2022, during a survey in commercial orchards in Mersin Province, symptoms consistent with bud union disorder were observed in Nagami kumquat trees. Molecular testing of symptomatic and asymptomatic samples excluded common citrus pathogens, including Citrus psorosis virus, Citrus exocortis viroid, and Hop stunt viroid. RT-PCR assays using specific primers revealed the presence of Citrus leaf blotch virus (CLBV), with expected amplicons obtained for both coat protein (CP) and RNA-dependent RNA polymerase (RdRp) genes only in symptomatic trees.

Sequencing confirmed CLBV identity, showing 96–97% nucleotide identity with GenBank sequences. Graft-inoculation onto Etrog citron and Dweet tangor confirmed pathogenicity, producing chlorotic blotching and stem pitting symptoms. This is the first confirmed report of CLBV in citrus in Turkey. Its likely introduction through infected grafting material highlights the importance of virus-tested propagation programs. Considering climate change and increasing movement of plant material, further surveys are needed to assess CLBV's distribution and potential impact on citrus production in the region.

Dynamics of the Flavescence dorée spread in South Tyrol (northern Italy). S. OETTL¹, Y. REYES-DOMINGUEZ¹, F. PERNTNER², A. SIMONCELLI², A. GALLMETZER¹. ¹Laimburg Research Centre, Laimburg 6 – Pfatten/Vadena, 39040 Auer/Ora, BZ, Italy. ²Office for Fruit and Viticulture and Phytosanitary Service, Brennerstraße 6, 39100 Bozen/Bolzano, BZ, Italy. E-MAIL: sabine.oettl@laimburg.it

Flavescence dorée, a serious and rapidly spreading threat to European viticulture, is caused by a phytoplasma associated with the 16SrV group (FDp). Although the disease has been known in Europe and neighbouring wine regions for decades and has been monitored in South Tyrol (northern Italy) since the early 2000s, the first outbreak in the province did not occur until 2016. Since then, its prevalence has increased significantly. As part of a province-wide monitoring program led by the phytosanitary service, over 5,500 leaf samples from symptomatic grapevines have been tested for FDp. Over the past nine years, the number of FDp-positive samples has risen steadily, with nearly 30% of symptomatic grapevines testing positive in 2024. The continued spread of the disease into new areas further exacerbates the situation. Due to the high phytosanitary risk, several municipalities have been designated as infestation zones, with legally mandated control measures implemented against *Scaphoideus titanus*, the insect vector responsible for transmission. A subset of 327 FDp-positive samples collected between 2016 and 2024 was further characterized using the method of Rossi et al. (2019). All grapevine-derived samples from South Tyrol were assigned to the *dnaK1* genotype, consistent with the FD-D strain found in other Italian wine-growing regions. However, seven samples from natural FDp reservoir plants, *Clematis vitalba* and *Alnus* spp., were classified either as *dnaK2* or *dnaK3* genotypes. These findings underscore the need for continued surveillance and the implementation of effective management strategies to mitigate the spread of

this destructive disease.

Carbon dot-mediated dsRNA delivery enhances RNAi efficacy against cucumber green mottle mosaic virus in susceptible and resistant cucumber. L. VELASCO, E. MARTÍNEZ, D. JANSSEN. Instituto Andaluz de Investigación y Formación Agraria (IFAPA), 29140 Churriana, Málaga, Spain. IFAPA, 04745 La Mojonera, Almería, Spain. E-MAIL: leonardo.velasco@juntadeandalucia.es

In this study, a dsRNA-based system for controlling cucumber green mottle mosaic virus (CGMMV) in cucurbits using carbon dots (CDs) or chitosan nanoparticles (ChNPs) as delivery nanoparticles were developed. In growth room trials, two cucumber varieties we evaluated: (i) CGMMV-susceptible and (ii) partially resistant. Three formulations were tested: (1) naked dsRNAs targeting CGMMV movement (MP) and coat (CP) viral transcripts, (2) dsRNA-CDs (1:0.5 w/w), and (3) dsRNA-chitosan nanoparticles (ChNPs) (1:0.5 ratio). Water-treated (mock) and uninoculated groups served as positive and negative controls, respectively. In susceptible plants, untreated inoculated controls exhibited symptoms, being severe in 11/12 individuals. Naked dsRNA reduced symptom severity (5/12 mild, 7/12 asymptomatic) and viral load by 25%, while dsRNA-CDs achieved 9/12 asymptomatic plants with >50% viral load reduction. DsRNA-ChNPs showed limited efficacy (3/12 severe symptoms) and no significant reduction in viral titers. Resistant plants remained asymptomatic across treatments but displayed distinct viral load patterns: dsRNA-CDs and dsRNA-ChNPs reduced titers by 68% and 81%, respectively, compared to untreated resistant controls. These results demonstrate that CD-complexation enhances dsRNA stability and uptake, providing effective protection against CGMMV in susceptible cultivars. The unexpected efficacy of ChNPs in resistant plants needs further investigation in nanoparticle-plant interactions. This nanotechnology-driven approach offers a promising strategy for integrating RNAi with host resistance in cucurbit disease management.

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Breaking innate resistance: transgenic RNAi strategies against tomato brown rugose fruit virus unintentionally promote viral systemic movement in tobacco hosts. L. VELASCO, E. MARTÍNEZ, C. RANGEL, D. JANSSEN. Instituto Andaluz de Investigación

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In our investigation of RNA interference-based resistance strategies against tomato brown rugose fruit virus (ToBRFV), we engineered transgenic *Nicotiana tabacum* lines expressing hairpin-derived dsRNAs targeting the coat protein (CP) and movement protein (MP) genes. These plants are intended as potential sources of dsRNAs, siRNAs and miRNAs for topical applications. Through primer walking, we identified the insertion loci, ensuring that transgene integration didn't disrupt endogenous genes - a critical consideration given tobacco's natural resistance against this pathogen. ToBRFV-mechanical inoculation in wild-type phenotype induces necrotic local lesions and limited systemic spread, with RT-qPCR revealing low viral titers in distal tissues. Our transgenic lines, however, subverted this pattern. Contrary to anticipated enhanced resistance, CP/MP-expressing plants displayed absence of local necrosis post-inoculation, accompanied by dramatic systemic spread - distal tissue viral loads averaged 570-fold compared to non-transgenic controls. This pattern suggests our engineered constructs might interfere with tobacco's native defense signaling pathways rather than directly inhibiting viral replication. Deep sequencing of small RNAs (18-24 nt) from the transgenic plants revealed construct-specific activity patterns. In CP-targeting lines TOCP2 and TOCP4, we detected modest siRNA production (139 and 169 CP-derived siRNAs per million total sRNAs, respectively). MP-targeting lines TOMP6 and TOMP14 exhibited markedly higher siRNA generation (1,245 and 2,676 MP-derived siRNAs per million), potentially reflecting differences in hairpin structure stability or transcript processing efficiency. Remarkably, the MP lines' heightened siRNA production didn't correlate with improved viral containment, suggesting ToBRFV might employ RNAi-independent movement mechanisms in transgenic plants, or that viral suppressors neutralize RNAi effects.

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Experimental evolution of sweet potato leaf curl del-tasatellite 1 in *Ipomoea setosa* and *Nicotiana benthamiana*. J.N. JAÉN-SANJUR, J. NAVAS-CASTILLO, E. FIALLO-OLIVÉ. Instituto de Hortofruticultura Subtropical y Mediterránea 'La Mayora' (IHSM-UMA-CSIC), Consejo Superior de Investigaciones Científicas, Algarro-

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Deltasatellites are circular single-stranded DNA molecules found in association with several begomoviruses, relying on their helper viruses for replication, movement, encapsidation, and transmission by *Bemisia tabaci*. Deltasatellites are approximately one-fourth the size of a typical begomovirus genome component. The genomes of deltasatellites do not encode any proteins, contain an adenine-rich region, a conserved satellite region, and two stem-loop structures. In nature, deltasatellites have been identified in association with both monopartite and bipartite begomoviruses in the Old and New Worlds, infecting both cultivated and wild plant species. Experimentally, some deltasatellites have been shown to be trans-replicated by their naturally associated helper virus and also by various begomoviruses and even curtoviruses. Typically, deltasatellites do not alter the symptomatology induced by their helper viruses, and their impact on viral DNA accumulation varies depending on the specific virus-host plant combination. To assess the adaptation of sweet potato leaf curl del-tasatellite 1 (SPLCD1) to different plant hosts, serial passage evolution experiments were conducted in *Ipomoea setosa* and *Nicotiana benthamiana*. Six plants of each species were agroinoculated with SPLCD1 infectious clones, followed by five successive passages using veneer grafting every four weeks. At each passage, young leaves were collected for DNA extraction, rolling circle amplification, and cloning. Sequencing and bioinformatic analyses of six clones per plant and passage were performed to investigate genetic changes associated with host adaptation. The results obtained from this study will be discussed, providing insights into the evolutionary dynamics of del-tasatellites in different plant hosts.

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Identification of whitefly-associated begomovirus 7 in *Achyranthes sicula* and its pathogenicity in tomato. E. DE LA LASTRA, I. TRUJILLO-ARTERO, E. FIALLO-OLIVÉ, J. NAVAS-CASTILLO. Instituto de Hortofruticultura Subtropical y Mediterránea 'La Mayora' (IHSM-UMA-CSIC), Consejo Superior de Investigaciones Científicas, Algarrobo-Costa, Málaga, Spain. E-MAIL: jnavas@eelm.csic.es

During systematic surveys to detect begomoviruses (genus *Begomovirus*, family *Geminiviridae*) in crops and wild plants in the province of Málaga (southern Spain), *Achyranthes sicula* plants were found to be infected with a begomovirus identified as whitefly-associated begomovirus 7 (WfaBV7). This virus had previously only been isolated from adults of the whitefly *Bemisia tabaci* MED, and its plant host was unknown. Phylogenetically, WfaBV7 is related to monopartite begomoviruses that infect tomato in several islands of southeastern Africa. An infectious clone of a WfaBV7 isolate from *A. sicula* was obtained and successfully agroinoculated into *Nicotiana benthamiana*, *A. sicula*, and tomato plants. In tomato, WfaBV7 caused mild leaf curling and a significant reduction in fruit yield. In *N. benthamiana*, WfaBV7 was able to act as a helper virus for sweet potato leaf curl delta-satellite 1. Interestingly, all attempts to transmit WfaBV7 between tomato plants using *B. tabaci* MED, MEAM1, and SSA2 as vectors were unsuccessful. Comparative analysis of the nucleotide sequences encoding the capsid protein (CP) of all available WfaBV7 isolates revealed the presence of a unique triplet of nucleotides not found in CP genes of phylogenetically related begomoviruses. Since the CP is known to be involved in vector transmission of begomoviruses, we investigated whether the additional amino acid encoded by this nucleotide triplet could be related to the lack of transmissibility by whiteflies. To this end, an infectious clone of a mutant WfaBV7 lacking this triplet was generated. The results of transmission assays performed with the mutant virus will be discussed.

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Viroid and Virus Infections in Tunisian Pistachio Orchards. M. ELAIR¹, C.E. KOURAICHI², M. DIGIARO³, N. MAHFOUDHI¹. ¹Laboratoire de Protection Des Végétaux, Université de Carthage, Institut National de La Recherche Agronomique de Tunisie, Rue Hedi Karray, 1004 ElMenzah, LR16INRAT04, Tunis, Tunisia. ²Institut Supérieur des Sciences Agronomiques de Chott Mariem, Université de Sousse, B.P 47, 4042 Chott Mériem. ³Istituto Agronomico Mediterraneo di Bari, Via Ceglie 9, 70100, Valenzano, Bari, Italy. E-MAIL: manel_elair@hotmail.com

Pistachio (*Pistacia vera* L.) is an economically important crop in Tunisia, but its production is increasingly threatened by viral and viroid infections. In spring 2022, field

surveys were conducted in the main pistachio-growing areas of Morneg, Sidi Bouzid, and Gafsa to investigate the presence of pistachio ampelovirus A (PAVA), hop stunt viroid (HSVd), and citrus bark cracking viroid-pistachio (CBCVd-pis). While chlorosis and leaf deformation were common symptoms, stunting and bushy growth were observed less frequently. A total of 144 leaf samples, collected from various cultivars and pollinator trees, were analyzed using RT-PCR. Of the analyzed samples, 116 (80.5%) tested positive for at least one of the targeted pathogens. Mixed infections of HSVd and CBCVd-pis were detected in 41% of samples, while PAVA was present in 15.9%. The highest infection rate was recorded in Sidi Bouzid (84.7%), followed by Morneg (84.2%) and Gafsa (70.7%). CBCVd-pis was the most prevalent pathogen, with incidence rates of 73.1% in Sidi Bouzid, 60.9% in Gafsa, and 57.8% in Morneg. In Morneg, female cultivars exhibited a higher infection rate (88%) compared to pollinator trees (77%), with frequent co-infections by both viroids. Among pollinator plants, 50% were infected with HSVd and 63.6% with CBCVd-pis. The higher incidence of HSVd and CBCVd-pis compared to PAVA may reflect efficient transmission through pruning tools and potential pollen-mediated dissemination. Further research is needed to understand their epidemiology, incidence, and symptom severity. Tunisia is currently implementing a certification program to produce virus- and viroid-free pistachio propagation material.

Preliminary identification of a novel Maculavirus infecting peach in California using High-Throughput Sequencing. R.A. KUBAA¹, A. OURO-DJOBO², K.A. STEVENS¹, O.J. ALABI², M. AL RWAHNIH^{1,3}. ¹Department of Plant Pathology, University of California, Davis, CA 95616, U.S.A. ²Department of Plant Pathology & Microbiology, Texas A&M AgriLife Research and Extension Center, Weslaco, TX 78596, U.S.A. ³Department of Plant Protection, School of Agriculture, The University of Jordan, Amman, Jordan. E-MAIL: raboukubaa@ucdavis.edu

The application of High-Throughput Sequencing (HTS) in routine virus screening has significantly improved the detection of known and novel viruses in vegetatively propagated crops. In 2020, Foundation Plant Services (FPS) at the University of California, Davis, received a peach selection from a public breeding program for evaluation and testing. Leaf tissue from grafted plants was analyzed via HTS as part of the plant health diagnostic pipeline. Bioinformatics analysis revealed the presence of a previously uncharacterized RNA virus showing

sequence similarity to members of the family *Tymoviridae*. Further analysis by HTS, followed by RT-PCR and rapid amplification of cDNA ends (RACE), enabled the complete sequencing of the viral genome which is 6,664 nucleotides long (excluding the poly(A) tail), and confirmed its affiliation with the genus *Maculavirus*, which includes monopartite, positive-sense single-stranded RNA viruses. The infected plant exhibited no visible symptoms at the time of sampling, suggesting a latent or asymptomatic infection. Preliminary genomic analysis indicated that the virus has a two-ORF genome organization and contains conserved domains such as methyltransferase, helicase, and RNA-dependent RNA polymerase. Phylogenetic trees based on both ORFs placed the virus within the *Maculavirus* clade. Subsequent RT-PCR testing confirmed the virus in both the scion and rootstock of grafted plants, while original rootstocks tested negative, indicating transmission occurred via bud grafting. This study highlights the importance of integrating HTS into certification programs for early detection of latent viral infections that may otherwise go unnoticed by conventional methods.

Emergence of Paprika mild mottle virus infecting pepper (*Capsicum annum* L.) crops in Türkiye. N. BUZKAN¹, B.B. ARPACI². ¹Plant Protection Department, Agriculture Faculty, Kahramanmaraş Sütçü İmam University, Kahramanmaraş, TÜRKİYE. ²Horticulture Department, Agriculture Faculty, Kilis Yedi Aralık University, Kilis, TÜRKİYE. E-MAIL: nbuzkan@ksu.edu.tr; nbuzkan@gmail.com

During August 2013 onwards, foliar (unfamiliar curling, distortion, yellowing) and fruit (reduced size, discoloration, mottle, pitting) symptoms were identified in pepper crops (*Capsicum annum* L.) in Aegean region in Turkey. Fruit and leaf samples were tested in DAS-ELISA for the most frequent viruses such as potato virus Y (PVY), tobacco etch virus (TEV), cucumber mosaic virus (CMV), tomato spotted wilt virus (TSWV) and cucurbit aphid-borne yellows virus (CABYV) in Turkey. The samples resulting positive for CABYV were subjected to RT-PCR with the general polerovirus primer pair. Direct sequencing of PCR amplicons and BLASTn search proved the presence of paprika mild mottle tobamovirus (PaMMV) in a mixed infection nature with a polerovirus detected. Extensive survey was later carried out in protected and open-field grown pepper plants out in 11 provinces throughout Thrace, eastern Mediterranean, Southeast Anatolia, Aegean, eastern Black Sea regions from June to August in 2013 – 2022. A total of

478 samples were tested with RT-PCR with primer pairs amplifying the corresponding region to the coat protein (CP) gene of PaMMoV. Only samples from northern Aegean region resulted positive for PaMMV. The nucleotide sequences corresponding to 451 nt-long the CP gene of the Turkish PaMMV isolates showed 97.7% sequence similarity with PaMMV pepper strain P11 from Spain. PaMMV as an emergent virus is firstly detected in pepper crops in Turkey and the results support its widespread in the region due to the infected seeds sold by local seed companies.

This research was partially granted by KSU BAP 2021/2-28D.

Image-based high-throughput phenotyping of grafted tomato plants upon virus or viroid infection. R. MARZIALE¹, L. VACCARO², R. SPANO¹, G. BUBICI¹, M. CHIUMENTI¹, A. PETROZZA³, F. CELLINI³, T. MASCIA², B. NAVARRO¹, F. DI SERIO². ¹Institute for Sustainable Plant Protection, National Research Council of Italy CNR, Via Amendola 165/A, 70126, Bari, Italy. ²Università degli Studi di Bari Aldo Moro, Via Amendola 165/A, 70126, Bari, Italy. ³Research Centre of the Lucana Agency for Development and Innovation in Agriculture (ALSIA) Metapontum Agrobios, Metaponto, Italy. E-MAIL: robertamarziale@cnr.it

Grafting is an agronomic technique used as a sustainable integrated strategy for the management of biotic and abiotic stresses. Besides improving the performance of several horticultural crops, grafting can enhance the tolerance to viral infections in tomato. Whether grafting may interfere also with viroid infections in this host plant is still unknown. Here, we investigated by high-throughput phenotyping (Scanalyzer 3D system) the responses of grafted and non-grafted tomato plants to infections with potato spindle tuber viroid (PSTVd) or a recombinant strain of potato virus Y (PVY^{c-to}), which elicit stunting and leaf curling or severe leaf distortion and mosaic in this host, respectively. Two tomato varieties, Manduria (Ma) and UC82 (UC), tolerant and highly susceptible to PVY^{c-to} infection, respectively, were tested. Non-grafted, self-grafted UC (UC/UC) and UC grafted onto Ma (UC/Ma) plants were mechanically inoculated with PVY^{c-to}, PSTVd or mock-inoculated. Quantitative data of morphological parameters were acquired at 9 time points up to 36 dpi. PCA analysis showed that grafting has a global effect on PSTVd and PVY^{c-to} infection, with a slightly positive effect of UC/Ma with respect to self-grafted tomato plants. Moreover, a delay in viroid-induced stunting and a reduced accumulation of the viroid were

observed in the grafted plants. A slight increase in plant area, silhouette and solidity values were measured in grafted-PVY^c-to plants compared to the non-grafted counterparts starting at 15 dpi, associated with the lower viral load. These preliminary results show that high-throughput phenotyping is a promising tool to estimate tomato response across PVY^c-to and PSTVd infections.

This research was financially supported by European Union-NextGeneration EU, Missione 4 'istruzione e ricerca', Componente 2, investimento 1.1 'progetti di ricerca di rilevante interesse nazionale (PRIN), project PRIN-2022 "DiVInGraft: dissection of molecular mechanisms underlying tolerance to virus and viroid infection in grafted tomato plants", ID: 2022BZW9PF, CUP B53D23017480006 and CUP H53D23005160006.

Characterization of two isolates of *Bougainvillea chlorotic vein banding virus* in Italian *Bougainvillea* plants. A. BEN SLIMEN, R. YAZBECK, T. ELBEAINO, M. DIGIARO. *International Centre for Advanced Mediterranean Agronomic Studies (CIHEAM IAMB), Bari, Italy.* E-MAIL: b.slimen@iamb.it

Despite their global distribution, ornamental *Bougainvillea* plants have received limited attention in virological studies. In this study, fifty-eight *Bougainvillea* samples, both symptomatic and asymptomatic, were collected from various nurseries across Apulia, Southern Italy. To investigate the presence of viruses, total RNA was extracted from pooled samples and subjected to high-throughput sequencing (HTS) using the Illumina platform. Analysis of the assembled contigs from the HTS data led to the identification of two distinct isolates of *Bougainvillea chlorotic vein banding virus* (BCVBV), genus *Badnavirus*, known to infect *bougainvillea*. Complete viral genomes were reconstructed by assembling contigs and bridging sequence gaps through PCR using newly designed primers. The first isolate, designated BCVBV Apulia 1 (PQ682518), comprised 8,694 nucleotides and showed highest sequence identity with the Brazilian isolate BCVBV-UNB-01 (MK473389), sharing 81.6% nucleotide and 84.0% amino acid identity. The second isolate, BCVBV Mini-Thai (PQ682517), was 8,822 nucleotides long and displayed high sequence identity to the Malaysian isolate BCVBV-UKM (MK816926), with 98.5% nucleotide and 98.7% amino acid identity. Genome structure analysis confirmed the typical BCVBV organization, featuring four open reading frames. Phylogenetic analysis placed the two Italian isolates in separate clades. Subsequent screening of individual samples revealed the presence of BCVBV in 12 out of 58 samples (20.7%), with the Apulia 1 isolate in

7 samples (12.1%) and BCVBV Mini-Thai in 5 samples (8.6%). This study represents the first report of BCVBV in Italy and Europe, identifying two genetically distinct strains and underscoring the need for further investigation into the epidemiology and impact of this virus.

This research was financially supported by the Regional Project "Pro.Di.Qua.Vi." (Trasferimento di Protocolli per organismi da quarantena e nocivi e per la selezione di materiali sanitariamente migliorati per il vivaismo pugliese). Misura 16 Cooperazione – Sottomisura 16.2).

CONCURRENT SESSION B2 – Emerging and re-emerging wood and vascular diseases of grapevine

SESSION KEYNOTES

Wood and vascular diseases of grapevine: emerging and re-emerging problems. L. MARTÍN. *Centro de Investigaciones Científicas y Tecnológicas de Extremadura (CICYTEX) Instituto de Investigaciones Agrarias Finca La Orden - Valdesequera, Área de Protección Vegetal, Autovía A-5 Km 372, 06187 Guadajira, Badajoz, España.* E-MAIL: laura.martin@juntaex.es

Vitis vinifera is one of the oldest and widely cultivated fruit crops globally, with over 7 million hectares under cultivation. In Europe, wine production constitutes a major socio-economic sector. Grapevines host a complex microbiota, including more than 900 fungal species reported to interact within the *Vitis* environment. Among these fungal community there are several well-known phytopathogenic fungi that cause diseases affecting leaves (e.g., downy and powdery mildew), fruit (e.g., grey mold and bunch rot), and roots (e.g., white rot and wilt). However, increasing attention is now being directed toward root and vascular diseases caused by soilborne fungi, particularly those affecting young vines. Decline syndromes in young grapevines have raised special attention among plant pathologists. These include root rot caused by *Fusarium* spp., black-foot disease caused by *Cylindrocarpus*-like species, and Petri disease. Both black-foot and Petri disease are part of the Grapevine Trunk Diseases (GTDs) complex. Since the early 2000s, the restriction of certain chemical treatments in Europe due to environmental and health concerns has coincided with the resurgence of GTDs, now considered as one of the major threats to the sustainability of viticulture. GTDs comprise a diverse group of diseases affecting both mature vineyards (e.g., Esca complex, Eutypa dieback, Botryosphaeria dieback, Diaporthe dieback) and

young plantations (e.g., Petri disease, black-foot disease). These diseases are chronic and often asymptomatic for years post-infection. However, they ultimately reduce vineyard longevity, yield, and grape quality, leading to plant death and decreased economic profitability of wine industry. The etiology of GTDs is complex, involving a wide range of taxonomically unrelated fungal pathogens—up to 30 genera and more than 100 species—isolated from the perennial wood of grapevines. While *Vitis vinifera* cultivars exhibit variable levels of tolerance and activate specific defense responses to vascular pathogens, complete resistance to GTDs has not been described. Recent advances in molecular technologies, particularly next-generation sequencing (NGS), have significantly enhanced our understanding of the grapevine mycobiome. Fungal community composition varies across the grapevine's vegetative cycle, between cultivars and plant tissues, and as a function of plant age. Additionally, microbial diversity is shaped by biogeographical factors and vineyard management practices. These insights highlight the complexity of fungal communities and emphasize the importance of understanding their ecological roles—including trophic modes and guild affiliations—as pathogens, endophytes, saprobes, or potential biocontrol agents. This growing knowledge provides a foundation for the development of innovative and sustainable disease management strategies. By integrating beneficial microorganisms, bioactive compounds, and improved agronomic practices, we can more effectively address wood and vascular diseases in vineyards and support growers in reducing losses caused by fungal pathogens.

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Grapevine Flavescence dorée: an update. E. ANGELINI. CREA Centro di Ricerca per la Viticoltura e l'Enologia, Via XXVIII Aprile 26, 31015 Conegliano, TV, Italy. E-MAIL: elisa.angelini@crea.gov.it

Flavescence dorée (FD) is a quarantine disease of grapevine associated to the presence of FD phytoplasmas (FDp). It is efficiently transmitted from vine to vine by *Scaphoideus titanus* Ball., a leafhopper of American origin, imported to Europe in the last century. FD is strongly epidemic in Central-Southern Europe, occurring from Portugal to Romania. Damages are very serious, spanning from lower production to the death of the

plants. Not all the varieties show the same susceptibility, with most of the rootstocks and a very few *Vitis vinifera* cultivars being less susceptible. Until few years ago the epidemiologic cycle FDp - *S. titanus* - grapevine has been thought to be the only one, while currently there are many scientific reports that demonstrated the presence of several secondary host plants and other secondary vectors, although with lower epidemiologic impact. The genetic variability of the pathogen plays an utmost role in FD epidemiology, with only a few isolates being epidemic, because transmitted by *S. titanus*. The main strategies to control the disease and the vector are mandatory and rely on insecticide treatments against the vector, uprooting of the infected plants, and planting of healthy material. However, several studies are ongoing to find out most sustainable and durable control strategies. Vibrational mating disruption of vector is one of these possibilities, since males and females communicate and mate via vibrations. It was demonstrated to be effective in laboratory conditions, and field trials are ongoing. Stimulation of plant natural defences is also a very interesting issue, however so far successful trials are missing. Gene silencing, based on RNA interference, can be used for controlling vectors: experiments with FD vectors (*S. titanus* and *Euscelidius variegatus*) were carried out in laboratory conditions and showed decreased fitness and lower egg development. Natural antagonists to the vector are not present in Europe. Some trials with the natural endosymbiont *Asaia* sp., already present in European *titanus* populations, gave good results in the model FD vector *E. variegatus*, leading to significantly reduced phytoplasma acquisition. Also a few studies on the resistance and susceptibility of grapevine varieties, against the vector and against the pathogen, are in progress, but they proceed slowly due to the complexity of the pathogen, which is not cultivable *in vitro* and transmissible only *in vivo* by means of the vector. Quantitative Trait Loci identification is indeed ongoing by classical approaches, such as breeding and phenotyping, and by modern tools, such as transcriptomic and Next Generation Sequencing. The genetic traits responsible for susceptibility and resistance to FD are thus not yet known, however their discovery could help a lot in producing grapevine plants resistant to the disease, by means of conventional breeding, genetic editing, and cisgenesis techniques.

ORAL PRESENTATIONS

Long-term establishment and active colonization of *Trichoderma atroviride* SC1 and *Pseudomonas chlo-*

roraphis M71 in the trunk of Esca-affected grapevine after their injection. G. BRUSSI¹, G. PUOPOLO^{1,2}, S. DI MARCO³, L. MUGNAI⁴, I. PERTOT^{1,2}. ¹Center Agriculture Food Environment (C3A), University of Trento, San Michele all'Adige, Italy; ²Research and Innovation Center, Fondazione Edmund Mach, San Michele all'Adige, Italy; ³Institute of BioEconomy, National Research Council, Bologna, Italy; ⁴Plant Pathology and Entomology Section, Department of Agricultural, Food, Environmental and Forestry Science and Technology (DAGRI), University of Florence, Florence, Italy. E-MAIL: greta.brussi@unitn.it

The increasing difficulties in protecting plant health pave the way for integrating endotherapy as a sustainable alternative, especially in contexts where conventional treatments are complex and/or ineffective, such as the Esca Complex, one of the primary wood diseases affecting grapevines. Despite numerous benefits, endotherapy is rarely used due to high application costs, the limited availability of effective formulations, and unclear outcomes. In this scenario, the use of plant-beneficial microorganisms for endotherapy could be a promising method for controlling plant diseases since this approach benefits from the plant's active and persistent colonisation of these microorganisms. Additionally, it enables them to reach areas that would otherwise be inaccessible to the passive movement of injected compounds, eliminating the need for multiple applications throughout the year. This work aims to verify if the plant-beneficial microorganisms *Pseudomonas chlororaphis* M71 and *Trichoderma atroviride* SC1 can systemically establish themselves once injected, both in rooted cuttings in the greenhouse and grapevine plants in the vineyard. Experiments were conducted to evaluate the colonisation efficiency, translocation within the grapevine plant, long-term persistence, and protection against the main Esca pathogens. The results confirmed the successful establishment of both microorganisms, with extensive translocation throughout the grapevine tissues, providing reassurance about the method's effectiveness. Long-term colonisation results showed that these plant-beneficial microorganisms could persist in the trunk for at least 60 days. Further investigation is needed to optimise injection protocols and assess the practical applications of this method under field conditions.

From *in vitro* screening to vineyard evaluation of endophytic biocontrol bacteria targeting *Fomitiporia mediterranea*, a key pathogen of esca, a grapevine trunk disease. O. MESGUIDA^{1,2}, R. TRAVA-

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Esca is a complex grapevine trunk disease that represents a major challenge to viticulture worldwide. Since the banning of sodium arsenite in European countries, the last approved pesticide, no curative treatment is available for its control. In this context, our study aimed to (i) select bacterial biological control agents (BCAs) effective against *Fomitiporia mediterranea* (Fmed), a major pathogen associated with Esca, (ii) investigate their modes of action, and (iii) evaluate their efficacy under field conditions. A stepwise *in vitro* screening of increasing complexity was performed to evaluate the inhibitory potential of 58 grapevine endophytic bacteria against Fmed. Three strains *Pseudomonas lactis* SV9, *Pseudomonas paracarnis* S45, and *Paenibacillus polymyxa* SV13 which exhibited a strong antifungal activity against Fmed were selected. Whole-genome and metabolome analyses of the three strains revealed that their biocontrol activity operates through both direct and indirect mechanisms. The efficacy of *P. paracarnis* S45 and *P. polymyxa* SV13 was evaluated in a vineyard trial by injecting them into the trunks of Sauvignon Blanc grapevines exhibiting Esca symptoms. Bacterial injections reduced the severity of Esca without affecting grape quality. The abundance of *Bacillus* increased in grapevines injected with *P. polymyxa* SV13, which correlated negatively with the abundance of *Phaeomoniliella* spp. and *Diplodia* spp.; a negative correlation between *Pseudomonas* spp. and *Fomitiporia* spp. was observed in grapevines injected with *P. paracarnis* S45. Furthermore, *P. paracarnis* S45 improved the defense mechanisms of the grapevine. These results highlight the potential of both strains as effective biological control agents for the management of Esca.

This work was supported by the Excellence Initiative of Université de Pau et des Pays de l'Adour–I-Site E2S UPPA [Project Biovine, seed funding], a French “Investissements d'Avenir” program and by the Industrial Chair “WinEsca” funded by the ANR (French National Research Agency, grant number ANR-22CHIN-0002-01), and the JAs Hennessy & Co and GreenCell companies.

One formulation for the control of downy mildew and grapevine trunk diseases in vineyard: The “no-copper” challenge of the Natural Agro project. V. MONDELLO, M. BOUVY, L. PARENT, J.F. GUISE, P. TROTEL-AZIZ, F. FONTAINE. *University of Reims Champagne-Ardenne, Research Unit Induced Resistance and Plant Protection – RIBP UCS INRAE 1488, Building 18, 51100 Reims, France.* E-MAIL: florence.fontaine@univ-reims.fr

The concerns about the impact of plant protection products (PPPs) on environment and human health lead the industry in finding new more sustainable ways to control diseases. This is particularly urgent in viticulture where the massive use of cupric-based PPPs has led to high level of copper contamination in soils and water. For these reasons, copper is now considered as “candidate for substitution” (art. 24 EC Regulation n. 1107/2009) by the European Community (EC) and its use is now limited at max. 28 kg-ha⁻¹ in 7 years (1981/2018 EC regulation). In this framework, the European LIFE project namely NaturalAgro (2023-2028), involving France, Italy and Portugal, aims to homologate one formulation for the control of two major grapevine diseases: the downy mildew and GTDs. The formulation is based on the carrier molecule hydroxyapatite (Microsap®), plant extracts and will contain no copper. The potential PPP will be tested, with different strategies according to the targeted disease, in several vineyards located in the three NaturalAgro project countries. Here we present some preliminary results obtained during our in vitro and in planta tests especially against some Esca-, Eutypa- and Botryosphaeria-dieback associated pathogens. These results were useful to identify the best formulation among those tested, differing in both components and relative ratios. The chosen potential PPP seems to be promising in the protection of pruning wounds from GTD-pathogens infections. Experimental plots in French vineyard have been initiated in 2024 and for 4 years.

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Exploring soil properties influence on Esca Disease dynamics: insights from a Bolgheri vineyard. F. BIGAZZI¹, G. CARELLA¹, S. DEL DUCA², S. MOCALI², S. PRIORI³, F. VITALI², L. MUGNAI¹. ¹*Department of Agricultural, Food, Environmental and Forestry Science and Technology (DAGRI), Plant pathology and Entomology section, P. le delle Cascine 28, 50142 Florence,*

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Esca complex of diseases remains one of the most damaging grapevine disorders, yet its causal factors are not fully understood. While numerous fungi are involved, and heavy rainfall in May and June is known to exacerbate foliar symptoms, the role of soil physicochemical properties in disease expression remains poorly investigated. This study aims to explore the influence of soil properties on leaf symptoms expression by comparing vineyard areas with high and low disease incidence over a five-year period. The research was conducted in a 1.5-hectare ‘Cabernet Sauvignon’ vineyard in Tuscany (Italy). Electromagnetic induction (EMI) sensor was employed to map soil spatial variability through soil electrical conductivity (ECa). Four sampling points were selected for profile excavation based on the combination of disease incidence and ECa. Soil features revealed significant differences between high- and low-incidence areas. Low-incidence soils showed clay-loam texture, high lime content, alkaline pH (8.3–8.5), and low assimilable manganese (6.8–12.3 mg kg⁻¹). In contrast, high-incidence soils showed deep soils with sandy-clay-loam texture, low lime content (< 3%), sub-alkaline pH (7.8–8.1), greater fertility (higher nitrogen) and good amount of organic matter. Assimilable manganese was slightly higher in high-incidence soils (13–18 mg kg⁻¹). These findings suggest that soil structure, pH, and organic matter buffering capacity may significantly influence micronutrient availability, particularly manganese, potentially enhancing the degradative activity of *Fomitiporia mediterranea*, a key Esca pathogen. This could result in increased production of byproducts and toxins, contributing to foliar symptoms. This study highlights the critical role of soil properties in Esca disease dynamics.

Grapevine trunk diseases in Greece: disease incidence and fungi involved in discrete geographical zones and varieties. S.I. TESTEMPASIS¹, E.A. MARKAKIS^{2,3}, G.I. TAVLAKI³, S. SOULTATOS^{2,3}, C. TSOUKAS⁴, A. SAMARAS¹, D. GKIZI⁴, A. TZIMA⁴, E. PAPLOMATAS⁴, G.S. KARAOGLANIDIS¹. ¹*Aristotle University of Thessaloniki, School of Agriculture, Faculty of Agriculture, Forestry and Natural Environment, 54124 Thessaloniki, Greece.* ²*School of Agricultural Sciences, Hellenic Mediterranean University, Stavromenos 71004, Heraklion, Crete,*

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The main objective of the present study was to estimate disease parameters and isolate fungi associated with grapevine trunk diseases (GTDs) in Greece. In total, 310 vineyards in different geographical regions in northern, central, and southern Greece were surveyed, and 533 fungal strains were isolated from diseased vines. Morphological, physiological and molecular analyses revealed that isolates belonged to 35 distinct fungal genera. The GTDs-inducing population structure differed significantly among the discrete geographical zones. *Phaeomoniella chlamydospora* (26.62%, *n*=70), *Diaporthe* spp. (18.25%, *n*=48) and *Fomitiporia mediterranea* (10.27%, *n*=27) were the most prevalent in Heraklion, whereas *Diplodia seriata*, *Alternaria* spp., *P. chlamydospora* and *Fusarium* spp. were predominant in Nemea. In Amyntaio and Kavala, *D. seriata* was the most frequently isolated species (>50% frequency). Multi-gene (*rDNA*-ITS, *LSU*, *tef1-α*, *tub2* και *act*) sequencing of selected isolates, followed by pathogenicity tests, revealed that *Neosetophoma italica*, *Seimatosporium vitis*, *Didymosphaeria variabile* and *Kalmusia variispora* caused wood infection, with the former being the most virulent. To the best of our knowledge, this is the first report of *N. italica* associated with GTDs worldwide. This is also the first record of *K. variispora*, *S. vitis* and *D. variabile* associated with wood infection of grapevine in Greece. The potential associations of disease indices with vine age, cultivar, GTD-associated population structure and the prevailing meteorological conditions in different viticultural zones in Greece are presented and discussed.

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POSTER PRESENTATIONS

Tolerance of cultivars and hybrid grapes against trunk diseases. A. CSÓTÓ¹, N. LAURINYECH¹, A. NAGY¹, N. RAKONCZÁS², K.E. NÉMETH³, C. NÉMETH⁴, Z. A. NAGY⁴, A. CSIKÁSZ-KRIZSICS⁵, E. SÁNDOR⁶. ¹Institute of Plant Protection, Faculty of Agricultural and Food Science and Environmental Management, University of Debrecen, H-4032, Debrecen, Böszörményi út 138, Hunga-

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Grapevine trunk diseases are among the most significant problems of grape plant protection worldwide, and there is no adequate curative procedure for its treatment. In Hungary, little is known about the resistance of the varieties in the environmental conditions of the region, neither that of domestic cultivars nor that of breeding lines. In our studies, we assessed the annual frequency of symptoms in 2022, and the degree of plant loss before that, for 305 varieties of four Hungarian grape variety collections. We compared the order of susceptibility of dominant domestic and internationally significant varieties. We formed groups of the varieties based on their descent as follows i) monophyletic varieties of *Vitis vinifera* origin and interspecific hybrids, and ii) monophyletic varieties of *V. vinifera* origin, Asian (*V. amurensis*) and American (*V. labrusca*, *V. riparia*, *V. rupestris*) origin hybrids. The tolerance of the resulting groups was compared based on their mortality. Our susceptibility results for the international varieties are supported by the previous literature data. Compared to the symptoms of the international varieties, the main Hungarian variety 'Juhfark' is the most resistant, while 'Furmint' can be classified as the most sensitive. The mortality of interspecific hybrids is significantly lower than that of monophyletic varieties, and the resistance of species hybrids derived from *V. amurensis* proves to be outstanding even within this group.

Bacteria and fungi colonizing grapevine wood issues: Taxa diversity and interactions in the context of grapevine trunk diseases. R. HAIDAR, A. YACOB, O. MESGUIDA, L. LEYMARIE, E. ATTARD, R. GUYONEAUD, P. REY. Université de Pau et des Pays de l'Adour / E2S-UPPA/ CNRS, Institut des Sciences Analytiques et de Physicochimie pour l'Environnement et les Matériaux - UMR 5254, Pau, F-64013 France. E-MAIL: rana.haidar@univ-pau.fr

Pathogenic fungi are considered as the major cause of Grapevine Trunk Diseases (GTDs) which threat wine and table grape industries worldwide. While fungal diversity in different cultivars is well studied, less information are available about bacterial diversity in grapevine wood. The function of these bacteria and their interactions with wood-inhabiting fungi is relatively less studied. Using metabarcoding, we identified the bacterial and fungal taxa composition of microbiota colonizing different wood tissues of five grapevine varieties (Ugni Blanc, Tannat, Baroque, Gros menseng and Arri-loba) from the Nouvelle-Aquitaine region in France. The results of next-generation sequencing are under analysed. These data will provide a better understanding of bacterial and fungal diversity, and their distribution in the various tissues of grapevine cultivars. To the best of our knowledge, this is the first report on the diversity of cultivable endophytic bacterial in the wood of these grapevine varieties. By using cultural dependent methods, 135 bacteria from the different grapevine varieties were isolated and assigned to bacterial species based on the 16S rRNA genes. 39 bacterial strains issues from different tissues of different cultivars and showed different ability to degrade cellulose, were selected to study their interaction, *in vitro*, with four of major pathogenic fungi of GTDs (*Fomitiporia mediterranea*, *Neofusicoccum parvum*, *Diplodia seriata* and *Eutypa lata*). Diverse types of bacterial/fungal interactions were observed which depended on bacterial and/or fungal species. However, three bacterial strains (*Bacillus amyoliquefaciens*, *B. subtilis* and *Priestia megaterium*) have showed a great potential as biocontrol agents against the studied fungi of GTDs.

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New potential BCA from table grape microbial community for Grapevine Trunk Diseases (GTDs) management. D. CORNACCHIA¹, F. DALENA¹, D. SALAMONE¹, A. AGNUSDEI¹, P. TANCREDI¹, R.M. DE MICCOLIS ANGELINI¹, G.L. BRUNO¹, F. MANNERUCCI¹, D. GERIN¹, F. FARETRA¹, S. MAVICA², D. AIELLO², S. POLLASTRO¹. ¹Department of Soil, Plant and Food Sciences, (DiSSPA), University of Bari Aldo Moro, Bari, Italy. ²Department of Agriculture, Food and Environment, University of Catania, via S. Sofia, 100, Catania, Italy. E-MAIL: francesco.faretra@uniba.it; dalia.aiello@unict.it

In this work, the antagonistic activity of members of table-grape microbial community, isolated from Apulia and Sicily, and the registered *Bacillus amyoliquefaciens* (D747), *Bacillus subtilis* (QST713), *T. atroviridae* SC1, *T. asperellum* ICC012 and *T. gamsii* ICC080 was assessed against Grapevine trunk diseases (GTDs) associated pathogens *Neofusicoccum parvum*, *Eutypa lata*, *Diplodia seriata*, *Cylindrocarpon destructans*, *Phaeomoniella chlamydospora*, *Phaeoacremonium minimum*, *Diaporthe neoviticola*. In dual culture assay, almost all *Trichoderma* strains differently affected the growth of tested pathogens, showing greater effectiveness than *T. gamsii* ICC080 and *T. asperellum* ICC012, and similar activity to *T. atroviridae* SC1. Previously, *Aphanocladium album* MX95 showed a deadlock at mycelial contact with *F. mediterranea*. *Clonostachys rosea* GP80 inhibited the growth of *P. minimum*. *B. amyoliquefaciens* (D747) and *B. subtilis* (QST713) showed a limited effectiveness against *N. parvum* and *E. lata*. Furthermore, the ability of the BCA to produce volatile organic compounds (VOCs) was evaluated. All the *Trichoderma* tested were effective against *P. chlamydospora*, *C. destructans* and *P. minimum*. The *Trichoderma* strains FV145 and FV185 were the best performant, showing similar activity to *T. atroviridae* SC1. The growth of *C. destructans* and *P. chlamydospora* was affected by *A. album* and *C. rosea*. *B. amyoliquefaciens* (D747) showed a moderate effectiveness against *P. chlamydospora*, *C. destructans* and *P. minimum*, while *B. subtilis* (QST713) light reduced the growth of *P. chlamydospora* and *C. destructans*. Moreover, *Trichoderma* TCH4 altered the colour of *N. parvum* and *D. seriata* mycelia. These results highlight the potential of native microorganisms as effective biocontrol agents against GTDs.

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Studying the evolution of Flavescence dorée symptomatology for the assessment of an early detection strategy. R. PINI, C. AGLIETTI, G. CARELLA, F. BIGAZZI, A. ROSSI, L. MUGNAI. Department of Agricultural, Food, Environmental and Forestry Science and Technology (DAGRI), Plant pathology and Entomology section, University of Florence, P.le delle Cascine 28, 50144 Florence, Italy. E-MAIL: chiara.aglietti@unifi.it

Flavescence dorée (FD) is an invasive grapevine disease reported in many European countries. Despite the quarantine measures mandatory for its control, concerning FD epidemic outbreaks continue to be reported. Investigating the FD symptoms development in 'Sangiovese', the most typical variety of the Chianti area in Tuscany, is useful to reduce delays between symptoms manifestation and management actions. In this work the evolution of FD was analyzed in three Sangiovese vineyards, where the development of symptoms was followed for 1 year, registering the presence and classifying each kind of symptom observed on leaves, shoots and trunks. Every 15 days, samples of each part of the analyzed plants were collected from each vineyard and tested for the presence of FD with a species-specific LAMP assay. The presence of vector insects, *Scaphoideus titanus* and *Dictyophara europaea*, was monitored to confirm flight periods and to correlate insect presence with disease incidence and severity. The higher percentage of positives to FD was found from leaf samples collected from shoots with zigzagging tips and curled leaves, while the lowest was observed in samples taken from shoots and leaves with a marbled lamina. The higher percentage of inoculum of FD was registered from mid-June to late July, while most insects were found between late July and end of August. Early symptom monitoring showed to be of great help, along with confirming sample positivity through molecular testing by the first ten days of July, to enable the early removal of infected plants before the arrival of adult *S. titanus*.

This research was partly financially supported by Enbiotech s.r.l.

Presence of GTD pathogens in young vineyards in Montenegro. B. KANDIĆ¹, V. MONDELLO², J. LATINOVIC¹, F. FONTAINE², N. LATINOVIC¹. ¹University of Montenegro, Biotechnical Faculty, Mihaila Lalića 15, 81 000 Podgorica, Montenegro. ²University of Reims Champagne-Ardenne, INRAE, RIBP USC 1488, 51100 Reims, France. E-MAIL: kandic.b@ucg.ac.me

Grapevine trunk diseases (GTDs) are caused by several fungi belonging to different genera and not phylogenetically related. They are widespread in all grapevine cultivation countries, causing significant economic losses. Due to the scarce knowledge about the presence of GTDs in Montenegro, a survey for the presence of these diseases in vineyard was carried out from 2021 to 2024. A total of 72,500 vines in 15 different vineyards were inspected. During the survey, samples of asymptomatic

vines were uprooted and analyzed to verify the presence of any internal GTD-like symptoms. A total of 10 samples from 13 different varieties were collected and processed in the laboratory of the Biotechnical Faculty in Podgorica. Generally, the survey for the GTD presence in the 15 Montenegrin vineyards showed the absence of foliar symptoms which resembles GTDs. However, both cross and longitudinal sections of the sampled vines highlighted the presence of black spots and small brown necrosis, especially in the wood of the upper third of the vine while the middle part and the grafting point did not show any symptoms of internal necrosis. The morphological and molecular identification of isolates obtained from asymptomatic grapevine plants showed the presence of GTD-associated pathogens *Phaeomonella chlamydospora*, *Diaporthe ampelina* and pathogens from the family *Botryosphaeriaceae*. The acquired results indicate the presence of latent infections by GTD pathogens in young grapevine plantings in Montenegro.

This research was financially supported by the Project "Biofungicides application in agriculture and urban areas (BIOAPP)", funded by Ministry of Education, Science and Innovation of Montenegro and abstract was created as a result of the ERASMUS + KA 171 project implemented between the University of Montenegro and the University of Reims Champagne-Ardenne.

In vitro insights into manganese-driven wood degradation by *Fomitiporia mediterranea*: are micronutrients abiotic triggers for foliar symptoms in Esca complex? F. BIGAZZI, G. CARELLA, L. MUGNAI. Department of Agricultural, Food, Environmental and Forestry Science and Technology (DAGRI), Plant pathology and Entomology section, P.le delle Cascine 28, 50142 Florence, Italy. E-MAIL: francesco.bigazzi1@unifi.it

Fomitiporia mediterranea (Fmed) is a basidiomycete fungus having a relevant role in grapevine wood tissue degradation associated with Esca complex of diseases and of its typical foliar symptoms. While abiotic factors such as soil drought suppress symptom expression, fertilization with specific microelements appears to exacerbate it, suggesting that soil chemistry plays an understudied role in disease dynamics. This study focuses on manganese (Mn), hypothesizing that its bioavailability in soils—through accumulation in vine tissues—could stimulate Fmed wood-degrading capacity by enhancing Mn-dependent enzymatic activity. We therefore evaluated whether Mn supplementation influences Fmed growth, wood decay, and secretome composition. Fmed was cultured on water agar with graded Mn concentrations. Mycelial growth rates and biomass production were

monitored on vine sawdust, while wood degradation was assessed via mass loss in pre-weighed wood chips. Results revealed statistically significant differences in fungal growth, biomass, and wood degradation between Mn-supplemented and control substrates. Moderate Mn enrichment accelerated mycelial growth, increased biomass production, and intensified wood mass loss compared to controls. However, elevated Mn concentrations suppressed fungal development, indicating a toxicity threshold. Visual analyses showed distinct color changes in the fungal colony under Mn-enriched conditions, suggesting altered metabolite profiles. These findings suggest that Mn availability modulates Fmed degradative activity, with optimal levels promoting enzymatic processes critical to wood decay. These results highlight the need to investigate micronutrient bioavailability as a potential abiotic driver of Esca disease progression, with Mn emerging as a key factor influencing pathogenesis through modulation of fungal enzymatic activity and wood degradation processes.

Phomopsis cane and leaf spot of grapevine is associated with *Diaporthe* dieback: first report in Italy. G. CARELLA¹, F. BIGAZZI¹, G. DARDANI², V. GUARNACCIA^{2,3}, L. MUGNAI¹. ¹Department of Agricultural, Food, Environmental and Forestry Science and Technology (DAGRI), Plant pathology and Entomology section, P.le delle Cascine 28, 50142 Florence, Italy. ²Department of Agricultural, Forest and Food Sciences (DISAFA), University of Turin, Largo Braccini 2, 10095 Grugliasco (TO), Italy; ³Interdepartmental Centre for Innovation in the Agro-Environmental Sector, AGROINNOVA, University of Turin, Largo Braccini 2, 10095 Grugliasco (TO), Italy. E-MAIL: giuseppe.carella@unifi.it

Phomopsis cane and leaf spot (PCLS), a disease caused by *Diaporthe* spp., is often considered secondary in viticulture. Recent studies classify *Diaporthe* species among Grapevine Trunk Diseases agents, causing *Diaporthe* dieback. This pathogen disrupts vascular tissues, obstructing sap flow and inducing spur decline. A three-year monitoring study (2022–2024) was conducted in a 20-year-old Cabernet Sauvignon vineyard located in San Casciano (Florence, Italy) to assess cortical necrosis incidence and severity. A representative number of plants were selected, and unproductive spurs—characterized by extensive tissue necrosis and PCLS symptoms—were sampled to assess the association between spur mortality and the presence of *Diaporthe* spp. in the spurs wood. Fungal isolates obtained from the selected spurs were characterized morphologically and through a multi-

locus phylogenetic analyses based on different genomic regions: internal transcribed spacer (ITS), *translation elongation factor-1 α* (*EF1- α*), *beta-tubulin* (*TUB2*) and *calmodulin* (*CAL*) genes were used. Results indicate that in 2022 spur dieback reached 69% of the vines spurs, with an incidence of PCLS of approximately 96%, while in 2023 spur dieback was 56% and PCLS 77%, and in 2024 the values were 41% and 31%. Across all three years, the ratio of spur mortality related to PCLS incidence remained consistently around 71%, underscoring the pivotal role of *Diaporthe ampelina* inoculum in the manifestation of *Diaporthe* dieback in vineyards, even if also Botryosphaeriaceae species were isolated. This study demonstrates the significant contribution of *D. ampelina* inoculum in PCLS in spur dieback, emphasizing the need for targeted management strategies to mitigate inoculum-driven disease spread.

Recent advances on emerging vascular pathogens of grapevines in Eastern and Southern Mediterranean countries. E. CHOUERI¹, F. JREIJIRI¹, T. ELBEAINO², R. ABOU KUBAA³, P. SALAR⁴, X. FOISSAC⁴. ¹Department of Plant Protection, Lebanese Agricultural Research Institute, Tal Amara, P.O. Box 287, Zahlé, Lebanon. ²CIHEAM Bari, Istituto Agronomico Mediterraneo di Bari, Italy. ³Department of Plant Pathology, University of California, Davis, USA. ⁴University of Bordeaux, INRAE, Fruit Biology and Pathology, Villenave d'Ornon, France. E-MAIL: echoueiri@lari.gov.lb

Eastern and southern Mediterranean countries are illustrious for the cultivation of grapevine, since the old antiquity. Emerging vascular pathogens of grapevines, including viral and phytoplasma diseases, significantly reduced grape yield and quality, in addition to shortening the productive life of grapevine canes, which prompted to conduct field surveys in grapevine growing areas over the past few decades in Lebanon, Syria, Palestine, Jordan, Tunisia, Morocco, Algeria, and Egypt. Bright yellow leaves, yellow mosaic, veinal mottling, and the malformed shoots, were the most common symptoms caused by GFLV in the vineyards. ToRSV and ArMV were rarely reported producing symptoms that resemble those described for fanleaf. Leaf reddening/yellowing with leafroll symptoms was consistently observed in different areas, mainly affected by GLRaV-1, GLRaV-2 and GLRaV-3, whereas other relevant viruses such as GLRaV-4 strain 6, GLRaV-5 and GLRaV-7 were less frequent. Rugose wood, corky bark and GVA were the most observed among the infrequent cases. In addition to the presence of GVB and GRSPaV, few cases of GPGV,

GVL and GVD were also reported. Later, typical grapevine yellows (GY) symptoms were observed in Lebanon, Jordan and Syria caused by '*Candidatus* Phytoplasma solani' associated with grapevine bois noir, '*Candidatus* Phytoplasma omanense', '*Ca. P. aurantifolia*', '*Ca. P. asteris*' and clover proliferation group (16SrVI). Finally, the heavy widespread of mealybug infestations and the outbreak of GY epidemic could be a real risk in the Arab vineyard agroecosystems. The use of certified seedlings and monitoring strategies are essential to prevent spread of epidemic viruses and phytoplasmas.

Genome and genetic diversity of *Lasiodiplodia* species associated with Mango dieback and stem-end rot diseases in Côte d'Ivoire. Y.S. YEO^{1,2,3}, A. DEREPPER³, D. KONE^{1,2}, D. FERNANDEZ³. ¹UPR of Plant Physiology and Pathology, University Félix HOUPOUËT-BOIGNY, UFR Biosciences, 22 BP 582 Abidjan 22, Côte d'Ivoire. ²Wascal/African Center of Excellence in Climate Change, Biodiversity and Sustainable Agriculture, University Félix HOUPOUËT-BOIGNY, 01 BP V 34 Abidjan 01, Côte d'Ivoire. ³UMR PHIM (Plant Health Institute), Univ. Montpellier, IRD, CIRAD, INRAE, Institute Agro, Montpellier, France. E-MAIL: diana.fernandez@ird.fr

Mango (*Mangifera indica*) is a highly value fruit in the global export market. However, its production faces significant biotic and abiotic challenges, notably Mango Dieback (MD) and Stem-End Rot (SER), which affect orchard yield and postharvest quality. To investigate the prevalence and fungal diversity associated with these diseases, 92 mango orchards were randomly surveyed across four agro-ecological zones (AEZ) in Côte d'Ivoire during 2020 and 2021. Symptomatic mango organs were sampled, leading to the identification of 17 fungal genera based on morphological traits. Among these, *Lasiodiplodia* (*Botryosphaeriaceae* family) was the most prevalent, with 49% isolation frequency in MD and 74% in SER cases. Multilocus sequence analysis targeting the rDNA internal transcribed spacer (ITS) and the translation elongation factor 1-alpha (*tef1-α*) gene revealed six *Lasiodiplodia* species: *L. pseudotheobromae*, *L. euphorbicola*, *L. brasiliensis*, *L. caatinguensis*, *L. theobromae*, and *L. citricola* associated to MD and SER. Genotyping-by-Sequencing (GBS) combined with Principal Coordinate Analysis (PCoA) and sparse Non-Negative Matrix Factorization (sNMF) identified 4 distinct genetic groups among 150 isolates of the AEZ *Lasiodiplodia* populations. Species assignment by GBS did not fully match ITS and *tef1-α* phylogenetic assignment. Genome sequencing of representative *Lasiodiplodia* spp.

isolates using Illumina HiSeq and subsequent annotation through multiple public databases allow comparative analysis, revealing both shared and species-specific gene clusters. These findings will enhance our understanding of disease dynamics in Côte d'Ivoire and will provide insight into the molecular determinants of *Lasiodiplodia* virulence, paving the way for developing effective disease management strategies.

This research was financially supported by the Regional Scholarship and Innovation Fund (Rsif) programme of the Partnership for Skills in Applied Sciences, Engineering and Technology (PASET).

Trunk disease and dieback pathogens in Australian almond orchards. B.J. OSWALD^{1,2}, E.S. SCOTT³, L. CARVALHAIS², M.R. SOSNOWSKI^{1,3}, T.J. WIECHEL⁴, S. KREIDL⁴, L. TESORIERO⁵, O.A. AKINSANMI². ¹South Australian Research & Development Institute, GPO Box 397, SA 5001. ²The University of Queensland, Centre for Horticultural Science, QAAFI, Brisbane, Australia. ³School of Agriculture, Food and Wine, The University of Adelaide, SA 5005. ⁴Agriculture Victoria, AgriBio Centre, Bundoora, Victoria. ⁵NSW Department of Primary Industries, Ourimbah, Australia. E-MAIL: brittany.oswald@sa.gov.au

Several fungal species cause trunk diseases in almond trees. Common symptoms include necrosis of the wood, discolouration of vascular tissues, gumming, and dieback of limbs from the point of infection. Infection may kill primary branches and entire trees if left untreated. Samples collected from symptomatic trees across Australia from 2015 to 2025 yielded numerous fungal species. Fungi were identified using ITS, *TEF1α*, *β* tubulin and *GAPDH* gene regions as required. *Botryosphaeriaceae* species were the most prevalent trunk disease pathogens, with *Diplodia seriata* being the most common. Less commonly isolated species included *Botryosphaeria dothidea*, *Lasiodiplodia* spp., *Dothiorella* spp., and *Neofusicoccum* spp. Other genera isolated included *Colleto-trichum*, *Collophora*, *Cytospora*, as well as species of families *Diaporthaceae* and *Diatrypaceae*. The distribution of these trunk disease pathogens differed among the growing regions. Detached branch assays and in planta experiments have shown that 18 *Botryosphaeriaceae* and 7 *Diaporthaceae* isolates are pathogenic to cvs Price and Nonpareil. Experiments towards developing a better understanding of how extrinsic factors influence disease severity are ongoing. Water-deficit stress applied to young trees in controlled conditions had no significant effect on the susceptibility of cv. Nonpareil to *D. seriata*.

ta. The experiment is being repeated. Disease severity in detached branches inoculated with *D. seriata*, *Neofusicoccum australe* and *C. acutatum* varied with incubation temperature. Of the six almond cultivars used in the experiment, none was consistently less susceptible to any of the three pathogens. Results from this research are helping to improve our understanding of fungal trunk disease pathogens and the factors that influence disease severity.

Evaluation of selected fungal biological control agents for the protection of grapevine pruning wounds against *Diplodia seriata*. A. FLOUDAS¹, S. TESTEM-PASIS^{1,2}, A. FLARI¹, E. DIMOU¹, A. ELEFTHERIOU¹, G.S. KARAOGLANIDIS¹. ¹Aristotle University of Thessaloniki, Faculty of Agriculture, Forestry and Natural Environment, Plant Pathology Laboratory, Thessaloniki, Greece. ²Department of Agriculture, School of Agricultural Sciences, University of Western Macedonia, 53100 Florina, Greece. E-MAIL: gkarao@agro.auth.gr

Grapevine Trunk Diseases (GTDs) are among the most destructive grapevine diseases, causing significant economic losses due to vineyard decline and replanting costs. The lack of effective chemical measures highlights the urgent need for environmentally and consumer-friendly alternatives. Biological Control Agents (BCAs) offer a promising, sustainable approach, targeting multiple GTD-associated fungi. This field study was conducted in the viticultural region of Nemea to evaluate the efficacy of four BCAs - *Trichoderma atroviride*, *T. citrinoviride*, *T. ghanense*, *Talaromyces pinophilus*—and a mixture (*T. ghanense* + *T. pinophilus*) in controlling infections by *Diplodia seriata*, a key GTD pathogen in Greece. Two Greek grapevine cultivars with contrasting susceptibility - Roditis (susceptible) and Limnio (tolerant) were used. BCAs were applied as conidial suspensions on fresh pruning wounds, followed 24 hours later by artificial inoculation with *D. seriata*. Wounds were covered for two days to maintain moisture and promote infection. After six months, samples were collected and analyzed using a culture-based method. Pathogen reisolation was conducted on PDA medium, and all obtained fungal isolates were identified molecularly. The results revealed that *T. pinophilus* and its mixture with *T. ghanense* provided strong protection in the susceptible cultivar Roditis, substantially reducing pathogen recovery. Conversely, *T. atroviridae* was more effective in the tolerant cultivar Limnio, indicating a potential cultivar-specific interaction. All BCAs demonstrated the ability to colonize and per-

sist on pruning wounds. These findings reinforce the potential of BCAs as effective, sustainable tools for GTD management and support their integration into future vineyard protection strategies.

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CONCURRENT SESSION C1 – *Phytophthora*–prominent, emerging threats to crops and forest ecosystems

SESSION KEYNOTES

***Phytophthora* in different ecosystems of the Mediterranean Basin: agriculture, horticulture, forestry, natural environments, and emerging *Phytophthora* threats from other regions.** A. PEREZ-SIERRA¹, T. JUNG², M. HORTA JUNG², J. BAKONY³, D. COOKE⁴, T. DOĞMUŞ-LEHTIJÄRVI⁵, R. LAHLALI⁶, I. MILENKOVIC⁷, C. MORALES-RODRIGUEZ⁸, S. MORICCA⁹, J. OLIVA¹⁰, B. SCANU¹¹, A. SOLLA¹², A. VANNINI⁸, S. WOODWARD⁵, S.O. CACCIOLA¹³. ¹Instituto Valenciano de Investigaciones Agrarias (IVIA), Centro de Protección Vegetal y Biotecnología, Moncada, Valencia, Spain. ²Department of Forest Protection and Wildlife Management, *Phytophthora* Research Centre, Mendel University in Brno, 613 00 Brno, Czech Republic. ³HUN-REN Centre for Agricultural Research, Plant Protection Institute, Fehérvári str. 132-144, 1116 Budapest, Hungary. ⁴The James Hutton Institute, Plant Pathology, Invergowrie, Invergowrie, Dundee, United Kingdom of Great Britain and Northern Ireland, DD2 5DA. ⁵Isparta University of Applied Sciences, Yenisarbademli Vocational School, 32950, Isparta, Türkiye. ⁶Phytopathology Unit, Department of Plant Protection and Environment, Ecole Nationale d'Agriculture de Meknès, Km 10, Rte Haj Kaddour, BP S/40, Meknes 50001, Morocco. ⁷Faculty of Forestry, University of Belgrade, 11030 Belgrade, Serbia. ⁸Department for Innovation in Biological, Agro-food, and Forestry Systems, Tuscia University, Viterbo, Italy. ⁹Department of Agricultural, Food, Environmental and Forestry Science and Technology (DAGRI), Plant Pathology and Entomology Section, University of Florence, Piazzale delle Cascine 28, 50144 Florence, Italy. ¹⁰Department of Agricultural and Forest Sciences and Engineering, University of Lleida, Lleida, Spain. ¹¹Department of Agricultural Sciences, University of Sassari, Viale Italia 39A, 07100 Sassari, Italy. ¹²Faculty of Forestry, Universidad de Extremadura, Avenida Virgen del Puerto 2, 10600 Plasencia, Spain. ¹³Department of Agri-

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The oomycete *Phytophthora* is one of the most important genera in plant pathology, with 260 species currently known, 60 descriptions in progress and an estimated number of 400 to be discovered worldwide. The social and economic impacts of *Phytophthora* in agriculture, horticulture, forestry and its wider environment are significant. It can cause important losses in crops and plant production, alters the structure of forests and has a negative impact on the landscape and biodiversity of natural ecosystems. The Mediterranean Basin is particularly vulnerable with many important crops, endemic plants and biodiversity that are at risk to diseases caused by *Phytophthora*. Several studies have demonstrated that these pathogens move accidentally mainly with infected plant material and infested soil through worldwide trade, and they are introduced into new plantations or restoration projects with devastating consequences. Moreover, *Phytophthora* can be spread by natural water courses and irrigation water. Other drivers are the introduction into environments with highly susceptible hosts, intensive cultivation, stressed hosts, climate change or the combination of all of them. Usually, *Phytophthora* is studied in one type of system, either agriculture, horticulture or forestry, however, these systems are interconnected and the spread between systems threatens crop health, plant production and biodiversity simultaneously. In many cases, the devastating effects are caused by exotic *Phytophthora* species, which are introduced into new areas where they have not coevolved with the hosts, becoming invasive even if in their natural habitats are unnoticeable. Understanding the interactions between *Phytophthora* species, their hosts, and the environment is crucial to mitigating their impact. Data is needed to start understanding *Phytophthora* interactions across landscapes and, in recent years, major surveys have been carried out around the world to find the origin of some of the most damaging *Phytophthora* species. These studies indicate most *Phytophthora* species are not native to Europe. Moreover, they have resulted in the description of a high number of species that are present in different geographical areas, which could thrive and be a possible threat to plants in the Mediterranean Basin. Evidence suggests there are still many unknown species whose potential impact cannot be predictable, and biosecurity measures and practices are essential to minimize the risk of introduction and spread of new or already introduced species. There are important biosecurity challenges with asymptomatic dispersal of the pathogen

and the broad host ranges. Therefore, future research should focus on improving surveillance, early detection, advanced diagnostics, data sharing, crop breeding for resistance, environmentally friendly control measures and good practices to safeguard global plant health and biodiversity, with particular attention to vulnerable regions, such as the Mediterranean Basin.

***Phytophthora* in public gardens- risks and their mitigation (Phyto-gard EUPHRESKO project).**

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Public gardens are characterised by a wide range of plant species, including rare plant specimens and collections. They have a high volume of incoming, including imported, plant stock and increasing numbers of international visitors annually. All these factors increase the risk of introduction and transmission of potentially pathogenic *Phytophthora* species. The Euphresco 'Phyto-gard' project, involves ten international partners, aiming to establish the main *Phytophthora* spp. found in selected public gardens and risk factors associated with their distribution within the garden, including association with incoming plants. All partners in the project collect and analyse samples with the same set of shared protocols, previously developed within ID-Phyt. Soil, water and plant samples from two gardens and each of their nurseries, were collected by each collaborating partner over two years. Sample analysis consisted of DNA extraction from the samples and nested PCR to verify the presence (or absence) of *Phytophthora* DNA. All *Phytophthora*-

positive samples were processed using an established metabarcoding approach involving Illumina sequencing. Our results indicate the presence of *Phytophthora* spp. in both garden and nursery in more than half of the samples. We detected differences in the total number of Oomycetes found in each country as well as in the predominant *Phytophthora* spp., probably related to both climatic conditions and host species present. The abundance of Oomycetes and *Phytophthora* spp. was related to both plant-host genera and the type of sample. The present project provided new records of *Phytophthora* species in both plant and soil samples. Overall, this work showed that gardens are hot spots of *Phytophthora* diseases. Disease mitigation could be improved by both stakeholder and public awareness. Improvement of best practice throughout the process of plant procurement, planting and garden management, is a key factor to prevent *Phytophthora* diseases.

Early detection of *Phytophthora* in EU and third country nurseries and traded plants (ID-PHYT).

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Invasive *Phytophthora* pathogens are causing significant economic damage to agricultural, horticultural and forestry crops worldwide, as well as ecological damage to native plant species in wider environments. Traded plants are a well-documented pathway for *Phytophthora* pathogens, facilitating their spread both nationally and internationally. The goal of the ID-PHYT project was to

develop a co-ordinated strategy for the early detection of *Phytophthora* pathogens in plant nurseries and traded plants for planting across EU and third countries to inform best practice. Protocols for nursery sampling and detection of *Phytophthora* using an eDNA metabarcoding method plus a traditional baiting method were successfully shared and validated across project teams in six partner countries (FR, GB, GR, IE, IT, USA). The final nursery sampling dataset consisted of 1011 samples with replicates collected from thirteen plant nurseries across six countries. This included 647 root samples and 364 water samples with 627 samples analysed by baiting and 384 samples analysed by metabarcoding. Sample metadata and a set of key nursery management data were also collected for downstream analyses conducted using hierarchical Bayesian mixed models. A high diversity of *Phytophthora* (65 known *Phytophthora* species including quarantine-regulated species and some first country records) was detected across the 13 sampled nurseries. *Phytophthora* was found in the irrigation water at several of the nurseries highlighting water management as a key priority area for improvement. High risk hosts with consistent *Phytophthora* associations included *Fagus*, *Ligustrum*, *Thuja*, *Lavandula*, *Quercus* and *Choisya* spp. Other nursery risk factors which increased the likelihood of *Phytophthora*-positive samples included reliance on greater than 50% imported plant stock and growing a high diversity of plant genera. Analyses were also able to identify *Phytophthora* species' sensitivities to substrate (water versus root), nursery latitude and detection method (baiting versus metabarcoding) which assists understanding of their lifestyle and habit and sensitivity to detection method, facilitating further prediction of risk. One outcome of the project was the co-design of a concise, best-practice guidance document based on scientific evidence translated from English into partner country languages and disseminated through each country's trade association channels. The best practice guidance highlights key plant biosecurity considerations for growers and focuses on the need to understand high risk hosts and pathways, improved water management and plant growing conditions, awareness of symptoms and the importance of having staff trained in plant health knowledge.

ORAL PRESENTATIONS

***Phytophthora palmivora*: A Serious Challenge for South African Papaya Growers.** F. JAMI¹, W.J. BOTHA¹, M. SCHOEMAN², M. DANEEL². ¹Agricultural Research Council, Plant Health and Protection,

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Phytophthora palmivora is an Oomycete pathogen widespread in tropical and subtropical regions, causing severe diseases such as fruit rot, bud rot, blight, cankers, and root rot. In South Africa, papaya is grown by both smallholder and commercial farmers, particularly in the Limpopo and Mpumalanga provinces. In 2019, widespread symptoms of crown and root rot were observed in South African papaya orchards, leading to significant tree collapse and loss of fruit production. This study aimed to identify and characterize *Phytophthora* species infecting papaya in South Africa, as well as to investigate the distribution and aggressiveness of identified species. Samples were collected from six commercial orchards, and diseased stems and soil samples were analysed, resulting in isolation of only one *Phytophthora* species. Molecular identification using rDNA-ITS sequencing confirmed the species identification as *P. palmivora*, and pathogenicity tests revealed the pathogen's aggressiveness on papaya fruits. This is the first report of *P. palmivora* in South Africa, where it is a regulated pathogen. Given its destructive potential, *P. palmivora* poses a serious threat to the papaya industry and other crops such as macadamia, citrus, and avocado. Understanding the distribution of *P. palmivora* and its potential introduction routes is critical for implementing effective management strategies to protect South African agriculture from further damage.

Emerging *Phytophthora* species in natural and agricultural ecosystems: a case study of wild and cultivated olives in Sardinia, Italy. A. DEIDDA, A. BRANDANO, C. MORITTU, G.G.A. SATTA, B. SCANU. *Department of Agricultural Sciences, Viale Italia 39/a, University of Sassari, 07100 Sassari, Italy.* E-MAIL: adeidda2@uniss.it

The genus *Phytophthora* includes numerous species widely recognised as destructive pathogens in agricultural and natural ecosystems worldwide. Several species have recently emerged as major threats to agricultural tree crops, particularly in areas with Mediterranean and semi-arid climates. Among these, cultivated olive has increasingly suffered from wilting and mortality in several olive-growing countries. Since 2022, severe decline and mortality of wild olive trees, traditionally used as rootstocks for cultivated olive, have been observed in Sardinia (Italy). Affected trees exhibited leaf chlorosis, wilting, shoot blight, defoliation and root necroses. Mul-

tipale *Phytophthora* taxa were isolated from rhizosphere soil and their pathogenicity on wild olive was confirmed. Similarly, since 2010, *Phytophthora*-induced decline has been reported in cultivated olive in the region, particularly in newly established commercial plantations. In addition to crown symptoms resembling those on wild olive, trees exhibited root and collar rot, with necrotic lesions often girdling the stem. Based on morphological and molecular analyses, isolates from infected tissues were identified as *P. heterospora*, a sister species of *P. palmivora*. More recently the number of *Phytophthora* species detected from young olive groves increased, including *P. citrophthora*, *P. inundata*, *P. nicotianae* and *P. niederhauserii*. Notably, *P. niederhauserii* and *P. citrophthora* were also detected in potted olive saplings coming from nurseries and ready for outplanting. These findings provide novel insights into the diversity and impact of *Phytophthora* species on wild and cultivated olives, highlighting the role of intensified cultivation practices and uncontrolled plant movement driving the spread of these pathogens.

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Influence of environmental and biological factors on *Phytophthora acerina* pathogenicity in almonds. B.J. OSWALD^{1,2}, E.S. SCOTT³, L. CARVALHAIS², M.R. SOSNOWSKI^{1,3}, O.A. AKINSANMI². ¹South Australian Research & Development Institute, GPO Box 397, SA 5001. ²The University of Queensland, Centre for Horticultural Science, QAAFI, Brisbane, Australia. ³School of Agriculture, Food and Wine, The University of Adelaide, SA 5005. E-MAIL: brittany.oswald@sa.gov.au

Phytophthora root rot, crown rot and trunk cankers in almond trees are caused by several *Phytophthora* spp., which are widespread in Australian orchards. Symptoms include limb dieback, chlorosis, gummosis, woody tissue discolouration and necrosis, root necrosis, tree decline and death, ultimately resulting in significant loss. *Phytophthora acerina* was the most commonly identified species, most frequently isolated from the trunks of mature trees. Other isolated species included *P. cactorum*, *P. hedraiaandra*, *P. multivora* and *P. niederhauserii*. Current research aims to understand how biotic and abiotic factors influence disease severity. Experiments assessed the susceptibility of commonly planted rootstocks and almond cultivars to *P. acerina*, and the

influence of water-deficit, temperature, pruning, and carbohydrate and lignin content on disease severity. These investigations revealed that although there was some variation, all almond cultivars tested were highly susceptible to infection. Lesions caused by *P. acerina* were shorter in trees subjected to deficit irrigation and longest at 30°C, compared to well-watered trees and lower temperatures, respectively. Further experiments showed that grafted nursery material subjected to pruning was more susceptible to colonisation by *P. acerina* than non-pruned trees. Experiments investigating rootstock susceptibility are underway, and compositional analysis is ongoing to determine how carbohydrates and lignin influence colonisation in almond wood, and results will be presented. An improved understanding of the effect of these factors on the severity of *Phytophthora* diseases in almonds will aid in developing effective management strategies to enhance orchard longevity and profitability.

Detection of *Phytophthora* and other oomycete pathogens in UK peat-free growing media and implications for plant health. D. FREDERICKSON MATIKA¹, S. GREEN¹, D. CRISP¹, K. SCHIFFER-FORSYTH², M. ELLIOT³, P.J.A. COCK⁴. ¹Forest Research, Northern Research Station, Roslin, Midlothian, EH25 9SY, UK; ²INRAE Grand Est-Nancy, 54280 Champenoux, France; ³Royal Botanic Gardens Edinburgh, 20a Inverleith Row, Edinburgh, EH3 5LR; ⁴Strathclyde Institute of Pharmacy & Biomedical Sciences (SIPBS), University of Strathclyde, Glasgow, G4 0RE, UK. E-MAIL: debbie.frederickson@forestryresearch.gov.uk

Peat-based growing media products are being phased out in the UK. Alternative constituents may be at elevated risk of harbouring oomycete plant pathogens. Thirty-six peat-free growing media samples were tested for oomycete pathogens by baiting and DNA metabarcoding. As evidence of live pathogens present, one *Phytophthora* species, *P. bilorbang*, was detected by baiting of wood chips and bark, and eight species of *Pythium*, *Phytophthium* and *Elongisporangium*, including four species not previously reported in the UK, were also baited. DNA of eleven *Phytophthora* species were detected by Illumina metabarcoding of the ITS1 across a range of samples including *P. citrophthora*, *P. cactorum* and *P. idaei* in compost mixes and *P. citrophthora* and *P. cinnamomi* in bark. Coir samples were found to contain DNA of *P. crassamura*/*megasperma* and also *Peronosclerospora*, a genus of tropical downy mildew pathogens. Of particular concern, DNA of six *Phytophthora* pathogens, including *P. bishii*, a pathogen of strawberry not previously report-

ed in the UK, were detected in imported, recycled coir previously used for strawberry production. We discuss the implications of the species detected for plant health and the need for further evidence-gathering to strengthen biosecurity.

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Silent Invaders: unmasking *Phytophthora* diversity in Mediterranean nurseries and gardens.

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Oomycete diseases, especially those caused by *Phytophthora* species, present a serious and growing threat to nurseries and gardens worldwide, often spreading silently through asymptomatic plants. With nearly 200 recognized species and an exceptionally wide host range, *Phytophthora* is behind some of the most destructive plant diseases globally. From 2021 to 2023, an extensive survey across Greece aimed to map the incidence and diversity of *Phytophthora* species in both symptomatic and asymptomatic plants. A total of 2,607 samples—including rhizosphere soil and irrigation water from ponds and canals—were collected from 34 plant genera, sourced from nurseries in Greece, France, and Italy, as well as public and private Greek gardens. Baiting assays produced 763 oomycete isolates, from which 129 were selected according to specific criteria for molecular identification. Targeting three genetic loci, 99 strains were confirmed as *Phytophthora* spp., representing 11 species and one interspecific hybrid. An additional 27 isolates were classified as *Phytophthium* spp., and three as *Pythium* spp. *Phytophthora nicotianae* emerged as the most prevalent species (35.36%), isolated from the rhizosphere of 20 host plants. Other species, such as *P. virginiana*, *P. parsiana*, *P. lacustris*, and the hybrid *P. lacustris* × *riparia*, were found exclusively in irrigation water. ITS motif

analysis revealed that even single nucleotide polymorphisms can effectively distinguish among *Phytophthora* species. Furthermore, under the Euphresco project “Phyto-gard” and national research initiatives, high-throughput sequencing (NGS) targeting the ITS region was performed on 163 samples, greatly enhancing detection sensitivity and offering a broader, more comprehensive view of oomycete diversity.

Survival of oomycetes and fungi during composing of horticultural waste: implications for compost biosafety. L. GUIDONI, A. VANNINI, C. MORALES-RODRIGUEZ. *Department for Innovation in Biological, Agro-food and Forest Systems (DIBAF), University of Tuscia, 01100 Viterbo, Italy.* E-MAIL: vannini@unitus.it

European countries are reducing peat use in horticulture due to its environmental impact. Research has demonstrated that compost can effectively replace peat without compromising the growth and development of potted horticultural plants. However, alternatives must meet strict quality standards, particularly regarding the risk of contamination by plant pathogens. Reliable diagnostic methods are essential to assess the survival of pathogens during composting. This study evaluated the survival of *Phytophthora cinnamomi*, a model plant pathogen, in woody chips from artificially inoculated *Castanea sativa* saplings used as bulking agents in composting. Detection methods included pure culture isolation, quantitative PCR (qPCR), and High Throughput Sequencing (HTS). At the start of composting, the pathogen was easily detected using baiting and molecular barcoding. By the end of the maturation phase, *P. cinnamomi* was no longer detectable by any method, indicating that effective composting can eradicate the pathogen. HTS also identified DNA from plant pathogenic fungal genera naturally present in green waste and bulking materials throughout the composting process. These findings support the development of diagnostic pipelines for compost biosafety certification. Overall, the study demonstrates that high-quality, pathogen-free compost can be produced from recycled horticultural waste without the need for complex or expensive composting infrastructure.

Unveiling intraspecific variability and population structure of the oak-pathogen *Phytophthora quercina* using genotyping-by-sequencing. A. BRANDANO¹, M. MULLETT^{1,2}, T. JUNG², M. HORTA-JUNG², A. DEIDDA¹, B. SCANU¹. ¹*Department of Agricultural Sciences,*

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Phytophthora quercina is an important root pathogen of oak trees. Its geographic distribution seems to be limited to Europe, although there have been reports in Asia Minor, North Africa and America. In this study a total of 75 isolates of *P. quercina* were selected to investigate its population structure and the outcrossing level through genotyping-by-sequencing (GBS) analysis. The STRUCTURE analysis suggested that eight clusters best fit the dataset, whereas the Bayesian Information Criterion from K-means clustering in Discriminant Analysis of Principal Components (DAPC) supported five groups. Among these, some clusters were consistent in sharing the same host (e.g. *Quercus suber*), while others had the same geographic origin (e.g. Portugal) but different hosts. Some clonal lineages were consistent sharing both the same host and geographic origin (e.g. *Q. suber* – Sardinia, Italy; *Q. suber* – Spain; or *Quercus robur* – Central Europe). Network analysis and the index of association show that this homothallic, selectively inbreeding *Phytophthora* species has undergone a significant amount of outcrossing, which may have generated distinct subgroups of admixed individuals. The estimation of the degree of linkage disequilibrium revealed that the results for these populations were significantly lower than those for a clonal. This study provides new insights into the phylogeography and evolutionary history of *P. quercina*, which seem to be mainly driven by the diversification of host oak species.

Big problems, big solutions: holistic approach and Integrated Pest Management for *Phytophthora cinnamomi* in the FAGESOS Project. C. MORALES-RODRIGUEZ, D. LIBERATI, R. CACCIA, A. VANNINI. *Department for Innovation in Biological, agro-food and forest systems (DIBAF) Tuscia University. Via San Camillo de Lellis snc, VT, Italy.* E-MAIL: cmorales@unitus.it

Phytophthora cinnamomi is a highly invasive soil-borne oomycete responsible for root rot in a wide range of woody species. In Mediterranean ecosystems, it poses a serious threat to biodiversity and forest stability, particularly affecting *Quercus ilex*, *Q. suber*, and *Castanea sativa*. The pathogen is facilitated by rising temperatures and changing precipitation patterns, which expand its geographic range and intensify disease outbreaks. In

September 2022, the European 101074466—LIFE21-CCA- IT-LIFE FAGESOS project was launched. The project has among its objectives the development of IPM protocols for the mitigation of damage caused by *P. cinnamomi* in *Quercus ilex*, *Q. suber*, and *C. sativa* as well as its large-scale application. By the end of the project, we expect to treat up to 1,070 ha (7 demonstrative sites) and to protect up to 18,119 ha of vulnerable areas in total. The project adopts a holistic and ecosystem-based approach, combining chemical, biological, and cultural practices over two consecutive years of treatment. In 2024, the first treatment cycle was applied across seven demonstration sites in different Mediterranean forest contexts. The protocol focuses on reducing pathogen inoculum in the soil, enhancing populations of beneficial microorganisms, and stimulating the plant's natural defense mechanisms. The contribution will present preliminary results of this initial phase. The LIFE FAGESOS project offers a model for integrated forest health management, promoting resilience against emerging diseases in the context of climate change.

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POSTER PRESENTATIONS

Disentangling the influence of Mediterranean *Quercus* species on *Phytophthora cinnamomi* infectivity. R. LÓPEZ, M.T. HIDALGO, M. GARCÍA, M. S. SER-RANO. *Department of Agronomy, Campus de Rabanales Edif. C-4, University of Córdoba, Córdoba, 14071, Spain.* E-MAIL: a12semom@uco.es

Mediterranean oak forests exhibit high *Quercus* species diversity with varying response against the pathogen *Phytophthora cinnamomi* (*Pc*). Recent studies using excised twigs inoculation have shown that *Q. pyrenaica* (*Qp*) was as sensitive to *Pc* as *Q. ilex* (*Qi*), whereas *Q. coccifera* (*Qc*) and *Q. faginea* (*Qf*) appeared more tolerant to infection. However, little is known about how these species could influence on *Pc* infective capacity. To address this, the ability of *Qc*, *Qf*, and *Qp* root exudates to stimulate zoospore release of *Pc* was assessed through *in vitro* experiments, using *Qi* as a reference. Aerial mycelium of *Pc* grown in Pea- Broth was transferred to glass beakers containing sterilized saline solution to stimulate sporangial production. Seedling rootlets from each species were immersed into the liquid and incubated in the dark at 23°C for 48 hours. Three beakers with one plant each

were prepared per species along with three more beakers without plants as controls. Zoospore production was quantified in a Neubauer chamber. The full experiment was repeated twice. Results showed that *Qi*, *Qc* and *Qp* induced significantly higher zoospores production than the controls whereas *Qf* did not. Additionally, although all species showed a similar effect to *Qi*, the infectivity of *Pc* was significantly less stimulated by *Qf* compared to *Qp*. These findings suggest that the high ability to induce *Pc* infective zoospores exhibited by sensitive *Quercus* species and even by the tolerant *Qc*, puts the survival of Mediterranean oak forests at greater risk.

Characterization of *Phytophthora* and *Pythium* species causing root rot in olive trees in Morocco. I. LEG-RIFI^{1,2}, A. LAZRAQ², S. AMIRI¹, A. TAHIRI¹, R. LAHL-ALI¹. ¹*Phytopathology Unit, Department of Plant Protection, Ecole Nationale d'Agriculture de Meknès, Km 10, Rte Haj Kaddour, BP S/40, Meknes 50001, Morocco.* ²*Laboratory of Functional Ecology and Environmental Engineering, Sidi Mohamed Ben Abdellah University, Route d'Imouzzer, P.O. Box 2202, Fez 30000, Morocco.* E-MAIL: rlahlali@enameknes.ac.ma

The olive tree (*Olea europaea*) is a vital fruit crop in Morocco; however, a widespread decline due to root rot has been observed in several regions. This study aimed to identify and characterize the oomycete species associated with olive root rot. During the 2021 and 2022 growing seasons, symptomatic root tissues and soil samples were collected, leading to the identification of 10 oomycete species through morphological characterization and sequencing of the internal transcribed spacer (ITS) region of rDNA. Seven species belonged to *Phytophthora* (*P. palmivora*, *P. plurivora*, *P. acerina*, *P. oleae*, *P. cactorum*, *P. gonapodyides*, and *P. megasperma*), while three species were from *Pythium sensu lato* (*P. schmitthenneri*, *P. aphanidermatum*, and *P. irregulare*). A pathogenicity assay using soil inoculation on one-year-old olive seedlings (cv. Picholine Marocaine) revealed that all species were pathogenic, causing root rot, vascular discoloration, and wilting, with successful re-isolation of the pathogens from necrotic roots, fulfilling Koch's postulates. These findings highlight the complex etiology of olive root rot, as multiple species induce similar symptoms, and represent the first detailed report of *Phytophthora* and *Pythium s.l.* species associated with this disease in Morocco.

Gene expression dynamics in olive drupes (*Olea europaea* L.) under *Phytophthora oleae* infection: RNA-Seq

insights for sustainable management. F. LA SPADA¹, S. CONTI TAGUALI¹, M. RIOLO¹, R. PARLASCINO¹, C. BUA¹, M.C. TAMBE¹, A. PANE¹, G. DIONISIO², S.O. CACCIOLA². ¹Department of Agriculture, Food and Environment Di3A, University of Catania, Via Santa Sofia 100, 95123 Catania, Italy. ²Department of Agroecology, Research Center Flakkebjerg, Aarhus University, Slagelse, 4200, Denmark. E-MAIL: olga.cacciola@unict.it

Rot of drupes caused by *Phytophthora oleae* is an emerging disease of olive (*Olea europaea* L.) in humid environments. This study assessed the effectiveness of two biocontrol means, a culture filtrate of *Trichoderma atroviride* and a biocontrol agent (BCA), a cell suspension of the yeast *Candida oleophila*, in controlling this disease. To better understand the mechanism of action of these two biocontrol means, the transcriptional reprogramming of the host as a consequence of *P. oleae* infection and treatments with either *T. atroviride*-culture filtrate or *C. oleophila* cells was investigated. Both treatments significantly reduced disease severity compared to the untreated control, with a reduction in the relative AUDPC of 52 and 56 percent, respectively. Transcriptomic analyses of olive drupes revealed noticeable changes of gene expression following pathogen infection and biocontrol applications. Infection with *P. oleae* alone modulated the expression of more than 2,200 genes, many of which were linked to defense regulation mechanisms, oxidative stress response, and hormonal signalling pathways. Treatment of non-infected drupes with either biocontrol means modulated genes involved in immune priming, stress responses, and cell wall remodelling. When applied to *P. oleae* infected drupes, both treatments influenced key defense related pathways, including jasmonic acid signaling and reactive oxygen species regulation, suggesting enhanced or redirected defense mechanisms. These findings highlight the bi-directional mode of action of both *T. atroviride* culture filtrate and *C. oleophila* as biocontrol means of rot of olive drupes, i.e. suppressing the disease development and modulating the expression of host genes promoting disease resistance.

This research was inspired by the projects “Nuove soluzioni tecnologiche per la filiera degli agrumi – NewCitrusTech” (cod. 1.3.1, Misura 19 PSR 2014/2022 – CLLD GAL Eoro) and “Azioni Innovative per la Produttività del Distretto dell’Ortofrutta di Qualità – INNOVAPROD” (cod. 1.3.2, Misura 19 PSR 2014/2022 CLLD).

Biotic and abiotic factors involved in kiwifruit decline syndrome (KVDS) in Spain. A. PÉREZ-SIERRA,

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Since 2020 decline symptoms have been observed in kiwifruit plantations in Valencia (Spain). Kiwifruit plants exhibited progressive dieback and died during summer. Initial investigations showed that a root rot was always associated with the decline. The symptoms were consistent with Kiwifruit Vine Decline Syndrome (KVDS), a disease of unknown aetiology affecting kiwifruit production in Italy since 2012. The aim of this study was to investigate the cause of the decline. A survey was conducted in 2024 in seven plantations where decline symptoms were detected. Soil and roots samples were collected. Isolations were performed on agar media and using baiting techniques and. In total, nine genera of fungi were recovered from the roots and four genera of oomycetes were recovered from roots and soil. The results were in agreement with the results obtained in Italy suggesting that the decline observed in Valencia could be the KVDS previously observed in Italy. As oomycetes have been shown to play an important role on the decline in Italy, an initial pathogenicity test was performed with four of the oomycetes detected: *Phytophthora nicotianae* and three *Phytophythium* species: *Ph. heliciodes*, *Ph. litorale* and *Ph. vexans*. One year old plants of two different rootstocks (*A. deliciosa* and *A. macrosperma*) were used. The results obtained indicated that this is a complex disease where abiotic factors could also be involved, and it is critical to identify the cause of this decline to develop appropriate control measures and to prevent the further spreading of the KVDS to new areas.

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***Phytophthora hedraiaandra* on the move: detection and characterization on imported *Hydrangea macrophylla* in Apulia, Southern Italy.** M. CARLUCCI¹, C. CARBOTTI¹, P.G. LUCCHESI¹, S. CONVERTINI^{2,3}, A. PACIFICO, F. NIGRO¹. ¹Department of Soil, Plant and Food Sciences (DiSSPA), Via Giovanni Amendola 165/A, University of Bari - Aldo Moro, Bari, 70126, Italy. ²Reagri srl, via Chiatona 62, 74016 Massafra (TA). ³Agrin scarl, via xxv Aprile 13, 36055 Nove (VI). E-MAIL: franco.nigro@uniba.it

The oomycete genus *Phytophthora* comprises highly destructive primary plant pathogens with significant economic impact. Moreover, *Phytophthora* species are responsible for extensive losses in ornamental plant production worldwide. Global trade facilitates the spread of these pathogens and the emergence of new hybrids suggests that novel *Phytophthora* species are evolving, likely in nursery environments where related but geographically isolated species come into contact. In spring 2024, several cases of decline and mortality of *Hydrangea macrophylla* were observed in nurseries in Apulia (southeastern Italy) on plants imported from Germany. Symptoms included collar and root rot, gradual wilting, leaf chlorosis, and stem necrosis. Isolations performed on potato dextrose agar (PDA) yielded colonies morphologically referable to *Phytophthora* spp., characterized by slow-growing, rosaceous to petaloid colony patterns on PDA, and the production of globose to broadly ovoid, caducous, papillate sporangia with short pedicels, typical of *P. hedraiaandra*. Multilocus phylogenetic analysis of two isolates, based on ITS, TEF1 α , TUB2, and COX1 sequences, confirmed their identity as *Phytophthora hedraiaandra*. Pathogenicity tests conducted by artificial inoculation of three-year-old *H. macrophylla* plants fulfilled Koch's postulates. The introduction of this pathogen through imported plants highlights the urgent need for the production of certified plant material, enhanced surveillance at points of entry, and improved local propagation practices to reduce the risk of introducing new invasive pathogens.

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Essential oils against *Phytophthora infestans*: A natural alternative to prevent potato from late blight disease. F. MARTINI^{1,2,3}, J. MUCHEMBLE², C. BURGEON^{1,4}, E. GONTIER³, M.H. JIJAKLI⁴, M-L. FAU-CONNIER¹. ¹Laboratory of Chemistry of Natural Molecules, UMRt BioEcoAgro INRAE-1158, Gembloux Agro-Bio Tech, Liege University, Passage des Déportés 2, 5030 Gembloux, Belgium. ²Plant Pathology and Biocontrol Team, UMRt BioEcoAgro INRAE-1158, ISA JUNIA, 2 rue Norbert Ségard, 59000 Lille, France. ³Laboratory of Plant Biology and Innovation, BIOPI-UPJV, UMRt BioEcoAgro

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The intensive use of synthetic and mineral (notably copper sulfate) pesticides depletes resources, pollutes water and threatens human health and ecosystems. As such, new and more sustainable alternatives for crop protection are urgently needed. This study explores the use of essential oils (EOs) – plant secondary metabolites known for their diverse biological activities – as natural alternatives for controlling potato late blight, a disease caused by the oomycete *Phytophthora infestans*. For this purpose, several EOs were chemically analysed, emulsified in water with Tween 20 and tested *in vitro* against three *P. infestans* genotypes (EU-13-A2, EU-36-A2, EU-37-A2). Oils rich in phenylpropanoids such as oregano, clove or cinnamon completely inhibited both mycelium growth and spore germination from 1 $\mu\text{L mL}^{-1}$, more efficiently than terpene-rich oils (bergamot, rosemary). *Ex vivo* assays on potato leaves confirmed the increasing susceptibility of three different potato varieties (Bintje, Fontane, Carolus) known to exhibit respectively no, low and high resistance against the disease. This highlights how varietal selection plays another key role in disease management. Phytotoxicity assessments showed no visible lesions or significant reduction in chlorophyll fluorescence caused by any EO under 2.5 $\mu\text{L mL}^{-1}$, although chlorosis was observed at higher concentrations. *In planta* experiments are currently underway to evaluate the protective properties of essential oils together with how they could induce plant systemic resistance. Together, these findings, together with deep comprehension of cellular sites and modes of action, will reveal promising EO for the development of effective and non-toxic biofungicides, paving the way for future applications in sustainable management of late blight.

CONCURRENT SESSION C2 – Major diseases that affect economic crops in the Arab and Mediterranean countries

SESSION KEYNOTES

Prevalence of emerging vascular diseases of stone fruit trees in Arab countries and progress in control strategies. E. CHOUEIRI. Department of Plant Protection, Lebanese Agricultural Research Institute, Tal Amara, P.O. Box 287, Zahlé, Lebanon. E-MAIL: echoueiri@lari.gov.lb

Stone fruit trees are a major industry in the Arab region characterized by climatic conditions favorable for fruit species providing economic, social and environmental services. Emerging vascular diseases had great influence on the quality and yield of these crops. Over 10 viruses have been reported in the Arab countries to affect stone fruits with a wide range of symptoms that consisted of mosaic, line patterns, mottling, yellow spots, distortion of the leaves, leaf shot-hole, fruit abnormalities, etc. The more common viruses and viroids detected in most Arab countries to infect stone fruits were PNRSV, PDV, ApMV, ACLSV, PPV, PLMVd and HSVd, whereas other viruses such as ApLV, APLPV, PBNSPaV were less widespread. Phytoplasma-like symptoms such as leaf yellowing/reddening and rolling, and witches' broom were observed, where '*Candidatus* Phytoplasma solani' infected plum, cherry and peach and '*Ca. P. omanense*' on plum in Jordan. In peach and plum orchards, the disease incidence ranged from 25% to 55% and from 15% to 55%, respectively, with a disease incidence of around 60% in sweet cherry. In addition, disease symptoms resembled those caused by phytoplasmas confirmed the presence of aster yellows phytoplasma (16SrI) affecting peach trees. In Egypt, European stone fruit yellows (ESFY) producing leaf curling, yellowing and fruit malformation caused serious economical losses on apricots and peaches. In Lebanon, almond witches' broom associated with '*Ca. P. Phoenicium*' destroyed more than 100,000 of almond and peach trees. Proliferation of slender shoots at several points on the main trunk of affected trees, or from the roots appeared on almond trees. Witches' broom and proliferation, perpendicular development of many auxiliary buds on the branches, leaf size reduction and yellowing (pale green), phyllody, stunted growth with affected fruit quality and yield reduction reaching 70%-100% and general decline of affected trees were also encountered. Intensified symptoms appeared after excessive pruning of infected trees. Infected almond trees generally die 3 to 4 years after the appearance of the first symptoms. Recently, *Xylella fastidiosa* has been reported on almonds with symptoms such as leaf scorch in Lebanon. However, monitoring efforts for *X. fastidiosa* in Palestine, Jordan and Morocco on stone fruit trees have been conducted, providing confirmation that these important horticultural crops are still free of *X. fastidiosa*. Control strategies have been initiated through several practices such as prohibiting imports of plant material from high risk countries, production of pathogen-free propagation materials, avoiding grafting scions from infected trees and strengthening human capacity in pathogens diagnosis. Large-scale surveys are

of utmost importance to detect newly established outbreaks, particularly for diseases that can be transmitted by insect vectors. Unfortunately, in most Arab countries, information on natural vectors of emerging vector-borne diseases is still limited or incomplete. Intensive training of technical staff and surveys to investigate the identity and distribution of the insect vectors and their natural hosts all over these countries is essential. Field surveys and continuous monitoring to assess the sanitary status of stone fruit crops and implement appropriate measures to limit the spread of these emerging vascular diseases is badly needed.

Management of major viral and fungal diseases affecting temperate food legume crops in the Arab and Mediterranean countries. S. G. KUMARI¹, S.-A. KEMAL². ¹International Center for Agricultural Research in the Dry Areas (ICARDA), Terbol Station, Beqa'a Valley, Zahle, Lebanon. ²ICARDA, Rabat, Morocco. E-MAIL: s.kumari@cgiar.org

The production, productivity and quality of temperate food legumes (faba bean, Kabuli chickpea, lentil, and field pea) are affected by several viral and fungal diseases in the Arab and Mediterranean countries. Surveys conducted over the past three decades have shown that the most important diseases affecting food legumes are wilt/root rot disease complex caused by *Fusarium* spp., *Rhizoctonia* and *Pythium* spp., and foliar diseases. In a few countries, parasitic nematodes (*Heterodera* and *Pratylenchus* spp.), and Ascochyta blights (*Didymella* spp.) are the major diseases of chickpea, faba bean and lentil, whereas faba bean suffers from chocolate spot (*Botrytis fabae*) and rust (*Uromyces viciae-fabae*). Viruses are emerging production constraints of food legumes where Faba bean necrotic yellows virus (FBNYV), Chickpea chlorotic stunt virus (CpCSV), Beet western yellows virus (BWYV), Bean yellow mosaic virus (BYMV), and Pea seed-borne mosaic virus (PSBMV) are the major ones. These viruses are affecting food legumes individually or as mixed infections. The importance of food legume viruses is mainly associated with increased in aphid activities due to changes in climate and cropping system in the regions. A positive linear correlation has been observed between virus prevalence and aphids that can transmit these viruses. The main aphid species in legume fields were *Acyrtosiphon pisum* (Harris), *Aphis craccivora* Koch. and *Aphis fabae* Scopoli. In addition, several wild species (annual or perennial) were found infected with these viruses and may play an important role in the spread of these viruses. In recent

years, virus epidemics have been reported in some Arab countries (such as Egypt, Tunisia, Syria, and Jordan), sometimes causing considerable yield reduction. Significant progress has been made in managing fungal diseases by integrating two or more management options, such as development resistant varieties, production of healthy seeds, adjusting planting dates, seed treatments, selective use of fungicide sprays, and cultural practices that reduce the impact on food legume productivity and quality. Over the past two decades, faba bean genotypes resistant to FBNYV and BLRV, chickpea genotypes resistant to CpCSV and BWYV, and lentil genotypes resistant to FBNYV, BLRV and PSbMV have been successfully identified. In addition, a relatively quick and simple plastic house technique was developed to identify resistant genotypes on the basis of relative virus movement and multiplication using Tissue blot immunoassay (TBIA). However, management options remain limited for emerging viruses, the emergence of virulent pathogens mainly for *Ascochyta* blight on chickpea, and complex soil-borne diseases caused by several parasitic pathogens and nematodes. Further efforts are still needed to develop varieties with multiple disease resistance, integrate new management options supported by digital innovations, establishing regional networks for food legume disease research for development, capacity development, and utilize modern tools, such as diagnostic tools, AI-based early warning and detection systems. Furthermore, more farmer-led participatory research for development is needed to effectively address the key plant health challenges arising from climate and farming systems changes facing food legume production systems in the Arab and Mediterranean countries which is leading to cereal monocropping.

ORAL PRESENTATIONS

Peacock's eye disease of olive, do we finally understand how to manage it effectively? D. EZRA¹, D. YGZAO^{1,2}, Y. GILAT^{1,2}, D. SHTIENBERG¹. ¹*Department of Plant Pathology and Weed Research, Institute of Plant Protection, Agricultural Research Organization – Volcani Institute, Rishon LeZion, 7528809 Israel.* ²*Robert H. Smith Faculty of Agriculture, Food and Environment, the Hebrew University of Jerusalem, Rehovot, Israel.* E-MAIL: dezra@volcani.agri.gov.il

Peacock's eye disease, also known as olive leaf spot, is caused by the fungus *Venturia oleaginea* and affects susceptible olive varieties globally. The disease symptoms appear as dark green to black spots on leaves, leading to

defoliation, twig desiccation, and significantly reduced tree productivity in subsequent seasons. Traditionally, chemical fungicides, mainly copper-based compounds, are applied to manage this disease. However, in recent years, olive growers in Israel have experienced severe disease outbreaks despite frequent fungicide applications (five to eight times per season). Our research aimed to develop a decision support system (DSS) to implement effective control measures against peacock's eye disease in Israel. We investigated the pathogen's biology and epidemiology, focusing on environmental conditions conducive to disease development. Our findings and the result of chemical control experiments we conducted over the past five years, led to the development of a DSS we named "Tavazit". This system facilitates precise and limited fungicide applications based on specific temperature and rainfall events, optimizing disease management strategies. Proper treatment for disease control based on the "Tavazit" DSS, over subsequent years, increased both the quantity of fruit yield and the quality of oil obtained from them, compared to untreated control trees. Implementing such DSS approaches aligns with integrated pest management principles, promoting sustainable agriculture by reducing unnecessary chemical usage and mitigating environmental impact.

This research was partially financially supported by the Israeli Council of Fruit Trees and by the Chief Scientist Ministry of Agriculture and Food Security.

Deciphering the European vine code: phenotypic and genetic insights into resistance to mildew. G. MADDALENA¹, V. RICCIARDI², O. FAILLA¹, G. DE LORENZIS¹, S.L. TOFFOLATTI¹. ¹*Department of DiS-AA, Via G. Celoria 2, Università degli Studi di Milano, Milano, MI 20133, Italy.* ²*IBBR, National Research Council CNR, Via Ugo La Malfa, 153, 90146 Palermo, PA, Italy.* E-MAIL: giuliana.maddalena@unimi.it

The sustainable management of grapevine diseases, such as downy mildew and powdery mildew, remains a major challenge for viticulture. Breeding varieties with genetic resistance provides an effective strategy for reducing chemical inputs. Although resistance loci to downy mildew (*Rpv*) and powdery mildew (*Ren/Run*) have primarily been introgressed from wild North American and Asian *Vitis* species, their use in breeding programs often results in the introduction of undesirable traits that compromise wine quality. Despite genetic erosion caused by human selection, exploring resistance within the Eurasian grapevine (*Vitis vinifera*) offers a promising alter-

native for breeding programs. Recent analysis has shown the potential of *V. vinifera* accessions from the Caucasus, which harbor a rich biodiversity, as source of disease resistance without compromising oenological quality. In this study, 88 *V. vinifera* accessions (wild and cultivated) from the Caucasus and neighboring regions were phenotypically evaluated for resistance to downy and powdery mildews over three consecutive years. Phenotypic screening allowed the identification of seven downy mildew-resistant and 31 powdery mildew-resistant accessions. A Genome-Wide Association Study (GWAS) performed on selected individuals revealed four novel resistance loci: *Rpv36* and *Rpv37* (downy mildew) and *Ren14* and *Ren15* (powdery mildew), located in genomic regions enriched with biotic stress response genes. These findings identify key targets for breeding disease-resistant grapevines within the *V. vinifera* species. Advanced tools like CRISPR and RNA interference could further enhance these loci potential, reducing chemical use and supporting sustainable viticulture. Ultimately, this approach promises lower production costs and improved environmental sustainability in the wine industry.

This research was financially supported by the European Union within the project titled 'FREECLIMB – Fruit Crops Adaptation To Climate Change In The Mediterranean Basin' under the Program Partnership For Research And Innovation In The Mediterranean Area (PRIMA), and the project titled 'INNOVINE - Combining innovation in vineyard management and genetic diversity for a sustainable European viticulture' (Grant agreement ID: 311775) under the Specific Programme 'Cooperation': Food, Agriculture and Biotechnology."

Characterization of Tunisian *Phyllosticta citricarpa* population and assessment of the effect of three *Trichoderma* species against the Citrus Black Spot. S. MANNAI¹, C. JEANDEL², J. AGUAYO², R. IOOS², N. BOUGHALLEB-M'HAMDI¹ ¹Institut Supérieur Agronomique de Chott Mariem, Laboratory of Plant Pathology, LR21AGR05 University of Sousse, 4042 Sousse, Tunisia. ²ANSES Plant Health Laboratory, mycology unit, USC INRAE 1480, Domaine de Pixérécourt, 54220 Malzéville, France. E-MAIL: n.boughalleb2017@gmail.com

Citrus black spot (CBS) caused by *Phyllosticta citricarpa* was first reported in Tunisia in 2019, making the first occurrence of this disease under a Mediterranean climate. *P. citricarpa* is a heterothallic fungus capable of sexual reproduction only when two mating types are present. Tunisian population revealed that strains belong to a unique mating type (MAT1.1.1). The aim of future investigations was to find some integrated tools for the

management of this fungus. In this sense, three *Trichoderma* species were evaluated against *P. citricarpa* development. The *in vitro* results obtained exhibited the effectiveness of these antagonists in reducing the mycelial growth of *P. citricarpa*, with varying levels of inhibition. *Trichoderma* species showed the highest mycelial growth inhibition with 54.33%, noted for *P. citricarpa* isolate CBS1 treated with *T. atroviride* (A3); 51.76 and 53.51% for *P. citricarpa* isolate CBS4 in presence of *T. atroviride* and *T. asperellum*, respectively. The *in vivo* assay on *Citrus limon* fruits confirmed the reduction in lesions development caused by the four *P. citricarpa* isolates. In fact, the reduction ranged from 28.28 to 72.83% for *P. citricarpa* CBS3 isolate treated with *T. kunmingense* and *T. atroviride*, respectively. Based on the current results, the *Trichoderma* species used reduced *P. citricarpa* mycelial growth *in vitro* and the lesions appearance due to *P. citricarpa* on *Citrus lemon* variety Eureka. Efficacy varies depending on the type of antagonist species and pathogen isolates. Therefore, these compounds have shown promising potential and could be used for CBS control.

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Identification of chickpea genotypes with multiple resistance to fusarium wilt and Ascochyta blight. F.Z. IBN EL MOKHTAR^{1,2}, I. MAAFA³, M. MITACHE^{2,4}, F. BELBOU^{1,5}, H. HOUMAIRI², O. IDRISI⁴, S. KRIMI BENCHEQROUN¹. ¹Plant Pathology Laboratory, Regional Center of Agricultural Research of Settat, National Institute of Agricultural Research (INRA), Morocco. ²Agri-Food and Health Laboratory, Hassan First University, Faculty of Science and Technology of Settat, Morocco. ³International Center for Agricultural Research in the Dry Areas (ICARDA), Agdal, Rabat 10080, Morocco. ⁴Laboratory of Food Legumes Breeding, Regional Center of Agronomic Research of Settat, National Institute of Agronomic Research, Avenue Ennasr, BP 415 Rabat Principale, 10090, Morocco. ⁵Biology department, Cadi Ayyad University, Faculty of Science Semlalia, Marrakech. Morocco. E-MAIL: sanae.krimibencheqroun@inra.ma

Chickpea (*Cicer arietinum* L.) is an important legume crop valued for its high protein content and soil-enriching nitrogen fixation. However, its production is threatened by biotic stresses, particularly Fusarium wilt (FW), caused by *Fusarium oxysporum* f. sp. *ciceris*

and Ascochyta blight (AB), caused by *Ascochyta rabiei* (Pass.) Labr. These diseases, especially under favorable environmental conditions, cause substantial yield losses and lack effective treatments, making genetic resistance the most viable solution. A total of 216 chickpea genotypes, including landraces and breeding lines, were evaluated for their resistance to FW and AB. FW screening was conducted in a sick plot field trial and a controlled growth chamber, using a completely randomized design with two replications. AB screening was conducted in a field trial (augmented RCBD) and a greenhouse trial (completely randomized design with two replications). Significant genotype, environment, and genotype-by-environment interaction effects ($P < 0.001$) were observed for both diseases. Using the Multi-Trait Genotype-Ideotype Distance Index (MGIDI) with a 30% selection intensity, 30 genotypes were identified as highly resistant to FOC, and 40 genotypes as moderately resistant to AB. Resistance levels varied between environments, highlighting environmental influence on disease expression. Notably, six genotypes exhibited resistance to both FOC and AB. The identified genotypes offer promising material for breeding programs aimed at developing dual-resistant chickpea cultivars. These efforts can enhance disease resilience and promote sustainable chickpea production.

POSTER PRESENTATIONS

Wheat Fusarium crown rot occurrence and disease strategy management in Algeria. H. BOUREGHDA, K. HACHEFA, K. DJEMAOU. *Ecole Nationale Supérieure Agronomique (ENSA), Département de Botanique, Laboratoire de Phytopathologie et Biologie Moléculaire, Hassan Badi street, El Harrach, Algiers, Algeria.* E-MAIL: hou.bouregghda@gmail.com

Crown rot (CR), caused mainly by *Fusarium* spp., is a globally widespread disease of wheat. CR may affect yield quantity and quality due to the accumulation of mycotoxin in wheat grains. Surveys conducted in Algeria have revealed the prevalence of CR in all cereal-growing regions in the north of the country, with *F. culmorum* being the dominant associated species. Within the framework of integrated disease management, the behavior of 17 durum and bread wheat varieties grown in Algeria were evaluated over two growing seasons (2022/23 and 2023/24) at the seedling (greenhouse) and maturity (field conditions) stages. The results showed that the bread wheat varieties were more resistant than durum wheat at both stages, with varying disease indi-

ces. At the seedling stage, disease indices ranged from 1.05 to 2.80. The bread wheat variety 'Rmada' was found to be the most resistant, while the durum wheat varieties 'Boussalem' and 'Manssourah' were considered the most susceptible. At maturity stage, disease indices ranged from 0.71 to 1.74, with Rmada recording the lowest index. In contrast, durum wheat varieties 'GTA' and 'Semito' were found to be the most susceptible. A strong positive correlation ($r=0.71$, $P \leq 0.05$) was observed between the behavior of the varieties at both stages over the two years. In conclusion, the variety 'Rmada', thanks to its behavior at both growth stages, represents a promising source for CR control and should be recommended in wheat breeding programs.

Diversity of fungal pathogens linked to dieback and canker diseases in Moroccan fruit trees.

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Fruit tree trunk diseases, caused by a wide range of fungal pathogens, affect the vascular tissues and bark of fruit trees, posing a major challenge to orchard management globally. This study aimed to investigate the diversity and distribution of trunk diseases affecting various fruit tree crops in the main production regions of Morocco. Symptomatic trunk samples were collected from infected trees, and the causal agents were identified through morphological and molecular phylogenetic analysis, targeting the internal transcribed spacer (ITS) region and the β -tubulin (β -tub) gene. The findings revealed a diverse range of fungal species associated with trunk diseases, including genera such as *Neoscytalidium*, *Lasiodiplodia*, *Botryosphaeria*, *Diplodia*, *Neofusicoccum*, *Diaporthe*, *Cytospora*, *Curvularia*, *Fusarium*, *Sordaria*, and *Biscogniauxia*. Some pathogens were found to be prevalent across different tree species and regions, indicating their widespread impact on orchard health. Pathogenicity tests confirmed that all isolates could induce characteristic lesions on inoculated shoots, with *Lasiodiplodia theobromae*, *Neoscytalidium hyalinum*, and *Diplodia insularis* causing the largest lesions. This study provides the first comprehensive report on species associated with apple, peach, cherry, plum, almond, and persimmon trees in Morocco, offering new species records and critical insights for developing effective control

strategies.

Olive diseases worldwide. V. SERGEEVA. *Western Sydney University Locked Bag 1797, Penrith NSW 2751, Australia.* E-MAIL: sergeeva@tpg.com.au

Plant diseases are of global concern and exact a heavy toll on food crop production. Olives are now one of the most extensively cultivated fruit crops in the world. Olive groves can be found in 60 countries around the world. However, growers face challenges due to many different pathogens that infect olive trees, reducing yields and increasing production costs. Harmful agents of the olive tree include fungi, bacteria, phytoplasm, viruses and virus-like agents attacking all parts of the plant. Different types of damage can be expressed qualitatively by describing the symptoms, or quantitatively through assessment of crown condition. The number of these occurrences has dramatically increased in recent years, causing serious damage to olive production. More than one factor can affect the health of a tree at any one time and differentiation can be made between the primary disease, which first and foremost affects the health of the tree, and the secondary disease, which has a less important influence and usually affects trees already weakened by an existing factor. An especially important concept is that olive-tree care attempts to manage plant health rather than just control disease problems. Due to the growth of the olive industry, it has become even more important to address the issues of product quality and the adoption of integrated pest and disease management. A proper understanding of olive variety, grove design, climate, soils and grove management are vitally important to the nurturing of trees, which affects health, quality and yield. A Guide to Olive Pests and Diseases published in 2024.

Sources of inoculum of *Alternaria* species associated with apple leaf and fruit spot disease. Z. EL KHATTABI, C. MORAGREGA, I. LLORENTE. *Plant Pathology, Institute of Food and Agricultural Technology, Universitat de Girona, C/ Maria Aurèlia Capmany, 61, 17003 Girona, Spain.* E-MAIL: zohra.elkhatabi@udg.edu

In recent years, a new apple disease caused by *Alternaria* has been reported in Europe, causing significant production losses. In Catalonia, this emerging disease was first detected in 2006 in commercial apple orchards in the province of Girona. Since then, it has progressively spread, leading to production losses ranging from 10% to 40%. The disease is usually associated with the *Alternaria alternata* and *Alternaria arborescens* species

complex. To develop an effective disease management program, it is essential to assess the inoculum potential. This requires identifying the specific sources of inoculum production within the apple orchards. This study aims to identify the sources of inoculum and model its production in apple orchards in Girona, with the objective of contributing to the development of effective management tools. The main sources of pathogen inoculum in apple orchards were identified through periodic sampling of leaf litter, canopy leaves, shoots, buds, and flowers. Additionally, the pathogenicity of the isolates obtained from these samples was assessed. The results indicated that *Alternaria* inoculum was produced in all examined organs; however, it was most abundant in leaf litter and other dead organic material on the soil.

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RNAi-mediated silencing of *ASI* and *B3* genes to reduce grapevine susceptibility to *Plasmopara viticola*. L. BOLOGNA, G. MADDALENA, E. SERGI, G. DE LORENZIS, S.L. TOFFOLATTI. *Department of DiSAA, Via G. Celoria 2, Università degli Studi di Milano, MI 20133, Italy.* E-MAIL: lucia.bologna@unimi.it

Downy mildew, caused by *Plasmopara viticola*, is one of the most widespread and economically impactful grapevine diseases in Europe and is primarily managed through fungicide applications. The increasing awareness of the negative impact of fungicides on human health and the environment is driving efforts to find more sustainable disease management strategies. RNA interference (RNAi) is emerging as a promising transgene-free biotechnological tool for pest management. In this study, RNAi was applied to protect grapevine from downy mildew by silencing two specific genes (*ASI* and *B3*), both implicated in nitrogen metabolism, which are associated with grapevine susceptibility to *P. viticola*. In the experimental setup, two susceptible cultivars, Pinot noir and Merlot, were sprayed with dsRNA specifically designed to silence the expression of the two genes. Plants treated with dsRNA targeting a gene not involved in nitrogen metabolism and with sterile water were used as controls. Leaves were sampled at 1, 3, 5, 7, and 14 days after treatment; three disks were obtained from each leaf and inoculated with a *P. viticola* sporangia suspension at a fixed concentration. A visual assessment of the inoculated disks was performed to evaluate disease severity by calculating the infection index per-

centage. Preliminary data indicate that the dsRNA-VvB3 treatment significantly reduced the infection index by day five post-treatment compared to both dsRNA-Vvi-ASI and control groups, although overall infection levels were modest, potentially due to non-ideal plant vigor. Future research will consider the use of in vitro-grown plants under controlled environmental conditions to further validate these promising findings.

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Impact of cropping practices on dry root rot and soil microbial diversity in durum wheat in semi-arid regions of Morocco. H. EL WAZZIKI¹, B. EL YOUSFI¹,

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Dry root rot is one of the most important wheat diseases worldwide, including Morocco. This study aimed to evaluate the health status of five no-till soils compared to a conventionally tilled soil in terms of dry root rot severity, microbial biodiversity, and the potential inoculum level of *Fusarium culmorum*. To assess disease severity, a root rot-susceptible variety 'Ourgh' was grown under greenhouse conditions in the six natural soils collected from three regions (Abda, Chaouia, and Ourdigha), each with different previous crops (lentils, fallow, wheat, barley, and forage). Disease severity was assessed at flowering on roots and stem internodes, and microbial biodiversity was estimated using microbial counting techniques. These assessments were also performed after artificial inoculation with *F. culmorum*. Root rot symptoms, confirmed by isolation of pathogen from infected roots and stems, were observed in all six soils at varying levels. Three pathogenic genera were identified: *Fusarium* spp., *Alternaria* spp., and *Bipolaris* (*B. sorokiniana*), with *Fusarium* spp. being present in all soils. Untilled soil showed higher microbial diversity than the conventionally tilled soil, which was dominated by bacteria, except in soil from Ourdigha with barley as the previous crop, where fungal and bacterial levels were similar. Inoculation increased microbial diversity and disease severity, and reduced plant height without significantly

affecting dry biomass. Except for this latter soil, microbial profiles shifted post-inoculation, with soils becoming either bacterial- dominant or balanced. Soil type, texture, and crop history (except barley) had no significant impact on microbial biodiversity. These findings highlight potential natural resistance mechanisms against *F. culmorum* for future biological control strategies.

This research was financially supported by the National Institute of Agricultural Research (INRA) of Morocco through the Aridoculture Centre (Settat) to conduct this study.

Assessing the Impact of *Fusarium culmorum* on agronomic performance and DON contamination in stems and grains of nine durum wheat varieties in Tunisia. S. GUERMECH^{1,2,3}, M. MASIELLO⁴, S. SOMMA⁴,

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Fusarium culmorum, the main causal agent of Fusarium Crown Rot (FCR) in Tunisia, represents a serious phytopathological threat under favorable environmental conditions. Its ability to produce the harmful deoxynivalenol (DON) mycotoxin, which can accumulate in plant tissues and grains, raises major concerns for human and animal health. Despite efforts over recent decades to breed resistant wheat varieties, many modern durum wheat varieties remain susceptible to FCR. This study aimed to evaluate the susceptibility of durum wheat varieties, widely grown in Tunisia to FCR. During two growing seasons (2020–2022), a field trial was conducted in Tunisia using artificial inoculation with *F. culmorum* at sowing. Agronomic traits such as tiller and plant density, kernels per spike, and grain yield were evaluated. FCR symptoms were present in all tested varieties, with significant differences in disease incidence, severity, and yield loss. The varieties Carioca, Maali, and Karim showed the highest incidence and severity, while Dhahbi, though susceptible, experienced the most severe yield loss (up to 42% in 2021). Strong correlations were observed between the incidence and severity of FCR and DON concentration in stems, and between the frequency of *F. culmorum* and DON concentration in grains. In addition, both latter parameters and FCR incidence were

correlated with yield loss. On the other hand, a weak correlation was observed between DON concentrations in stems and grains, indicating limited translocation of the toxin. These findings highlight the impact of *F. culmorum* on wheat health, yield, and mycotoxin contamination, emphasizing the importance of resistant selection and integrated disease management.

Study of the behavior of different potato varieties in response to infection with major viruses in most regions of Algeria. L. ALLALA-MESSAOUDI¹, I. MOURAKEB², M. BENDANBRI², M.S. SHAHID³.

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In Algeria, viruses are among the most important biotic factors that significantly influence the growth and productivity of potato crops, both in terms of quality and quantity. This study is part of an investigation into the behavior of potato cultivars in response to viral infections, conducted under controlled conditions and in open fields. The objectives are twofold: first, to update data on the prevalence of the major potato-associated viruses (PVY, PVX, PVA, PVS, and PLRV); and second, to provide insights into the degree of resistance or susceptibility expressed by three registered potato cultivars (Kennebec, Désirée, and Red Pontiac) in the region. The results showed that most of the studied viruses were present, with the highest infection rates for PVX and PVY viruses in open fields, where infection rates reached 95% and 82%, respectively. However, under controlled conditions using an insect-proof film, the infection rates of these two viruses were significantly reduced, with PVX at 37% and PVY at 49%. Furthermore, the findings indicate that Kennebec is more resistant to PVY, despite a relatively high infection rate of 68%, while Red Pontiac proved to be the most susceptible of the other three varieties tested.

Pathogenic, morphologic, and genetic studies of *Colletotrichum* spp. associated with anthracnose of citrus in Algeria. S. ALI-AROUS¹, M. MEZIANE¹, K. DJELOUAH², S.M. SANZANI³. ¹Laboratory of production

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The economic importance of citrus fungal diseases is constantly increasing, and they are caused by several species, some of which may be linked to the widespread citrus decline observed in Algerian citrus areas. Citrus anthracnose is associated with mild to severe symptoms on leaves, fruits, and twigs, likely due to host susceptibility, climatic conditions, and possibly affected species. For many years, this species was known as *Colletotrichum gloeosporioides*. The latter was genetically characterized by using the universal ITS regions as a marker, and it was the only species reported to be associated with citrus anthracnose in Algeria. By isolating 58 fungal colonies from infected leaves, fruits, and twigs collected from citrus orchards, four different isolates of *Colletotrichum* (CHDZ-1, CHDZ-2, CHDZ-3 and CHDZ-4) were distinguished from different aboveground parts of infected citrus fruits of different species and cultivars grown in the Chlef Valley. Based on Koch's postulate, all of them were involved in the development of the disease. Multi-locus sequencing analyses of the internal transcribed spacer ITS and glyceraldehyde-3-phosphate dehydrogenase (GAPDH) revealed that species belonging to the *C. gloeosporioides* species complex were associated with the disease. Further molecular sequencing of other genomic regions is recommended for the precise identification of these *Colletotrichum* isolates. During this research, we observed severe anthracnose infection in the most visited citrus region, causing visible damage to all aerial parts of trees. Special attention must be paid to this emerging disease to prevent its fatal establishment.

The foliar black spot caused by *Neodidymelliopsis ranunculi* (Ascomycota: Didymellaceae) a new fungal disease in the Algerian citrus orchards. S. ALI-AROUS¹, K. DJELOUAH², A. HOUARI¹, O.J. ALABI³, M. SÉTAMOU⁴, S.M. SANZANI⁵. ¹Laboratory of Production and Protection of Crops of Chlef region, University of Hassiba Ben Bouali, Chlef, Algeria. ²CIHEAM Bari, Via Ceglie 9, 70010 Valenzano, Bari, Italy. ³Department of Plant Pathology & Microbiology, Texas A&M AgriLife Research & Extension Center, Weslaco, TX 78596, USA. ⁴Texas A&M University-Kingsville Citrus Center,

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During routine surveys in the summer of 2017, severe leaf defoliation was observed in four citrus orchards, two sweet orange ('Thompson Navel' and 'Washington Navel') and two mandarin ('Nova' and 'Oro Grande'), located in the Chlef valley in the northwestern Algeria, one of the most important citrus region of the country. Visual inspection of defoliated green leaves and leaves on tree canopy revealed unusual symptoms consisting of greasy black protuberant spots on the abaxial side and yellowing on the adaxial surface. A quick survey was conducted to determine the disease incidence. The percentage of trees infected with the disease reached 50% and 35% in sweet orange and mandarin orchards, respectively. The incidence of the diseased leaves ranged from 30% to 60% for sweet orange, while 50–70% of leaves showed disease symptoms in mandarin orchards. Morphological identification such as size, shape, status of otiole(s) and aggregation of pycnidia then a multi-locus sequencing approach was used for identity confirmation based on the 5.8S region of rDNA with flanking internal transcribed spacers (ITS), a portion of the 28S large subunit of rDNA (LSU), part of RNA polymerase second largest subunit (RPB2) and of β -tubulin gene (TUB2). The outcome of the identification process has associated this disease to the fungus *Neodidymelliopsis ranunculi*. Koch's postulates also confirmed *N. ranunculi* to be the causal agent. This is the first report of black spot symptoms caused by *N. ranunculi* on citrus in Algeria.

Recent findings of new citrus tristeza virus strains in the Chlef valley, Algeria. M. MEZIANE, S.A. AROUS, N.A. BENTURQUIA. *Research Laboratory of Crop Production and Protection, Hassiba Benbouali University of Chlef, Algeria.* E-MAIL: m.meziane@univ-chlef.dz

Citrus production plays a vital economic role in the Chlef Valley. However, this important crop faces ongoing challenges from diseases that threaten yield, fruit quality, and the long-term sustainability of orchards. For decades, several citrus orchards have shown decline and severe symptoms associated with tristeza disease, caused by the new, virulent exotic VT strain first reported in 2019, affecting both old and young orchards. To update the current distribution and strain occurrence of this disease, an additional survey was conducted in the valley

in 2021. Samples collected from various citrus areas were tested using DTBIA/DAS-ELISA. The survey revealed an increased infection rate of 4.24%, up from 3.21% in 2019. Molecular genotype characterization using multiple molecular markers and CP gene sequencing confirmed the presence of the preexisting T30 and VT genotypes. In addition, two new genotypes were detected in the S1 and T3 strains. This study confirmed the widespread dissemination of the virulent VT genotype and the emergence of new alien strains. The other CTV isolates were found to be Mediterranean, with a nucleotide identity of 98.6% to 99.1% compared to the worldwide T30 mild CTV isolates recorded in the Mitidja region. These findings raise serious concerns: the discovery of new strains alongside the continued spread of the severe VT genotype, combined with the presence of efficient aphid vectors, poses a significant threat to the citrus industry in Algeria. This situation requires proactive measures by the National Phytosanitary Services, including extending surveillance to other citrus production regions.

First report of *Septoria pistaciarum* on pistachios in Basilicata, Italy. M. GALLO¹, W. MELLIKECHE¹, R. DHAHRI¹, V.M. RICCIUTI², A.M. D'ONGHIA¹, F. VALENTINI¹. ¹Centre International de Hautes Etudes Agronomiques Méditerranéennes Bari (CIHEAM Bari), Via Ceglie 9, 70010 Valenzano, Bari, Italy. ²Azienda agricola - Pistacchio di Stigliano, C.da Sauro – Capalbi, 75018 Stigliano, Matera, Italy. E-MAIL: gallo@iamb.it

In May–June 2024, leaf spots were observed on 'Bianca' pistachio trees from a Stigliano (MT) farm, Basilicata, Italy. Leaves showing distinct red-brown spots with black margins were brought to the laboratory for isolation. Small portions (0.2–0.3 cm²) of symptomatic leaves were surface-sterilised, air-dried in a laminar flow hood, and placed on antibiotic-amended Potato Dextrose Agar (PDA), then incubated at 25°C for 7–10 days. Slow-growing, *Septoria*-like colonies were transferred to PDA plates, and pure cultures were obtained from single hyphae. The isolate produced sulcate and immersed grey/black colonies. A yellow mucilaginous matrix exuding from black conidiomata, often covered by white mycelium, was observed. Conidia were hyaline, curved to falcate, with 1–5 septa, measuring $(21.8\text{--}) 45.2 \pm 8.5$ (–64.4) \times $(1.7\text{--}) 2.9 \pm 0.5$ (–4.2) μm . A comparison with data from the literature confirmed the identification of *Septoria pistaciarum* Caracc. Traditional identification was further confirmed by sequencing the ITS region of rDNA. A BLAST search using the ITS sequence of the *S. pistaciarum* isolate revealed an exact match

with several reference sequences in the database, mainly from pistachio hosts. Pathogenicity tests were conducted on two-year-old pistachio plants, and the fungus was successfully re-isolated from leaf lesions, fulfilling Koch's postulates. While *S. pistaciarum* had previously been isolated in Sicily, this is the first confirmed occurrence on the continental mainland of Italy.

Preventing food-borne threats at the source: A one health approach to crop and animal Biosecurity. F. ALI¹, M. SADIQ², I. AHMAD². ¹*Pet Street Veterinary Clinic Abu Dhabi UAE.* ²*University of Agriculture Peshawar Pakistan.* E-MAIL: drfarzand_2011@yahoo.com

Ensuring food safety begins long before animal production starts in the field, with healthy plants. As a veterinarian, the link between plant health and animal health is clear: contaminated crops can lead to unsafe feed, compromised animal health, and ultimately impact human food safety. Recent advances in phytopathology offer new tools to detect and control plant diseases that threaten the safety and quality of crops used in animal feed and the broader food chain. By integrating veterinary and plant sciences, we can develop more effective strategies to prevent foodborne illnesses, reduce toxin exposure (such as mycotoxins), and enhance biosecurity. Strengthening education and collaboration across disciplines is key to training professionals who can address these challenges holistically. This approach supports a safer, more sustainable food system, one where plant, animal, and human health are protected together.

Incidence and severity of early blight affecting *Solanaceae* family crops. R. DUMBADZE, G. MEPARISHVILI, L. GORGILADZE, N. JABNIDZE, L. KOIAVA, M. MURADASHVILI. *Department of Plant Diseases Monitoring, Diagnostics and Molecular Biology, Institute of Phytopathology and Biodiversity, Batumi Shota Rustaveli State University, Kobuleti 6200, Georgia.* E-MAIL: rusudandumbadze6@gmail.com

In the conditions of today's globalization, there is a great danger that the spread of some highly harmful microorganisms will take a very wide scale and adapt to different ecological conditions in many regions. Some microorganisms were not previously considered to be among the causes of diseases threatening agricultural crops, but today they are already considered important phytopathogens that significantly reduce the quantity and quality of crops. Among them, *Alternaria* fungi are note-

worthy, which cause especially great damage to the crops of the family of *Solanaceae*. They cause spots of Potatoes, Tomatoes, Eggplants, Peppers, Tobacco - leaves, stems, tubers and fruits. In 2024, the monitoring of the crops of the family of *Solanaceae* was carried out in the municipalities of the Adjara region, to detect *Alternaria* spp. Private homesteads and farms were visited during the vegetation period of the plant through a scientific expedition. About 186 disease samples were collected and analysed. The Incidence and Severity of *Alternaria* spp. was uneven on *Solanaceae* crops, the Incidence on potatoes was 35%, and the severity was 25%, on tomatoes 30% and 20%, the disease was spread with the same frequency on eggplant and pepper, and it was 20% and 20%. It should be noted that the mentioned disease was not observed at all on tobacco. The identification of *Alternaria* spp. isolates was performed exclusively based on morphological criteria, with the most significant characteristics being the morphology of the conidia and the formation.

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CONCURRENT SESSION C3 – Huanglongbing of citrus and bacterial plant diseases

SESSION KEYNOTES

Sustainable control of bacterial diseases: Current advances and safety concerns. E. STEFANI. *Department of Life Sciences, University of Modena and Reggio Emilia, via Amendola 2, Pad. Besta, 42122 Reggio Emilia, Italy.* E-MAIL: emilio.stefani@unimore.it

Bacterial diseases represent a challenge in the current scenario of climate change, innovative agricultural systems, increased consciousness of sustainable production, need to minimise chemical inputs into the environment. Basically, the management of bacterial diseases rely on a plethora of copper compounds, a few antibiotics (where allowed), and, in case of insect transmission, use of insecticides. Emerging technologies, such as CRISPR and TALEN gene editing or the use of RNAi vectors, offer the potential to develop disease resistant plants or to modify the plant response to a pathogen attack; however, both technologies raise ethical concerns for their possible off-target effects, potential unintended consequences and unforeseen and unpredictable impacts on human health and the environment. The use

of copper-based pesticides has been thoroughly reduced, and the European Commission adopted the regulation (EU) 2018/1981 to restrict copper use in agricultural systems (i.e. only uses resulting in a total application of maximum 28 kg of copper per hectare over a period of 7 years are authorised), and copper compounds are candidates for substitution. Then, the biocontrol of bacterial diseases appears the most prospective approach to ensure crop productivity, remuneration to farmers, environmental safety and consumers' satisfaction. Many beneficial microbes are described in the scientific literature as possible solutions for farmers: basically, these are bacteria and fungi; additionally, bacteriophages are described as prospective biocontrol agents that can be used to control phytopathogenic bacteria. The biosafety of plant pathogen biocontrol agents is a crucial consideration for those researchers and companies intending to develop commercial biopesticides as these agents, while generally considered safe, once introduced into the environment to control diseases may pose potential risks, such as unintended effects on non-target organisms, host-switching, and the potential for the biocontrol agent to become a new pathogen. Therefore, no authorisation will occur in the EU if the putative microbial biocontrol agent is suspected to be pathogenic to humans or terrestrial vertebrates, if it has effects on other non-target organisms and if it has toxicological effects on humans, animal health and the environment. Currently no guidance is available on the risk assessment for microbial consortia specifically developed for plant protection purpose: indeed, despite microbiome manipulation in agricultural systems may provide a system approach to increase sustainability and safety, studies on microbial dynamics in the topsoil are scarce. Bacteriophages as anti-bacterial agents are generally considered non-toxic, safe for users and target-specific, leading to them being possibly used as biopesticides in the coming future: a document issued by OECD is currently available to provide guidance for a regulatory framework. Finally, nanoparticles are being explored as antimicrobial agents, nanocarriers and microbe-mediated particles, despite there are insufficient reliable ecotoxicological data for risk assessment purposes and to establish safety doses. To conclude, microbial antagonists, bacteriophages, nanoparticles and RNAi vectors as putative and prospective plant protection agents are under evaluation for their biosafety: indeed, their foreseen increasing use requires additional studies to confirm safety for consumers and lack of possible adverse effects to non-target organisms and the environment, prior to their registration as plant protection products.

ORAL PRESENTATIONS

Exploring the potential of antimicrobial peptides, bacteriophages, and bacteriocins in managing bacterial diseases, with a focus on *Xylella fastidiosa*. M. SABRI¹, O. CARA^{1,2}, F. VALENTINI¹, A. DE STRADIS³, C.D. CALVANO⁴, A. M. BIANCO⁴, K. EL HANDI¹, T. ELBEAINO^{1,5}. ¹International Centre for Advanced Mediterranean Agronomic Studies (CIHEAM BARI), Bari, Italy. ²University of Bari, Department of Soil, Plant and Food Science, Bari, Italy. ³Institute for Sustainable Plant Protection (IPSP), National Research Council of Italy (CNR), Bari, Italy. ⁴University of Bari, Department of Chemistry, Bari, Italy. ⁵Institute for Sustainable Plant Protection (IPSP), National Research Council of Italy (CNR), Portici, Italy. E-MAIL: elbeaino@iamb.it

The increasing threat of *Xylella fastidiosa* (Xf), alongside other significant bacterial plant pathogens affecting Mediterranean agriculture, such as *Erwinia amylovora* (Ea), *Agrobacterium tumefaciens* (At), and *Xanthomonas campestris* pv. *campestris* (Xcc), which cause leaf scorch, fire blight, and black rot diseases, respectively, highlights the need for sustainable, effective, and eco-friendly control strategies. Our research investigates the use of antimicrobial peptides (AMPs), bacteriophages (phages), and lactic acid bacteria-derived bacteriocins as advanced nanobiocontrol approaches to combat these pathogens. At the AMPs level, potent antimicrobial agents against Xf subspecies *pauca* (Xfp), including Nisin A from *Lactococcus lactis*, and Ascaphin-8, DASamp2 from frog skin, and MK-45, MR-53, MW-56, MG-58 from *Leuconostoc mesenteroides* strain MS4 have shown strong bactericidal activity against Xfp with different lethal concentrations. Notably, Nisin A rapidly disrupted bacterial membranes, effectively controlling infections in *Nicotiana benthamiana* with systemic translocation lasting up to nine days. Similarly, the four *Leuconostoc mesenteroides* MS4-derived bacteriocins were efficient at 0.2–0.4 mg/mL to combat Xfp infections *in planta*, proving to be cost-effective, eco-friendly biocontrol agents. Furthermore, a newly identified lytic bacteriophage (MATE 2) exhibited broad-spectrum activity against Xfp and Xcc, a major cruciferous crop pathogen. When combined with *L. lactis* and Nisin A, MATE 2 showed a synergistic effect, reducing Xcc infection symptoms in broccoli by 71%, outperforming individual treatments (64% and 38%). Additionally, a novel bacteriophage targeting Ea effectively reduced fire blight disease in pear plants, highlighting phage-based strategies' broader poten-

tial. These outcomes demonstrate that nano-biocontrol agents; bacteriocins, bacteriophages, and AMPs, can serve as sustainable, eco-friendly alternatives to chemical pesticides, offering a multifaceted approach to managing *Xfp* and other bacterial plant diseases while promoting agricultural sustainability.

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***Ralstonia pseudosolanacearum*, an emerging threat in Europe.** R.A.M. VREEBURG, M.J.C. PEL, T.M. RAAY-MAKERS, M. BERGSM-VLAMI. *Netherlands Institute for Vectors, Invasive plants and Plant Health (NIVIP), National Plant Protection Organization (NPPO), Netherlands Food and Consumer Product Safety Authority (NVWA), Geertjesweg 15, 6706 EA Wageningen, The Netherlands.* E-MAIL: r.a.m.vreeburg@nvwa.nl

Until recently, *Ralstonia pseudosolanacearum* was absent from outdoor agricultural systems and natural aquatic environments in Europe and was only reported in restricted greenhouse cultivation systems, such as in the greenhouse cultivation of roses in the Netherlands and some other European countries. Since a few years now, *R. pseudosolanacearum* has been frequently found in Europe: in imported plant material (e.g. ginger and curcuma), plants cultivated outdoors or under semi-protected conditions (e.g. tunnels) - often after importation of infected plant material, and in waterways. Given the broad range of host plants that can be infected by *R. pseudosolanacearum* major awareness is currently needed in Europe. Potato is a major arable crop in Europe and a known host of *R. solanacearum*, we therefore studied the virulence of three *R. pseudosolanacearum* strains in potato. When potato plants are inoculated with *R. pseudosolanacearum*, they develop symptoms at high temperatures (28°C). At lower temperatures (20°C) however, less or no symptoms develop while the bacteria could be found back in the progeny tubers, increasing the risk for spread of the bacteria. Genomic analysis of recent European isolates shows that there is a high genetic diversity among the *R. pseudosolanacearum* strains that have been found in Europe. This makes it difficult to predict how well the virulence results obtained with the three tested *R. pseudosolanacearum* strains can be generalized to all *R. pseudosolanacearum* strains.

Population Genetic signatures revealed contrasted outbreak histories of bacterial diseases: the case of Bacterial wilt, Huanglongbing and Citrus Canker. G. CELLIER. *Anses-Plant Health Laboratory, Saint Pierre F97410 – Reunion Island.* E-MAIL: gilles.cellier@anses.fr

As long-distance traveling of human beings and movement of goods drastically increase over time in relation to globalization of trade and exchanges, and so is the spread of bacterial pathogens and associated infectious diseases across the globe. Key for improved disease control lies into acquiring a thorough knowledge on factors shaping pathogen populations at fine scales and how they interact with their environment. In order to show much more clearly how infectious agents are spreading and evolving other than genetic drift alone, phylogenetic and epidemiological techniques are often used. Bacterial lineage-centered molecular genotyping techniques, such as multilocus variable number of tandem repeats analysis (MLVA). They are of interest especially when they provide high throughput, a sound phylogenetic signal, and a resolution fitting the spatiotemporal scale investigated. In the case of complex plant pathogens, such as the *Ralstonia solanacearum* species complex (RSSC), the 'Candidatus liberibacter spp.' species complex associated with the huanglongbing disease, and *Xanthomonas citri* pv. *citri*, several studies achieved molecular characterization of outbreak strains during the last decade, and brought light on its epidemiology. Nevertheless, selecting proper genetic marker and analytical algorithm is vital to apply molecular genetics in a given biological population. We will present examples of epidemiological results produced within the framework of these three pathosystems, which revealed the need for a proper sampling and methodological approach, along with the adoption of complementary multiscale analyses from gene to genome in order to apprehend the evolution complexity as a whole.

Citrus huanglongbing in California: current status and progress. D. GUTIERREZ¹, L. KUMAGAI¹, K. OKASAKI¹, D. MORGAN¹, G. VIDALAKIS². ¹*California Department of Food and Agriculture, Sacramento, CA 95814, United States of America.* ²*Department of Microbiology & Plant Pathology, Citrus Clonal Protection Program, University of California, Riverside, CA 92521, United States of America.* E-MAIL: vidalg@ucr.edu

The spread of *Diaphorina citri*—the Asian citrus psyllid (ACP) and vector of Huanglongbing (HLB)—has posed a persistent threat to California's citrus industry. First identified in San Diego County in 2008, ACP

has since established itself throughout all major citrus-producing areas. The first detection of HLB occurred in 2012 in Los Angeles County. By 2024, state agencies tested more than 650,000 citrus trees and 330,000 psyllid samples, confirming and removing over 9,800 HLB-positive trees from urban landscapes across six southern California counties. In response, quarantine zones now span over 6,215 square kilometres, limiting the movement of nursery stock and commercial fruit to mitigate further spread. The California Department of Food and Agriculture (CDFA), in collaboration with the USDA and the Citrus Pest and Disease Prevention Committee, manages a \$47 million program encompassing detection, regulatory, and biological control strategies. Targeted surveys, incorporating travel and human activity data, inform inspection priorities. Since 2011, more than 31 million *Tamarixia radiata* (*Eulophidae*) parasitoids have been released to suppress ACP populations, particularly around new HLB finds and high-risk corridors. Regulatory zones were restructured in 2017 to restrict the movement of citrus from southern to central California, while nursery stock production is confined to protective structures with mandatory testing. Ongoing efforts include public outreach campaigns, homeowner engagement, and multi-agency collaborations to eliminate unmanaged citrus. Future program goals include scaling HLB diagnostic capacity through private and industry-led laboratories, while state and federal funding continues to drive research in detection, epidemiology, and sustainable management of the HLB-ACP complex.

Whole Genome Sequencing and Fourier transform Infra-Red typing bring new insights to Portuguese *Erwinia amylovora* population diversity.

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Following the first outbreaks of fire blight affecting Portuguese pear orchards in 2010, intensive surveys and control of nursery materials resulted in the collection of a large set of strains of *Erwinia amylovora*. Isolates obtained from distinct apple, pear and quince varieties, from different production areas were identified fol-

lowing EPPO standard PM7/20(3). Characterization of *E. amylovora* pathogenicity and virulence features, assessed through biological tests by inoculation of apical pear twigs and immature fruits, displayed distinct virulence levels, even within the same CRISPR group. A set of 44 selected isolates was sequenced by Illumina Whole Genome Sequencing (WGS) and exhibited the expected conservative patterns with high levels of similarity for the whole set (>99.87%). In an attempt to understand these differences in virulence patterns, the bacterial collection was further characterized by Fourier transform infrared spectroscopy (FTIR). FTIR showed reproducibility and congruence of cluster composition for the biomolecules within the wavelength range of 800-1300 cm⁻¹, including lipids, proteins and polysaccharides. As expected, *E. tasmaniensis* and *E. pyrifoliae* grouped as outliers after clustering, by Euclidean UPGMA. Interestingly, the Portuguese strains of *E. amylovora* revealed specific features distinct from the reference strain CFBP 1430. Integrative analysis of WGS data and FTIR high-resolution fingerprinting profiles unveiled the diversity associated with the expression of disease severity, often hidden by the homogeneity of WGS profiles. These diagnostic and epidemiological features derived from differential expression of specific biomolecules, may contribute to better understanding new outbreaks and alternative therapies.

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***Xanthomonas euroxanthea* expands its host range: first occurrence in sunflower crops in Bulgaria.**

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Sunflower (*Helianthus annuus*) is a major oilseed crop globally, with Bulgaria being the leading producer within the European Union. During a 2021 field survey in Kavarna (Dobrich district, Bulgaria), sunflower plants exhibiting necrosis and longitudinal cracking of the petiole were observed. Microscopic examination suggested a bacterial infection, prompting further etiological investigation. Strains belonging to the genus *Xanthomonas* were isolated, and phylogenetic and phylogenomic analyses revealed their clustering with *Xanthomonas euroxanthea* CPBF 424^T, a pathogenic strain identified in walnut buds in Portugal, responsible for walnut bacterial blight. The sunflower isolate exhibited five of eight *X. euroxanthea*-specific markers (XEA4-XEA8), a pattern also observed in strains from *Solanum lycopersicum*, *Phaseolus vulgaris*, and rainwater sources, suggesting host adaptation and a potential emerging lineage. This is the first report of *X. euroxanthea* affecting sunflower. Given sunflower's economic importance and widespread cultivation in Mediterranean countries, this study highlights the need for monitoring the pathogen's distribution and potential impact on sunflower production. As many sunflower crops are also used in farming systems for crop rotation systems, alternating with rice, beans, or maize, early detection and management strategies are crucial to mitigate risks associated with its spread to other plant hosts.

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POSTER PRESENTATIONS

Survey of bacteria affecting stone fruits and almond in Montenegro. T. POPOVIĆ¹, J. ADAMOVIĆ², A. PROKIĆ², N. ZLATKOVIĆ³, A. OBRADOVIĆ². ¹Administration of Food Safety, Veterinary and Phytosanitary Affairs, Serdara Jola Piletića 26, Podgorica, Montenegro. ²Faculty of Agriculture, University of Belgrade, Nemanjin 6, Belgrade, Serbia. ³Institute for Plant Protection and Environment, Teodora Dradžera 9, Belgrade, Serbia. E-MAIL: tamara.popovic@ubh.gov.me

The production of stone fruits and almonds in Montenegro is often compromised by various pathogenic bacteria. The infection and disease spread are facilitated by favourable environmental conditions as well as extensive cultivation practices and inadequate control measures. Symptomatic samples of stone fruit and almond plant material were collected from different locations across Montenegro during the 2017–2020 survey. Not all of them resulted in bacterial isolation indicating that some symptoms could not be a reliable diagnostic criterion. From apricot, peach and sweet cherry samples 48 strains were isolated, and based on their morphological, pathogenic, biochemical, and molecular characteristics identified as *Xanthomonas arboricola* pv. *pruni* (Xap). Additionally, 29 strains, identified as *Pseudomonas syringae* pv. *syringae* (Pss), were isolated from apricot, peach, nectarine, sweet cherry, Japanese plum, and almond, suggesting a wider spread of this pathovar across the country. No mixed infections were registered. Phenotypic and genetic variations within the Pss population could be a consequence of multiple strain origin, and adaptation to different hosts and climatic conditions. In contrast, the genetic homogeneity observed in the Xap population suggests a spread of the local population without new introductions. To prevent spread of Xap to other areas and susceptible hosts, strict phytosanitary measures are implemented. However, Pss as a wide host range pathogen should be controlled by cultural practices. All tested strains showed *in vitro* resistance to copper sulphate and sensitivity to streptomycin sulphate, indicating that integrated management strategies have to be implemented to protect orchard productivity and stone fruit yield.

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First report of *Pectobacterium carotovorum* causing potato soft rot in Georgia. M. MURADASHVILI¹, C. LOMBAERS², M. KRIJGER², P. VAN DER ZOUWEN², G. MEPARISHVILI¹, Z. SIKHARULIDZE¹, R. DUMBADZE¹, J.M. VAN DER WOLF². ¹Batumi Shota Rustaveli State University, Institute of Phytopathology and Biodiversity, Kobuleti, 6200, Georgia. ²Wageningen University & Research, Droevendaalsesteeg 1, 6708 PB Wageningen, the Netherlands. E-MAIL: makamuradashvili25@yahoo.com

Today, bacterial diseases still remain a significant limiting factor for potato harvest in Georgia. During the 2022 year has been revealed that potato soft rot diseases caused by the *Pectobacterium carotovorum* in the various municipalities of Georgia: Marneuli, Kobuleti Tsalka, and Akhaltsikhe. Under the research were observed many cases of rotted potato tubers with cream and dark black colored tissue, as in the field, also storage. Since 2020, potato soft rot and blackleg diseases, caused by *Dickeya solani* were particularly prevalent in the regions of Khulo, Akhaltsikhe, Akhalkalaki, and Kobuleti (Muradashvili *et al.*, 2023). 19 isolates from collected 28 samples were identified as a gram-negative, facultative anaerobes. They have produced cavities on the crystal violet pectate media (Perombelon & Van Der Wolf, 2002) were negative for oxidase, urease, and indole production, did not produce acid from dulcitol or sorbitol, could not utilize malonate and citrate. The isolates were catalase positive, and were able to grow at 37°C. Pathogenicity tests were conducted by the fulfill Koch's postulates. Observed potato soft rot similar tissue maceration on the inoculated potato slices and whole potato tubers and causal pathogen were reisolated. 11 isolates from them were selected for genetic study. DNA was extracted from test bacterial isolates and analyzed using gapA PCR with the primer pair gapA-7-F/gapA-938-R (Cigna *et al.*, 2017). The amplicon size was 932 bp. GapA gene sequence alignment fragments showed a uniform group of isolates 100% identical to gapA sequences of *Pectobacterium carotovorum* strains MBr2, isolated from *Brassica rapa* subsp. *chinensis* in Taiwan (OR651974), and strain ZB18121, isolated from China (OP793220). The gapA sequences of the strains from Georgia are represented by accession numbers: PP060483; PP060484; PP060485; PP060486; PP060487; PP060488; PP060489; PP060490; PP060491; PP060492; PP060493. This is the first report of *Pectobacterium carotovorum* causing potato soft rot in Georgia.

This work was provided by Shota Rustaveli National Science Foundation (SRNSF) of Georgia [grant № FR- 21-1778].

First report of *Pseudomonas avellanae* causing hazelnut decline in Serbia. T. POPOVIĆ MILOVANOVIĆ¹, S. LORETI², P. MILOVANOVIĆ³, A. JELUŠIĆ⁴, R. ILIČIĆ⁵, M. SCORTICHINI⁶. ¹Institute for Plant Protection and Environment, Teodora Drajzera 9, 11040 Belgrade, Serbia. ²CREA - Research Centre for Plant Protection and Certification, Via Carlo Giuseppe Bertero 22, 00156 Rome, Italy. ³Agrounik doo, Krnješevačka BB, 22310 Šimanovci, Serbia. ⁴University of Belgrade, Insti-

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During July 2024, a decline was observed on hazelnut (*Corylus avellana* L.) cultivar Tonda di Giffoni, 5-years old. Developed symptoms coming out when trees started to yield in form of rapid wilting of leaves on branches. Diseased stems exposed a brown discoloration of vascular tissue. Isolations from wood tissue resulted in the forming of large, convex, levan-type, mucoid, cream-whitish *Pseudomonas*-like bacterial colonies on Nutrient agar supplemented with 5% sucrose. Results of LOPAT tests (Levan positive, Oxidase negative, Potato soft rot negative, Arginine dehydrolase negative, Tobacco hypersensitivity positive) ranged isolates into *Pseudomonas syringae* group Ia. Pathogenicity test was performed on shoots of potted 1-year-old hazelnut (Tonda di Giffoni) plants inoculated by bacterial suspension through leaf scars in the early autumn. First necrosis at the points of inoculation was observed after ten days, and continued to spread longitudinally through the stem, reaching a length of 15–20 cm after two months. Koch's postulates were fulfilled by reisolation of the same pathogen from the inoculated stems. Genetic identification was performed by sequencing of *gyrB* (DNA gyrase subunit B) gene. Nucleotide BLAST analysis of the partial *gyrB* gene sequence of the two Serbian isolates showed 100% identity with *Pseudomonas avellanae* strains from the NCBI database. Phylogenetic analysis indicated genetic homogeneity among the tested and five reference *P. avellanae* strains (BPIC631, CFBP4960, CIP 105176T, NCPPB 3491, and NCPPB 4222) and confirmed identification by placing them within the same tree cluster. This finding indicates on possibility of wider spread of this bacterium in the territory of EU.

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CONCURRENT SESSION D1 – Nematodes of crops and forestry

SESSION KEYNOTES

Microbiome-assisted agriculture for the management of nematode pests in vegetable crops. S. MOLINARI.

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Plant Microbiome is the collective communities of plant-associated microorganisms, also referred as plant second genome. Such microorganisms colonize the rhizosphere that is the zone of soil influenced by plant roots. Plant microbiome in rhizosphere can improve root architecture, enhance nutrient uptake and tolerance to environmental stresses, and stimulate plant immune system. Benefits from enrichment of soil by rhizosphere microorganisms in agricultural practices have brought to the development of a more sustainable Microbiome-Assisted Agriculture (MAA) to establish low input and low impact crop systems. Massive use of chemical fertilizers and pesticides has caused farmers to face severe problems of soil degradation associated with decline in quality and yield in crop production. Soil is a non-renewable resource on human time scales with high vulnerability to degradation processes such as erosion, depletion of the soil organic carbon (SOC) pool and loss in biodiversity, in soil fertility caused by nutrient scarcity and uptake difficulty, acidification and salinization. Natural undamaged soils are rich in beneficial rhizosphere microbes, such as arbuscular mycorrhizal fungi (AMF), opportunistic fungi, plant growth promoting rhizobacteria (PGPR), that are specifically selected by plants to have benefits in terms of growth and tolerance to abiotic and biotic stresses. One of the main tools for regeneration of degraded soils is the addition of suitable microbiome-generating formulations in the attempt to establish beneficial relationships in the rhizosphere between microorganisms and roots for having higher yield and product quality. Different commercial microbiome-generating formulations or low scale green waste composts have been used in our laboratory to test their activity as biofertilizers and biocontrol agents (BCAs) against attacks of soil-borne parasites, such as plant parasitic nematodes, to vegetable plants. Pilot programs involve vegetable crop plants grown in pots or small plastic containers located in glasshouses under controlled environmental conditions, and single pre-treatments before artificial nematode inoculation. Commercial formulations are differently effective as BCAs, whilst a specific low scale food waste compost has been found to be both a good fertilizer and bio-stimulant against root-knot nematodes (RKNs). Best formulations can reduce infection factors to the 40-50% of controls. Paramount to have an effective increase of pest resistance under our experimental conditions is the amounts of added formulations with respect to plant species, age and size. Inhibi-

tory compounds are used to select specific microorganisms and study their specific role. Controls of nematode infection involves mycorrhization of roots with the activation of plant immune system, monitored through detection of defense gene expressions and enzyme activities. Target genes are those encoding for pathogenesis-related proteins (PR-proteins) and enzymes regulating the Reactive Oxygen Species (ROS) production and degradation. Primed plants react to pest attack by high and prompt defenses, such as Hypersensitive Reaction characterized by cell death. Microbiome-generating formulations and composts are currently being studied as agro-products that can associate bio-fertilizing and bio-stimulating effects. Moreover, composted materials have their own microbial and fungal community and may enhance soil microbial life of plants to which they are added for benefits in terms of improvement in bulk density, water content, and nutrient availability.

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The pinewood nematode - a nightmare for Europe. I. ABRANTES, L. FONSECA. *University of Coimbra, Centre for Functional Ecology - Science for People & the Planet (CFE), Associate Laboratory for Sustainable Land Use and Ecosystem Services-TERRA, Department of Life Sciences, Calçada Martim de Freitas, P-3000 456 Coimbra, Portugal. E-MAIL: isabel.abrantes@uc.pt*

The pinewood nematode (PWN), *Bursaphelenchus xylophilus*, the causal agent of the pine wilt disease (PWD), a quarantine organism by the European and Mediterranean Plant Protection Organisation (EPPO), is recognised worldwide as a major forest pest, endangering European pine forests and the forestry industry. The PWD results from a complex interaction among three organisms: the PWN, its insect vector (*Monochamus* spp.), and a conifer tree. The nematode is the common element in this interaction, while insect vectors and host trees vary from one location to another. This nematode is originated from North America and in the early 20th century has spread first to Japan and later to China, Korea, and Taiwan. In Europe, it was first identified in mainland Portugal, in 1999, and later in Spain and Madeira Island. More recently, it was reported in the Republic of Armenia (Asia). The nematode is transmitted to new hosts by insects, mainly from the genus *Monochamus* (Coleoptera: Cerambycidae), being *M. gal-*

loprovincialis the most widespread species in Europe. The primary susceptible hosts of PWN are coniferous species within the genus *Pinus*, which is widely distributed across the Northern Hemisphere including European species such as *P. mugo* (dwarf mountain pine), *P. nigra* (black pine), *P. pinaster* (maritime pine) and *P. sylvestris* (Scots pine). Nevertheless, the list of susceptible plants also includes coniferous species from the genera *Abies*, *Chamaecyparis*, *Cedrus*, *Larix*, *Picea*, and *Pseudotsuga*. After being transmitted to the host tree by the insect vector, the PWN reproduces and migrates through the vascular system, causing the tree's death. Symptoms in infected plants include reduced resin exudation, yellowing and wilting of needles, and partial or total drying of the crown. The spread of the PWN into non-native areas occurs predominantly due to human activity. Uncontrolled dispersion of this nematode in Europe could lead to severe economic consequences for the forestry industry. Furthermore, the PWN spread could be exacerbated by climate change, which has altered environmental conditions in ways that facilitate the establishment and expansion of both the nematode and its vector. Warmer temperatures, increased droughts, and milder winters create favourable conditions for the nematode reproduction and increase tree susceptibility. Severe symptoms of PWD are typically observed in regions where average summer temperatures exceed 20°C. As climate change continues to progress, the expansion of PWN infected areas across Europe is expected. Urgent adaptive management strategies (mechanical, cultural, biotechnical, chemical, biological, or genetic) are needed. The outbreaks in Europe, along with their negative economic, ecological, and social impacts, serve as a warning to guard against the introduction of invasive alien species into new ecosystems.

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ORAL PRESENTATIONS

Status of plant-parasitic nematodes associated with Citrus in Morocco: spotlight on *Tylenchulus semipenetrans* and a call for Integrated Management. B. ZOUBI^{1,2,3}, A. QADDOURY², A. IRAQI HOUSSEINI¹, K. KHFIF¹, F. MOKRINI³. ¹Laboratory of Biotechnology, Environment, Agri-Food, and Health, Faculty of Sciences Dhar El Mahraz, Sidi Mohammed Ben Abdellah University, Fez 30050, Morocco. ²Center of Agrobiotechnology

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Plant-parasitic nematodes (PPNs) represent a major threat to citrus production worldwide being particularly problematic in Moroccan orchards. Among them, *Tylenchulus semipenetrans*, the causal agent of citrus slow decline, is considered the most destructive species due to its widespread distribution and significant impact on tree vigor, fruit yield, and quality. As a semi-endoparasitic nematode, it colonizes citrus roots, leading to gradual weakening of the tree and long-term yield reduction. To address the lack of recent data on the distribution and diversity of citrus-associated nematodes in Morocco, extensive surveys were conducted between 2018 and 2021 across major citrus-growing regions including Gharb, Moulouya, Souss-Massa, Tadla, Loukkos, Haouz, Marrakech-Safi, Béni Mellal-Khénifra, and Berkane. Soil and root samples collected during the 2018- 2020 campaigns were analyzed for nematode identification and soil physicochemical characterization. A total of 11 genera and 10 species of PPNS were identified. *T. semipenetrans* showed the highest prevalence and reached infestation rates of up to 100% in regions such as Gharb, Loukkos, Moulouya, Souss-Massa, and Tadla. The survey carried out in 2021 reaffirmed these findings, highlighting *T. semipenetrans* as the dominant species in Souss-Massa (88%), Gharb (67%), Berkane (63%), Marrakech-Safi (62%), and Béni Mellal-Khénifra (57%). The distribution of PPNS was significantly correlated with soil physicochemical properties such as soil texture, pH levels, and mineral content. The findings highlight the urgent need for region-specific integrated nematode management programs, incorporating monitoring, sustainable bio-control approaches, and agronomic practices tailored to local soil and climate conditions.

Evaluation of *in vitro* nematocidal effects on *Meloidogyne incognita* of laboratory extracted and commercial essential oils of *Artemisia annua*. E. AJOBIEWE^{1,2}, E. FANELLI², M.P. ARGENTIERI³, F. DE LUCA². ¹Department for Humanistic, Scientific and Social Innovation, University of Basilicata, Via Lanera 20, 75100 Matera, Italy. ²IPSP, National Research Council CNR, Via Amendola 122/D, 70126 Bari, Italy. ³Department of Pharmacy-Drug Sciences, University of Bari Aldo Moro, Bari 70125, Italy. E-MAIL: francesca.deluca@cnr.it

Root-knot nematodes, *Meloidogyne* spp., are considered the most harmful nematodes because of their worldwide occurrence and heavy yield losses caused to a large range of crops. In the last two decades, many experiments have been conducted using essential oils (EOs) against different genera of plant parasitic nematodes. Sweet wormwood, *Artemisia annua* shows great potential of application to produce new nematocidal products due to the presence of a high number of bioactive phytochemicals. The aim of this work is to evaluate the effects against *M. incognita* of laboratory extracted and commercial EOs of *A. annua* collected from different geographical locations. One EO extracted from dried leaves of *A. annua* from South Italy and one commercial EO purchased from India were tested on juveniles (J2s) of *M. incognita*. J2s were exposed for 24 and 48 h to 1000, 2000 and 4000 $\mu\text{g mL}^{-1}$ concentrations of both EOs in an orbital incubator at 70 rpm, at 25 °C. J2 mortality caused by the lab-extracted EO was about 18% at 4000 $\mu\text{g mL}^{-1}$ concentration after 24 h, and 44% after 48 h. J2 mortality caused by the commercial EO was 38% at both 1000 and 2000 $\mu\text{g mL}^{-1}$ after 24 h, and 48% at 2000 $\mu\text{g mL}^{-1}$ after 48 h. Our findings show that the activity profiles of *Artemisia* EOs could be associated with distinct phytochemical compositions, different geographical origin and season of collection. The chemical profiles of the EOs and the molecular mechanisms they trigger in J2s are being studied.

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Investigating biotic and abiotic factors causing a hitherto unknown disorder of olive trees in Southern Italy.

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Olive trees showing extensive leaf yellowing, reddening of the foliage followed by defoliations have been observed at the end of the autumn or in the winter in several new olive plantations in the Southern part of Apulia on FS17 and Leccino. Only these 2 cultivars

resistant to *Xylella fastidiosa* are authorized for planting to restore the devastation caused by the *Xf* epidemic until 2024. Visual inspections and sampling were conducted for 2 consecutive years in 20 three-four years old groves that showed this unusual discoloration of the canopy affecting a limited number of trees or very large areas of the groves. Two or three symptomatic plants per orchard were completely uprooted and mycological analysis were carried out on roots and crown; phytoparasitic nematodes were searched on roots and in the soil. Indeed, samples included symptomatic and asymptomatic leaves and soil for chemical analysis. *Meloidogyne javanica* (root-knot nematodes, RKNs) was the most frequently species of plant parasitic nematodes detected on roots. *Cylindrocarpon* spp. *Fusarium* spp., *Macrophomina phaseolina*, *Rhizoctonia solani* were the mainly soil fungi pathogens detected in highly RKN-infested plants. On the other hand, chemical analyses revealed high sodium content (3062 to 9267 mg kg⁻¹) and potassium deficiency (0.16 to 0.45%) in symptomatic leaves, regardless of the content of these elements in the soil, most likely, as a consequence of the impairment of the root systems caused by the nematode and fungal occurrence. Overall, the symptoms on canopies were more severe than those in soil, with high values of total (351-372 g kg⁻¹) and active (53-135 g kg⁻¹) calcium carbonate.

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In vivo biocontrol efficacy of *Streptomyces violascens* AS2 strain against *Meloidogyne* spp. in tomato.

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Conventional agriculture relies on chemical inputs, disrupting soil health and biodiversity. Climate change has exacerbated ecological collapse. Biocontrol offers an eco-friendly approach by harnessing natural antagonistic mechanisms, notably through filamentous actino-bacteria that produce bioactive compounds. Their application to plant-parasitic nematodes, a major agricultural

threat, demonstrates a significant potential of population suppression. The adoption of actinobacteria-based biocontrol supports sustainable practices, ensuring long-term food security without compromising ecosystem integrity. In the current study, 14 extracts of Actinobacteria were screened *in vitro* for their nematocidal activity against *Meloidogyne* spp. Afterwards, suitable aqueous and ethyl acetate extracts of *Streptomyces violascens* were selected for *in planta* assays to: (i) assess the *in vivo* bionematocidal activity against root-knot nematodes (RKNs), (*Meloidogyne* spp.); (ii) evaluate the plant growth-promoting effect on tomato (*Solanum lycopersicum*); at a gradient of concentration ranging from 400 to 1000 $\mu\text{g ml}^{-1}$. After 3 months of treatment, the *in vivo* trials showed that *S. violascens* aqueous and ethyl acetate extracts significantly reduced the disease index at 1000 $\mu\text{g mL}^{-1}$, by reducing gall formation by about 84% and 69 %, respectively, along with about 65% and 40% decrease in egg numbers, and a substantial decrease of 78% and 54% in second-stage juveniles numbers in the root systems.. *S. violascens* treatment also resulted in a significant enhancement of plant-growth parameters and photosynthetic pigments, indicating a positive impact on overall plant health and vigor. The present findings suggest that the tested actinobacterial extracts could serve as a sustainable resource for managing RKNs, offering a viable alternative to conventional chemical nematicides.

This work was financially supported by the CNRST Labeled Research Unit N°4 grant and the Moroccan (CNRST)- Italian (CNRi) Scientific and Technical Cooperation Program.

New solution to control nematodes affecting key vegetables: Reklmel™ (fluazindolizine). A. FENIO¹, T. THODEN², S. YANNIS³, S. ELLIS⁴, M. BRADASCIO⁵, M. ALIQUO⁶. ¹IFS Field Scientist, Corteva Agriscience Italia srl, via dei Comizi Agrari 10, Cremona, Italy. ²Global Biology Program Leader, Corteva Agriscience Germany GmbH. ³Zonal Biology Leader, Corteva Agriscience Hellas S.A. ⁴Global Regulatory Ecotoxicologist, Corteva Agriscience Limited UK. ⁵Country Regulatory Lead Italy, Corteva Agriscience Italia srl. ⁶Country Marketing Manager, Corteva Agriscience Italia srl. E-MAIL: antonino.fenio@cor-teva.com

Phytopathogenic nematodes cause significant damage to many agricultural crops, especially horticultural ones. The most widespread species in Italy are those belonging to the root-knot genus *Meloidogyne* and the cyst-forming *Globodera* and *Heterodera*, to which some invasive alien species have recently been added. The damage caused to

crops and the impact on farmers income is increasing. The intensification of nematode activity can be traced back to multiple factors such as the lack of availability of effective means of defense, limitations in their use, the emergence of resistance and more recently the effects of climate change. To address these problems farmers have resorted to soil fumigation creating the conditions for the impoverishment of biodiversity and consequent impact on the health of the soil itself. Reklmel is a new sulfonamide nematicide developed by Corteva Agriscience and applied at lower rates than many traditional nematicides. It provides a novel mode of action which specifically targets plant-parasitic nematodes which is vital importance to extending the usefulness of nematode resistance management programs. Nematodes affected by Reklmel show a significant decrease in their activity and ability to infect plant roots which has a notable benefit on plant health and crop yield. Within our talk, we will present also recent data on the compatibility of Reklmel, with various beneficial organism including free-living nematodes, nematode & disease suppressive soil fungi as well as beneficial soil bacteria.

This research was financially supported by Corteva Agrisciences

POSTER PRESENTATIONS

Molecular nematode community analysis reveals novel and known foes in Portuguese vineyards. P. FRAGA¹, J. WILTEN², L. CONCEIÇÃO¹, M.J. CUNHA³, A. BERTAN². ¹University of Coimbra, Centre for Functional Ecology - Science for People & the Planet (CFE), Department of Life Sciences, P-3000 456 Coimbra, Portugal. ²HLB B.V. R&D Molecular Diagnostics Department, Wageningen, The Netherlands. ³Polytechnic University of Coimbra, Research Centre for Natural Resources Environment and Society (CERNAS), High School of Agriculture, Bencanta, P-3045-601 Coimbra, Portugal. E-MAIL: luci@zoo.uc.pt

Concerning vineyards in Portugal, the knowledge of plant parasitic nematodes (PPN) affecting this important crop is scarce. To survey PPN and free-living nematodes associated to this crop, 13 root-adjacent soil samples from vineyards in Portugal were collected and their nematode communities extracted. In collaboration with University of Coimbra, HLB B.V. received nematode communities and performed DNA isolation, full-length 18S rDNA SSU amplification, Nanopore sequencing, nematode SSU database searches and data analysis for these 13 samples. Primer sequences were trimmed

from the demultiplexed data using Porechop and used as input for the Decona pipeline. Further, reads were filtered, clustered (min. 20 reads per cluster) and polished cluster consensus sequences generated. The BLAST analysis of the reads against a nematode SSU database comprising more than 5000 sequences retrieved 151 amplified sequence variants (ASVs) matching 125 distinct taxa. The average number of mapped reads per sample with a minimum identity percentage of 97% was 67,240 (avg. 42 ASVs per sample) and the avg. read quality (mean Q) was 17.85. Further, beta diversity and Shannon's biodiversity indices were calculated and nematode community similarity across samples was assessed based on PCoA, NMDS and hierarchical clustering methods. *Helicotylenchus* spp. and *Tylenchulus semipenetrans* were found in 10 and 11 of the 13 samples, respectively. *Xiphinema pachtaicum*, *Ditylenchus destructor* and *Meloidogyne hapla* were also found. Based on the NGS data, molecular detection tests are being developed for each of the 5 species of the genus *Helicotylenchus* present in the samples.

This work was supported by FCT - Fundação para a Ciência e Tecnologia, I.P., in the framework of the Project UIDB/04004/2025 - Centre for Functional Ecology - Science for the People & the Planet and Instituto do Ambiente, Tecnologia e Vida.

Resistance of *Solanum linnaeanum* to *Meloidogyne* spp. B. SIOPA, L. CONCEIÇÃO. *University of Coimbra, Centre for Functional Ecology - Science for People & the Planet (CFE), Department of Life Sciences, P-3000 456 Coimbra, Portugal.* E-MAIL: luci@zoo.uc.pt

Root-knot nematodes (RKNs), *Meloidogyne* spp., are important crop pests that cause severe losses in crop production worldwide, reducing both productivity and crop quality. Some species are considered quarantine organisms by the European and Mediterranean Plant Protection Organization. The development of nonchemical and sustainable management strategies to reduce nematode damage is crucial. *Solanum linnaeanum* was already considered resistant to *M. chitwoodi*, with an average of 519 small galls/plant and with 45% adult nematodes, all males, inside the roots. The resistance of this *Solanaceae* species was assessed against the most common RKN species, *M. hapla*, *M. incognita* and *M. javanica*. Five replicates of *S. linnaeanum* and 5 replicates of *S. lycopersicum* cv. Coração de Boi (control) were inoculated with a suspension of eggs and juveniles of each RKN species. Two separated bioassays were performed for each RKN species. Gall index, the Bridge and

Page rating chart and reproduction factor are being estimated and will be presented. The use of *S. linnaeanum* as a new source of resistance is a good alternative for the management of RKNs in search of nonchemical and sustainable strategies to protect crops.

This work was supported by FCT - Fundação para a Ciência e Tecnologia, I.P., in the framework of the Project UIDB/04004/2025 - Centre for Functional Ecology - Science for the People & the Planet and Instituto do Ambiente, Tecnologia e Vida.

PINEMARK – Pine resistance/susceptibility proteomic biomarkers to *Bursaphelenchus xylophilus* and *B. mucronatus* infection under climate change scenarios.

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The pinewood nematode (PWN), *Bursaphelenchus xylophilus*, the causal agent of pine wilt disease, is a quarantine organism in Europe. Its closely related species, *Bursaphelenchus mucronatus*, which shares similar ecological and morphological features with PWN, is considered to have low pathogenicity but is also capable of killing trees under specific environmental conditions. Climate changes have raised concerns about how these nematode species would interact with stressed pine hosts. Understanding these interactions is crucial for developing successful management strategies. This study aims to assess the effects of drought and high temperatures on the proteomic profiles of pine species with differing susceptibilities to *B. xylophilus* infection and to identify susceptibility and resistance biomarkers. These objectives have been approached through comparative proteomics of PWN and *B. mucronatus*-infected and non-infected *Pinus pinaster* (highly susceptible) and *P. pinea* (low susceptible) under different temperatures (25–30°C) and varying water availability conditions (well-watered/water-stressed). The identification of differentially expressed proteins and molecular pathways associated with pines response to infection will reveal resistance and susceptibility biomarkers, providing new insights into host-nematode interactions. Identified host resistance and susceptibility biomarkers will be useful for selecting more resistant trees to nematode infection under climate change

scenarios and improving the management strategies for this important forest disease.

This research was financially supported by FEDER-Portugal 2020, COMPETE 2020 and by FCT projects, PineWALL, PTDC/ASP-SIL/3142/2020; UIDB/04004/2025 (DOI: 10.54499/UIDB/04004/2025); UIDP/04004/2025 (DOI: 10.54499/UIDP/04004/2025); TERRA - LA/P/0092/20205 and Instituto do Ambiente, Tecnologia e Vida. Hugo Silva (Grant - 2023.03527.BD) is funded by FCT, European Social Fund (ESF), under the Programa Demografia, Qualificações e Inclusão (PDQI) – Portugal2030.

Effect of temperature and water availability on *Bursaphelenchus xylophilus* and *B. mucronatus* pathogenicity. H. SILVA¹, J.M.S. CARDOSO¹, B. MANADAS², L. FONSECA^{1,3}. ¹Centre for Functional Ecology - Science for People and the Planet, TERRA Associate Laboratory, Faculty of Science and Technology, University of Coimbra, Portugal. ²CNC - Center for Neuroscience and Cell Biology, University of Coimbra, Portugal. ³FITOLAB - Laboratory for Phytopathology, Instituto Pedro Nunes (IPN), Coimbra, Portugal. E-MAIL: hugoffs.95@gmail.com

Pine wilt disease, caused by pinewood nematode (*Bursaphelenchus xylophilus*), poses a significant threat to forests, particularly under climate change scenarios. This study evaluated the pathogenicity of *B. xylophilus* and its closely related species, *B. mucronatus* that is a species with similar morphological and ecological traits although considered as a weaker pathogen. Impact of these nematodes was assessed in *Pinus pinaster* and *P. pinea* under different temperature and water availability conditions. Two- to three-year-old seedlings of both pine species were inoculated with 3,000 nematodes (*B. xylophilus* or *B. mucronatus*) and exposed to 2 temperature conditions (25°C and 30°C) and 2 water availability regimes (water- stressed and well-watered). Disease symptoms were monitored for 40 days, and soil water content was recorded. At the end of monitoring, nematodes were extracted from aerial parts and roots, and quantified per gram of dry wood. Results showed that *P. pinea* exhibited no symptoms, and few nematodes of both species were recovered. However, *P. pinaster* was highly susceptible to *B. xylophilus*, showing symptoms few days after inoculation with high number of nematodes recovered. In *B. mucronatus*-inoculated *P. pinaster*, heavy symptoms appeared at 30°C under water-stressed conditions, with high nematode populations recovered. This study highlights the significance of temperature and water availability in determining the pathogenicity of *B. xylophilus* and *B. mucronatus*. Additionally, it

reveals that *B. mucronatus*, despite being considered a relatively weak pathogen, may exhibit high pathogenicity to *P. pinaster* under certain conditions, suggesting that this nematode species could emerge as a significant threat to pine forests in a context of climate change.

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Diversity and role of plant-parasitic and free-living nematode communities in Italian vineyards. A. VOVLAS¹, A. TROCCOLI¹, E. FANELLI¹, V. PAPESCHI², F. DE LUCA¹. ¹IPSP, National Research Council CNR, Via Amendola 122/D, 70126 Bari, Italy. ²Servizio Tecnico di Condifesa Lombardia Nord Est, Via Malta 12, Brescia, Italy. E-MAIL: alessiovovlas@cnr.it

Nematodes represent a diverse and widespread group of invertebrates, largely studied for their economic and ecological relevance. Their identification is important to arrange the proper management strategies that should be designed to maintain soil health and fertility, as well. A wide survey was conducted in 2024 in North Italy to study plant parasitic (PPNs) and free-living nematodes (FLNs) associated with *Vitis vinifera* vineyards. This study aimed to determine the prevalence and distribution of PPNs and FLNs in vineyards. Moreover, explanatory variables were used, to describe the influence of soil, climate and agricultural management factors in structuring the variation of nematode community composition. Fifty- three sampling sites from ten different vineyards were surveyed, and nematode species and genera were identified using an integrative approach. A total of approximately 3000 soil nematode individuals were found belonging to 16 different genera. The diversity of nematode communities associated with soil samples was determined using several ecological indices such as Dominance, Shannon-Wiener Index, Evenness, Margaleff, Equitability, Enrichment Index (EI), and Maturity Index (MI). The Nematode Indicator Joint Analysis (NINJA) tool, structural nematode dynamics, Correspondence and Food web analyses were also applied. The results obtained provide novel insights into the biodiversity and dynamics of terrestrial nematodes associated with Italian vineyards. The contribution of

such biodiversity as an effective soil health bioindicator will be presented.

This research was partially funded by NEMAGEST project funded by Lombardy Region (Bando 2018 per Progetti di ricerca in campo agricolo e forestale – d.d.s. n. 4403 del 28/03/2018).

Morphological and molecular characterization of reniform nematodes in the olive orchard of central Italy.

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Reniform nematodes belonging to the genus *Rotylenchulus* are semi-endoparasites that affect the roots of various herbaceous and woody plants. The genus *Rotylenchulus* Linford & Oliveira, 1940 comprises 10 valid species, showing high intraspecific variability of some diagnostic features, making identification, based only on morphology, a difficult task. Nematode surveys of plant-parasitic nematodes in Italy, undertaken during the last few years on several crops, including sugarcane, sorghum, maize, grapevine, olive, and wild olive have revealed feeder-root infections and heavy soil infestations of *Rotylenchulus* spp. The main objective of this study was to investigate the biodiversity of reniform nematodes in olive orchards of central Italy, conducted with a traditional management strategy. Reniform nematodes recovered from two olive groves were characterized using a polyphasic approach, combining morphological and molecular characterization. Morphological and morphometric analyses identified at species level reniform nematodes in both sampling sites as *Rotylenchulus macrodorus*. Molecular analyses, based on D2-D3 segments of 28S rRNA gene and mitochondrial COI confirmed the high intraspecific and interspecific variability within Italian *Rotylenchulus* populations. Blast search based on the D2-D3 sequences revealed that one population showed a 97-98% similarity with the corresponding Italian populations of *R. macrodorus* from GenBank, while the second population showed 95% similarity suggesting the occurrence of a species complex for *R. macrodorus* or a high intraspecific variability. Further molecular analyses are ongoing to correctly identify these nematodes that are frequently ignored or misdiagnosed as pests, and this may potentially pose a threat to food security.

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O-Live", under grant agreement No. 101091255 (Soil Deal for Europe – HORIZON-MISS- 2021-SOIL-02-03) [AV] and partially by Agritech National Research Center funding from the EU Next- Generation EU (Piano Nazionale di Ripresa e Resilienza (PNRR) – Missione 4 Comp. 2, Invest 1.4 – D.D. 1032 17/06/2022, CN00000022) [EA].

Nematocidal activity of the alpine medicinal plant *Peucedanum ostruthium* against the root-knot nematode *Meloidogyne incognita*.

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Restrictions on the use of chemical pesticides have raised growing interest in the study of plant-derived nematocidal compounds. The potential nematocidal properties of *Peucedanum ostruthium* (*Apiaceae*) were tested by 2% ethanol plant extracts *in vitro* and in pot experiments. Infective juveniles (J2s) were exposed for 4, 8, 24, 48 and 96 h to 62.5, 125, 250, 500, 1000 and 2000 µg mL⁻¹ concentrations of both root and leaf extracts, including distilled water and a chemical nematicide as controls. Nematode mortality reached more than 98% after 24-hour treatment with 500, 1000 and 2000 µg mL⁻¹ solutions of root extract and 94% after 48-hour treatment with 1000 and 2000 µg mL⁻¹ solutions of leaf extract. To assess the ability to infect, juveniles were exposed for 24 h to the LD50 of both extracts, then rinsed and inoculated in tomato plants kept in growth chamber. Thirty days after inoculation, nematode infection was significantly reduced in roots inoculated with root extract treated J2s. The suppressiveness of *P. ostruthium* extracts was also investigated by irrigating tomato plants in 50 mL pots with 5 mL of 4000 µg mL⁻¹ solutions at inoculation and 2 weeks later. At the end of nematode life cycle, infestation on tomato roots was reduced and, particularly, a significant reduction of the eggs/egg mass numbers was observed. Results from this study show a high nematocidal potential of *P. ostruthium* root extract, which can be exploited for the formulation of new effective nematicides.

This study was funded by PNRR Agritech, task 3.2.5 'Nature-based solutions for natural resources and environment protection'.

Toxicity of Asteraceae plants from the Anatolian region to the Root-Knot Nematode *Meloidogyne incognita*. G. AYDINLI¹, T. D'ADDABBO², M. KOÇ³, M.P. ARGENTIERI⁴, S. MENNAN⁵, P. VERONICO². ¹Ondokuz Mayıs University, Bafra Vocational High School, Samsun, Türkiye. ²Institute for Sustainable Plant Protection, National Council of Research, Bari, Italy. ³Ankara Yıldırım Beyazıt University, Public Health Institute, Department of Traditional, Complementary and Integrative Medicine, Ankara, Türkiye. ⁴Department of Pharmacy—Drug Sciences, University of Bari, Bari, Italy. ⁵Ondokuz Mayıs University, Faculty of Agriculture, Department of Plant Protection, Samsun, Türkiye. E-MAIL: pasqua.veronico@cnr.it

Asteraceae plants from the endemic flora of the Anatolian region are a reservoir of still unexplored sources of bioactive compounds that may be used for the control of root-knot nematodes (*Meloidogyne* spp.). The aim of this study was to assess the toxicity to the infective juveniles (J2s) of the root-knot nematode *M. incognita* of 2 extracts from Asteraceae spp.: one from *Helichrysum noeantum* and one from *Tanacetum nitens*. The nematode J2s were exposed for 24 and 48 h to 62.5, 125, 250, 500, 1000 and 2000 µg mL⁻¹ concentrations of each plant extract, including distilled water and a 2% ethanol solution as controls. J2 motility and mortality were checked microscopically at the end of each exposure time. About 90% of the *M. incognita* J2s were immobilized by a 24-hour exposure to the 3 and 2 highest concentrations of *T. nitens* and *H. noeantum* extracts, respectively. At the same concentrations, all the J2s treated with *H. noeantum* extract recovered their mobility after a 24-hour permanence in water, while 13–26% mortality rates occurred for the J2s exposed to *T. nitens* extract. Nematode mortality reached 93 and 55% after 48-hour treatment with 1000 µg mL⁻¹ solution of *T. nitens* and *H. noeantum*, respectively. Both extracts did not show any toxicity to J2s when used at concentrations ≤ 250 µg mL⁻¹. The results indicate a higher suitability of *T. nitens* plant extracts to a potential formulation of nematocidal products. This study was carried out within the frame of a bilateral project CNR-TUBITAK “The nematocidal potential of endemic species of Asteraceae in Türkiye”

CONCURRENT SESSION D2 – The role of pathogens in forest tree mortality in times of global change

SESSION KEYNOTES

Climate acts as an environmental filter to forest pathogens. M. CABALLOL^{1,2,3}, M.A. REDONDO⁴, N. CATALÁN⁵, T. CORCOBADO^{6,7}, T. JUNG⁶, B. MARÇAIS⁸, I. MILENKOVIĆ^{6,9}, M. NEMESIO-GORRIZ¹⁰, J. STENLID⁴, J. OLIVA^{1,2}. ¹Department of Agricultural and Forest Sciences and Engineering, University of Lleida, 25198, Lleida, Spain. ²Joint Research Unit CTFC – AGROTECNIO-CERCA, 25198, Lleida, Spain. ³Current address: Forest Science and Technology Centre of Catalonia (CTFC), 25280, Solsona, Spain. ⁴Department of Forest Mycology and Plant Pathology, Swedish University of Agricultural Sciences, Box 7026, 750 07, Uppsala, Sweden. ⁵Institute of Environmental Assessment and Water Research, IDAEA-CSIC, 08034, Barcelona, Spain. ⁶Mendel University in Brno, Faculty of Forestry and Wood Technology, Department of Forest Protection and Wildlife Management, Phytophthora Research Centre, 613 00, Brno, Czech Republic. ⁷Austrian Research Centre for Forests (BFW), Seckendorff-Gudent-Weg 8, 1131, Vienna, Austria. ⁸Université de Lorraine - Inrae, UMR Interactions Arbres/Microorganismes, 54000, Nancy, France. ⁹University of Belgrade-Faculty of Forestry, 11030, Belgrade, Serbia. ¹⁰Forestry Development Department, Teagasc, D15DY05, Dublin, Ireland. E-MAIL: maria.caballol@ctfc.cat

Global change and forest pathogens pose a combined and often synergistic threat to natural ecosystems worldwide. The number of reports of forest diseases and tree mortality has increased worldwide over the past decades. Global change has contributed to this increase by introducing forest pathogens to new areas and favouring their range expansion due to changes in the climate. Understanding how the environment influences the distribution of forest pathogens is paramount given the upraising effects of climate change. Many studies have predicted an expansion of the distribution of forest pathogens with increasing temperatures. However, predictions based on single-species models considering pathogens as independent and static entities may not fully represent the complexity of their interactions in nature. The functional trait-based approach has emerged as a powerful tool for explaining community patterns and could be used to predict how pathogen communities may respond to environmental changes based on their functional traits. While this approach has been applied to plant-associated microbes such as bacteria and mycor-

rhizal and endophytic fungi, the role of climate on plant pathogens has been rarely explored from a community ecology perspective. The genus *Phytophthora* comprises some of the most damaging plant pathogens of agricultural, forestry and natural ecosystems. The ecologically diverse array of species included in this genus provides a good opportunity to study how the environment influences the composition of communities. We explored the role of climate in the assembly of *Phytophthora* species at >250 river sites across two gradients, a latitudinal gradient spanning from Mediterranean to Arctic conditions, and an altitudinal gradient including the Spanish Pyrenees. *Phytophthora* communities were obtained by sequencing the full internal transcribed spacer (ITS) region of river filtrates. We tested whether the biogeography and seasonal variation of *Phytophthora* was linked to climatic variables and whether other environmental factors such as water chemistry and vegetation diversity played a role. In this talk, we present how climate acts as an environmental filter for the assembly of *Phytophthora* communities. We show how different mechanisms occur in southern and northern latitudes, and how functional traits related to morphological and physiological characteristics of *Phytophthora* species explain community assembly processes. At northern latitudes, the cold climate restricts species diversity and acts as a filter for *Phytophthora* distribution, selecting species with a low minimum temperature limit for growth. By contrast, at southern latitudes, a dry climate poses a strong environmental filter for *Phytophthora* communities which results in the dominance of drought-tolerant species with thick oospore walls and a high optimum temperature for growth, and a high maximum temperature limit for growth. The ability to survive under cold conditions seems to be key for colonising northern latitudes while the ability to cope with dry conditions seems crucial for colonising southern latitudes. These findings indicate that considering functional traits can improve the predictability of how pathogens will respond to future climate change scenarios.

Forest pathogens in the era of global change: disrupting the holobiont and reshaping ecosystems. C. MORALES-RODRIGUEZ. *Department for Innovation in Biological, agro-food and forest systems (DIBAF) University of Tuscia. Via San Camillo de Lellis snc, VT, Italy.* E-MAIL: cmorales@unitus.it

Forest ecosystems are under mounting pressure from global change, which acts through a convergence of drivers such as climate shifts, biological invasions, increased

human mobility, and landscape fragmentation. These factors do not merely add new stressors; they profoundly reshape the structure and function of ecological networks, including those that govern the relationships between forest trees and their associated microorganisms. The increasing frequency and severity of tree mortality events linked to pathogens underscore the need for a deeper and more integrated understanding of forest health. In this context, the holobiont framework, provides an innovative perspective for interpreting the outcomes of host-microbe-pathogen interactions in dynamically changing environments. Global change can disrupt these interactions, tipping the balance between symbiosis, commensalism, and pathogenicity. Climate-induced shifts in temperature regimes are driving the expansion of geographic ranges and phenological shifts in pathogens. Simultaneously, biological invasions introduce novel species that can destabilize established microbial communities, with cascading effects on ecosystem resilience. Understanding how these microbial networks respond to global change is essential for predicting emerging disease outbreaks and implementing effective forest management strategies. This contribution emphasizes the need to transcend classical paradigms, advocating for a more systems-level approach that integrates microbial ecology, climate science, invasion biology, and adaptive forest management. Such approaches are increasingly facilitated by advances in high-throughput sequencing, metagenomics, and modelling tools that can unravel the complexity of host-microbiota-pathogen systems. As a representative example, we explore the chestnut forest system of central Italy, which has experienced the compounding effects of global change drivers over recent decades. In this system, the introduction of the gall wasp *Dryocosmus kuriphilus* has indirectly influenced the fungal communities already present, such as *Gnomoniopsis castanea*, concurrently altering the native microbial communities associated with chestnut tissues. In addition to the effect of the introduction of a new species, we will see the influence of global warming on the distribution of soilborne pathogens such as *Phytophthora*, which cause chestnut ink blight. These changes have led to unexpected disease patterns and declines in tree vitality, illustrating how biotic and abiotic stressors can synergistically destabilize forest holobionts. In conclusion, forest pathogens should be studied within the broader context of microbial and environmental interactions that define the holobiont. Addressing forest health in the Anthropocene requires not only pathogen surveillance but also a comprehensive understanding of the host-associated microbiota and its role in ecosystem resilience. By embracing this integrated framework, researchers and

forest managers can better anticipate future disease scenarios and design more effective mitigation strategies.

The rise of emerging pathogens in a changing world: the case of *Cryptostroma corticale*. R. SCHLOESSER^{1,2}, S. BIEN², J. BUSSKAMP², G.J. LANGER², E.J. LANGER³, L. GHELARDINI^{4,5}, A.L. PEPORI¹, A. SANTINI¹. ¹IPSP, National Research Council CNR, Via Madonna del Piano 10, 50127 Sesto Fiorentino, FI, Italy. ²NW-FVA, Northwest German Forest Research Institute, Grätzelstraße 2, 37079 Göttingen, Germany. ³University of Kassel, Department of Ecology, Heinrich-Plett-Straße 40, D-34132 Kassel, Germany. ⁴University of Florence, Department of Agriculture, Food, Environment and Forestry (DAGRI), Florence, Italy. ⁵National Biodiversity Future Center (NBFC), Palermo, Italy. E-MAIL: rebe-kaschloesser@cnr.it

Climate change is progressively stressing and debilitating trees, encouraging the emergence of pathogens previously considered secondary as, for example, *Diplodia sapinea*, *Biscogniauxia nummularia*, *B. mediterranea* and *Cryptostroma corticale*. *C. corticale*, the causal agent of the Sooty Bark Disease (SBD), has been present in Europe at least since the middle of the 20th century. The pathogen had been reported from several central European countries in the 1950s and 1960s, causing mostly minor and localised damage, however, started causing large scale damages after the extreme drought years 2018-2020. According to literature, the pathogen emerges in cyclic patterns following years with above-average summer temperatures. SBD mainly causes damage on *Acer pseudoplatanus* but can also infect other native *Acer* species as well as *Aesculus hippocastanum*. As temperatures are expected to rise further, especially in the Mediterranean area, further outbreaks of secondary pathogens can be envisaged in the future. Since the start of the latest large-scale outbreak of SBD in Europe, interest in SBD has increased, and several papers regarding dispersion and potential management have been published. The disease has also come to the attention of the general public, as the spores produced by *C. corticale* can cause an allergic reaction in humans referred to as 'maple bark strippers' disease. As breakouts of SBD have been reported from many different regions, it was of interest to investigate whether *C. corticale* might have an endophytic life stage and thus be dispersed much farther than observed by symptomatic trees.

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sumer Protection and located at the Northwest German Forest Research Institute (NW-FVA), and by the Phytosanitary service of the Regione Toscana, Italy.

ORAL PRESENTATIONS

Integrative analysis of bacterial pathogens causing acute oak decline on Portuguese fagaceae forests. L. CRUZ^{1,2}, L. DUARTE¹, D. MCGUIRE^{1,2}, A. MADURO¹, E. SOUSA^{1,3}, P. NAVES^{1,3}. ¹National Institute for Agrarian and Veterinarian Research, Oeiras, Portugal. ²BioSystems & Integrative Sciences Institute (BioISI), University of Lisbon, Lisbon, Portugal. ³GREEN-IT – Bioresources for Sustainability. E-MAIL: leonor.cruz@iniav.pt

Since the first report of acute oak decline agent *Brenneria goodwinii* affecting *Quercus suber* in Portugal (2018), an increasing number of similar findings on other Fagaceae species, namely *Quercus ilex* and *Quercus pyrenaica*, have been observed in distinct geographic areas of the country. Over 100 samples collected from trunks, twigs, acorns and insects from the Buprestidae, Dermestidae and Cerambycidae (Coleoptera) families were laboratory tested using isolation and biomolecular tools to identify the putative agents involved in this decline syndrome. Additionally, cross pathogenicity tests were made for selected strains under confinement conditions on healthy plantlets of *Q. suber*, *Q. ilex* and *Q. pyrenaica* using reference strains of *B. goodwinii* and *Lonsdalea* spp., as controls. Whole Genome Sequencing (WGS) was further implemented on a set of strains selected based on the host species, location and pathogen. Relative frequency of positive isolation and PCR tests showed differences between host species. *Q. ilex* displayed the lowest infection level (5.71%), *Q. suber* 14.29% and *Q. pyrenaica* showed the highest disease rate (17.24%). These tests highlighted the presence of distinct bacterial pathogens affecting stands and isolated trees, while cross pathogenicity tests disclosed specific host-pathogen interaction patterns. Additionally, WGS confirmed the presence, among other bacteria, of pathogenic strains of *Lonsdalea iberica*, *B. goodwinii* and *Rahnella victoriana*, affecting these hosts. This study brings to light relevant epidemiological aspects of acute oak decline, contributing to clarify the presence, diversity and risk of dispersion of these pathogens in south-western Europe Mediterranean forests, where distinct Fagaceae species coexist.

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Phylogenetic structure, host abundance, and micro-climate predict establishment of *Fusarium dieback* - invasive shothole borers in California. S. LYNCH¹, M. BUSTAMANTE¹, K. EL FAR¹, C. CARACHURE¹, A. ADASKAVEG¹, J. KABASHIMA², C. SHOGREN², A. ESKALEN¹. ¹University of California, Davis, United States of America; ²University of California, Agricultural and Natural Resources, University of California, United States of America. E-MAIL: sclynch@ucdavis.edu

Pine ghost canker is a recently described disease affecting multiple pine species in urban forests of Southern California. Symptoms include wedged cankers with irregular margins and cryptic discoloration on cross-sections of branches, which can lead to severe dieback and potentially tree death. In this study, we identified and characterized five *Neofusicoccum* species (*N. luteum*, *N. mediterraneum*, *N. parvum*, *N. stellenboschianum*, and *N. vitifusiforme*) as the primary etiological agents of pine ghost canker. These pathogens were consistently isolated from multiple symptomatic pine samples (n = 41) and identified by morphology and phylogenetic analyses using four DNA barcodes (rDNA ITS, *tef1*, *tub2*, and *rpb2*). Pathogenicity was confirmed on healthy branches of 15-year-old Monterey pines, where the five *Neofusicoccum* species, caused vascular lesions that were not significantly different in length. Secondary fungi (*Diaporthe*, *Diplodia*, *Neopestalotiopsis*, and *Pestalotiopsis* spp.) were also recovered from symptomatic tissues but did not cause vascular lesions in pathogenicity tests. The optimal temperature for mycelial growth of *N. luteum* and *N. parvum* was 30°C, whereas for *N. mediterraneum*, *N. stellenboschianum* and *N. vitifusiforme*, it was 25°C. All five species were able to resume growth at room temperature (20°C) after showing no growth during a 7-day exposure to 5°C and 40°C. This study constitutes the first report of *N. luteum*, *N. stellenboschianum*, and *N. vitifusiforme* causing pine ghost canker in California. Environmental factors such as warmer temperatures, irrigation, and pest infestations are discussed as drivers

of disease expression in pine trees. Management practices are also proposed.

First detection of acute oak decline associated bacteria in holm oak of Salento: environmental drivers and disease implications. A. BENE¹, G. CARLUCCIO¹, M. VERGINE¹, E. SABELLA¹, A.G. DELLE DONNE², L. DE BELLIS¹, A. LUVISI¹. ¹Department of Biological and Environmental Sciences and Technologies, University of Salento, 73100 Lecce, Italy. ²Department of Agriculture, Rural Development and Environment, Sec. Osservatorio Fitosanitario, 70125 Bari, Italy. E-MAIL: alessandro.bene@unisalento.it

Acute Oak Decline is a complex disease that affects oak trees, including Holm oak (*Quercus ilex* L.), representing a new challenge for the protection of European and Mediterranean forests. Pathogens related to AOD in Holm oak were analysed in the Salento peninsula, southern Italy, in two different contexts: urban and rural environment. The results showed a difference in symptomatology where urban trees showed crown desiccation and insect attacks, such as *Kermes* spp. and *Nidularia* spp., while rural trees showed characteristic AOD symptoms, including vertical bark cracks with dark exudates, larval galleries, and insect exit holes associated with Buprestidae. In addition, the presence of three key disease bacteria *Brenneria goodwinii*, *Gibbsiella quercinecans*, and *Rahnella victoriana* was confirmed, but only in samples from rural trees. Pathogenicity tests confirmed *B. goodwinii* and *G. quercinecans* as the causative agents of AOD in Holm oak, having fulfilled all of Koch's postulates. In addition, phylogenetic tests showed a high genetic similarity between these strains and those widespread in Europe and Iran, suggesting the possibility of wide-spread dissemination via vector insects or plant material. Thus, while bacteria play a fundamental role, the multifactorial nature of AOD imposes the need to investigate all other possible abiotic, such as drought and poor soil conditions, and biotic actors as well. Future investigations should focus on monitoring the spread of AOD, determining its impact on Mediterranean forests.

Hidden phytosanitary threat within internationally exchanged tree seed. I. FRANIĆ¹, M. CLEARY², R. ESCHEN³, A. PEREZ-SIERRA⁴, S. PROSPERO¹. ¹Swiss Federal Research Institute WSL, Birmensdorf, Switzerland. ²Swedish University of Agricultural Sciences, Alnarp, Sweden. ³CABI, Delémont, Switzerland. ⁴Instituto Valenciano

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Although several major tree disease outbreaks in forests and plantations have been caused by pathogens that are also known as seed-borne, tree seeds are generally considered a low-risk pathway for introducing plant-pathogenic fungi, and their movement remains largely unregulated. However, our recent analysis of the international tree seed trade revealed that approximately 2.5 million kilograms of tree seeds are exchanged between countries each year. Due to climate change-driven seed shortages, we anticipate that the volume of international seed trade will increase in the future. This is particularly concerning because our recent molecular studies, which examined multiple tree species originating from several continents, demonstrated that tree seeds harbour diverse fungal communities, including known pathogens. Furthermore, these seed-associated fungal communities were primarily structured by the host species rather than environmental factors, indicating strong host specificity, likely due to confirmed frequent vertical transmission. These findings suggest a significant risk of pathogen introduction through seed trade, with a higher likelihood of establishment in new areas if suitable host species are present. Nonetheless, further research is needed to understand the impact of seed-borne fungi in new environments and to develop effective risk management strategies.

POSTER PRESENTATIONS

Emerging fungal pathogens associated with holm oak decline in Southern Italy. C. DEL GROSSO^{1,2}, D. PALMIERI¹, L. MARCHESE¹, L. MELISSANO³, G. LIMA¹. ¹Department of Agricultural, Environmental and Food Sciences, University of Molise, 86100 Campobasso, Italy. ²Institute for Sustainable Plant Protection, National Research Council (CNR), 70126 Bari, Italy. ³Department of Agriculture, Environment and Rural Development, Sustainable Management and Protection of Natural and Forest Resources, Apulia Region, 70100 Bari, Italy. E-MAIL: lima@unimol.it, carmine.delgrosso@cnr.it

Holm oak (*Quercus ilex* L.) forests, iconic elements of the Mediterranean landscape, are increasingly threatened by complex decline phenomena. Since 2016, extensive dieback symptoms, such as cankers, branch necrosis, and crown thinning, have been observed in holm oak stands across the Salento Peninsula (Southern Italy), raising concerns about forest resilience under cli-

mate change. This study aimed to identify fungal isolates associated with holm oak decline and assess their pathogenic potential. Several fungal species of the Botryosphaeriaceae family, as in particular *Diplodia corticola* (Dc), *Diplodia quercivora* (Dq), and *Neofusicoccum vitifusiforme* (Nv), were identified. Pathogenicity tests on artificially stressed and inoculated holm oak seedlings revealed that Dc and Dq induced severe subcortical and leaf margin necrosis, while Nv induced similar but milder symptoms, suggesting its behaviour as a weak pathogen. These findings confirm the important role of Botryosphaeriaceae in holm oak decline but report for the first time Dq and Nv as *Q. ilex* pathogens in Italy. These pathogens may act within a broader decline syndrome in which abiotic stress factors, particularly alternating periods of droughts, heavy rainfall and heat waves, severely compromise tree vitality, favouring opportunistic infections. Furthermore, recent reports identifying bacterial agents associated with Acute Oak Decline (AOD) in the same geographical area, highlight the need for integrated and multidisciplinary approaches to monitor and manage this complex forestry issue and to develop the most appropriate strategies to counteract holm oak decline in Mediterranean forests.

The findings presented are based on a study published in *Journal of Fungi*, DOI: <https://doi.org/10.3390/jof10010035>.

Plasticity responses of *Hymenoschyphus fraxineus* populations to thermal stress in the Italian peninsula. L. STAZIONE^{1*}, C. AGLIETTI^{1*}, A.L. PEPORI², F. PECORI², G. GRAHAM¹, L. GHELARDINI¹, A. SANTINI². ¹Department of Agricultural, Food, Environmental and Forest Sciences and Technologies (DAGRI), University of Florence, Piazzale delle Cascine 18, 50144 Florence, Italy. ²National Research Council (CNR), Institute for Sustainable Plant Protection, Via Madonna del Piano, 10, 50019 Sesto Fiorentino, Italy. E-MAIL: leoneldaniel.stazione@unifi.it. *Equal contribution

Global change is a complex trend affecting the spread and establishment of non-native species. Among these, the invasive fungal pathogen *Hymenoschyphus fraxineus* was reported as a challenge threatening natural ash (*Fraxinus* sp.) populations of the whole Europe. In Italy, *H. fraxineus* was recently reported on *Fraxinus excelsior* at the southward of the country, being able of colonizing mediterranean habitats previously considered far from the growth optimum of the species. There is a gap of knowledge about *H. fraxineus* adaptability and distribution in Italy, also due to the highly fragmented geo-

graphic presence of the host, especially in the south of the country. The aim of this work is to characterize populational adaptive traits of *H. fraxineus* along the Italian peninsula. Samples were collected from symptomatic *F. excelsior* along the distribution of ash populations and fungal strains were identified by sequencing of nuclear and mitochondrial loci. The phenotypic responses were studied to assess optimal range and thermal stress tolerance by testing the mycelial growth of each strain against heat and cold thermal treatments, as well as their virulence by inoculation into the host tissues. Preliminary results clarify the possibility of further spread of the pathogen in the Mediterranean area, contributing to the understanding of the distribution and potential adaptive responses of invasive fungal pathogens in the context of fluctuating temperatures. Results will be useful to build an effective prevention and forest management strategy, helping the conservation of ash biodiversity richness at the southern edge of the species' natural distribution.

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Preliminary findings of ophiostomatoid fungi from bark beetle galleries of the Bakhmaro Resort Forest, Georgia. T. ABRAMISHVILI¹, D. GAGANIDZE¹, M. GIORBELIDZE¹, A. DADEGASVILI¹, N. KHARABADZE², M. BURJANADZE², J. FOIT³, K. TOMANKOVA⁴. ¹Department of Plant Pest Diagnostic, State Laboratory of Agriculture, Tbilisi, Georgia. ²Vasil Gulisashvili Institute of Forestry, Agricultural University of Georgia, Tbilisi, Georgia. ³Department of Forest protection and management of environmental, Mendel University, Brno, Czech Republic. ⁴Central Institute for Supervising and Testing in Agriculture, Olomouc, Czech Republic. E-MAIL: tea.abramishvili@sla.gov.ge

Bark beetles are dangerous pests of forest crops and vectors of pathogens causing phytopathogenic fungal diseases. They introduce the fungal spores with their body into the passages made in the plant, where they cover the vascular fiber cones and hinder the physiological processes. Finally, the plant withers. The aim of the study is to detect and identify bark beetles as vectors of phytopathogenic fungi and the fungi spread by them in the coniferous forests of the resort zone Bakhmaro of Guria region of Western Georgia. The bark beetles and fungi were identified by morphological and molecular

methods (sequencing the ITS 1 and ITS2 2 regions of the ribosomal RNA operon 5.8S gene). Seven bark beetle species were identified: *Ips typographus*, *Cryphalus* sp., *Cryphalus asperatus*, *Phytoptorus polygraphus*, *Phytokteines spinidens*, *Pityophthorus* sp., *Cryphalus piceae*. From these species and their galleries, 114 fungal isolates were isolated: *Leptographium* sp. Was 55.2%, *Ophiostoma piceae* and *Ophiostoma ainoae* were each 3.5%. *Graphilbum* sp. was 15%. *Trichoderma* sp. 18.4% and *Lecanicillium* sp. 4.3%. This research first recorded two species of *Ophiostoma*—*O. piceae* and *O. ainoae*, as well as representatives of *Leptographium* and *Graphilbum* genera on spruce, in galleries of bark beetles. These findings expand the known diversity of ophiostomatoid fungi in the Caucasus region and underscore the need for continued monitoring of vector-pathogen relationships in Georgian coniferous forests, especially under climate-induced stress conditions.

Emerging association between *Neofusicoccum parvum* and the invasive *Ailanthus altissima*: a first report from Italy. I. GIUBILEI, M.I. DRAIS, D. COS, A. MAZZAGLIA. Department of Agriculture and Forest Sciences, University of Tuscia, Viterbo, Italy. E-MAIL: irene.giubilei@unitus.it

Ailanthus altissima (Tree of Heaven) is among the most aggressive invasive plant species worldwide, capable of outcompeting native flora and altering ecosystem dynamics. In 2024, a widespread dieback phenomenon affecting *A. altissima* was observed in Rome province, Italy. Symptomatic trees exhibited canopy decline, wood discoloration, and necrotic lesions. Fungal isolates obtained from infected tissues were identified morphologically and molecularly as *Neofusicoccum parvum*, an aggressive pathogen within the Botryosphaeriaceae family. The identity of the fungus was confirmed through multilocus sequencing (ITS, *tef1-α*, *tub2*, and *LSU*), and its pathogenicity was validated via inoculation trials, which reproduced disease symptoms and fulfilled Koch's postulates. This represents the first report of *N. parvum* infecting *A. altissima* in Italy. The occurrence of this pathogen on an invasive host species raises intriguing questions about potential biocontrol implications. As *N. parvum* is known for its virulence on a broad range of woody plants, its natural emergence on *A. altissima* may offer novel avenues for controlling this difficult-to-eradicate species. These findings provide new insights into the pathogen's ecology and underscore the importance of monitoring emerging host-pathogen interactions, particularly in

the context of invasive species management and changing environmental conditions.

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Impact of climate change on olive dieback diseases in Tunisia and beneficial effect of *Bacillus velezensis* OEE1 in biocontrol of fungal pathogens. M.A. TRIKI, M. CHEFFI, Y. GHARBI, K. ENNOURI, S. KRID, M. ENNOURI. *Laboratory of Genetic Resources of Olive tree: Characterization, Valorization and Phytosanitary Protection, Olive Tree Institute, University of Sfax, Road of the airport km 1.5, P.O. Box 1087, Sfax, 3000, Tunisia.* E-MAIL: mohamedalitriki2@gmail.com

Olive tree (*Olea europaea* L.) is of great economic importance in Tunisia. Field surveys showed increased incidence of severe dieback and mortality of olive trees increased in many olive-growing area. These regions might be particularly affected by climate change, which could have extensive impact on the olive agrosystem including the development of diseases. Olive dieback is characterized by rapid browning of leaves, dieback of shoots, twigs and branches followed by death of the entire tree. Laboratory diagnosis and pathogenicity tests, revealed the occurrence of pathogens in branches and occasional attacks by soil-borne fungi. *Neofusicoccum australe*, *Phoma fungicola*, *Nigrospora* sp., *Biscogniauxia mediterranea* and *Lasiodiplodia theobromae* were detected in branches. These species are reported for the first time to occur in olive trees in Tunisia. To limit the use of agrochemicals against these pathogens, different biocontrol strategies have been developed using endophytic bacteria. *Bacillus velezensis* OEE1 represented an efficient and effective alternative in the control of these pathogens as it was able to reduce mycelial growth up to 60% either by diffusible or volatile compounds. The mechanisms used by this strain include competition for space and nutrients (siderophore), production of cell wall degrading enzymes (chitinase and glucanase) and several antimicrobial compounds (fungicin, utirin, surfactin, bacillomycin, macrolactin).

B. velezensis OEE1 can also activate the plant's defensive responses. Besides biocontrol ability, *B. velezensis* OEE1 is an effective plant growth promoting bacteria due to atmospheric nitrogen fixation, phosphate solubilisation and phytohormone production.

Holm Oak (*Quercus ilex*) decline in Apulia, Southern Italy, a phytopathological overview. P.G. LUCCHESI, E. CHIAROMONTE, D. SALAMONE, E. TARASCO, S. POLLASTRO, F. FARETRA, F. NIGRO. *Department of Soil, Plant and Food Sciences, (DiSSPA), University of Bari Aldo Moro, Bari, Italy.* E-MAIL: stefania.pollastro@uniba.it

Holm oak (*Quercus ilex*) is a keystone species in Mediterranean agroecosystems, valued for its ecological, ornamental, and landscape functions. Over the last decade, widespread and progressive decline has been observed in Apulia (southern Italy). This study aims to investigate the underlying causes of this phenomenon. Systematic surveys and sample collections were carried out across different Apulian locations. Fungal isolates were identified by morphological and molecular techniques; bacterial communities were characterized, and insect presence was monitored in urban settings. Multiple xylem-inhabiting fungi were identified, notably *Diplodia corticola*, a known canker pathogen of *Quercus* spp. Bacterial strains of the genera *Brenneria* and *Gibbsiella* were also isolated from symptomatic tissues. Concurrently, infestations by phytophagous insects such as *Kermes vermilio* and *Nidularia pulvinata* were frequently observed. These findings, along with meteorological data under analysis, suggest a potential link between drought stress and disease severity. Preliminary evidence indicates a complex etiology, with holm oak decline likely resulting from the interplay of biotic (fungi and insects) and abiotic (climatic) stressors. The study highlights the importance of integrated monitoring strategies to address forest tree decline in the context of global change.

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Genetic characterization and evolution dynamics of *Ophiostoma novo-ulmi* on wych elm along the Italian peninsula. A.L. PEPORI, H. BERTO, S. TORRE, S. VILLA, C. DE QUATTRO, F. SEBASTIANI, F. PECORI, A. SANTINI. *National Research Council, Institute for Sustainable Plant Protection, Via Madonna del Piano 10, 50127 Sesto Fiorentino, FI, Italy.* E-MAIL: alessia.pepori@cnr.it

Dutch elm disease (DED), caused by the fungus *Ophiostoma ulmi* and later *Ophiostoma novo-ulmi*, is one of the most aggressive plant diseases that has been devas-

tating elm populations throughout Europe for almost a century. Two subspecies of *O. novo-ulmi* are present in Europe: subsp. *novo-ulmi* (formerly Euro-Asiatic race) and subsp. *americana* (formerly North American race). Since the 1980s, hybrids between these subspecies have also been found. *Ulmus glabra* Hudson (wych elm) is present along the Italian peninsula in small, scattered populations, separated by hundreds of kilometres. The survival of these populations is at risk due to the spread of DED and for the changing climatic conditions. Wych elm is a species with average DED resistance, but which suffers considerable damage under stress conditions. A sampling of symptomatic plants was carried out throughout the Italian peninsula to perform the genetic characterisation of the *O. novo-ulmi* populations and to monitor the relative abundance of the two subspecies in selected populations of *U. glabra*. We also compared two sets of samples available at the IPSP-CNR from the pre-epidemic phase (2001) and the epidemic phase (2013). Sixty-three isolates of *O. novo-ulmi* obtained from different elm populations were selected and genetically characterised. The results of this study are providing a clear and detailed picture of how these two subspecies and their hybrids evolved in this area. Information regarding the evolution of *O. novo-ulmi* populations, the dynamics of the spread over time and space were analysed and suggestions for future scenarios were developed.

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Etiology of Pine Ghost Canker in Southern California Urban Forests. L. SHANNON¹, B. MARCELO¹, E. KARINA¹, C. CARLOS¹, A. ADAM¹, K. JOHN², S. CHRISTOPHER², E. AKIF¹. ¹University of California, Davis, United States of America; ²University of California, Agricultural and Natural Resources, University of California, United States of America. E-MAIL: sclynch@ucdavis.edu

Pine ghost canker is a recently described disease affecting multiple pine species in urban forests of Southern California. Symptoms include wedged cankers with irregular margins and cryptic discoloration on cross-sections of branches, which can lead to severe dieback and potentially tree death. In this study, we identified and characterized five *Neofusicoccum* species (*N. luteum*, *N. mediterraneum*, *N. parvum*, *N. stellenboschianum*, and

N. vitifusiforme) as the primary etiological agents of pine ghost canker. These pathogens were consistently isolated from multiple symptomatic pine samples (n = 41) and identified by morphology and phylogenetic analyses using four DNA barcodes (rDNA ITS, tef1, tub2, and rpb2). Pathogenicity was confirmed on healthy branches of 15-year-old Monterey pines, where the five *Neofusicoccum* species, caused vascular lesions that were not significantly different in length. Secondary fungi (*Diaporthe*, *Diplodia*, *Neopestalotiopsis*, and *Pestalotiopsis* spp.) were also recovered from symptomatic tissues but did not cause vascular lesions in pathogenicity tests. The optimal temperature for mycelial growth of *N. luteum* and *N. parvum* was 30°C, whereas for *N. mediterraneum*, *N. stellenboschianum* and *N. vitifusiforme*, it was 25°C. All five species were able to resume growth at room temperature (20°C) after showing no growth during a 7-day exposure to 5°C and 40°C. This study constitutes the first report of *N. luteum*, *N. stellenboschianum*, and *N. vitifusiforme* causing pine ghost canker in California. Environmental factors such as warmer temperatures, irrigation, and pest infestations are discussed as drivers of disease expression in pine trees. Management practices are also proposed.

CONCURRENT SESSION E1 – Recent and future developments in plant pathogen diagnostics

SESSION KEYNOTES

Using High Throughput Sequencing to resolve cases of mistaken identity. A.R. FOWKES^{1,2}, N. BOONHAM^{1,2}, A. FOX¹. ¹Fera Science Ltd, York Biotech Campus, York, United Kingdom, YO41 1LZ. ²Newcastle university, Newcastle upon Tyne, Tyne and Wear, United Kingdom, NE1 7TU. E-MAIL: aimee.fowkes@fera.co.uk

Traditionally, virus discovery, has been driven by viruses causing charismatic, conspicuous symptoms on hosts. Viruses were characterised by host range, serology, physical characteristics etc. Partial sequence data was only generated from the advent of PCR based methodologies. Since 2009, use of high throughput sequencing (HTS) has enabled detection and molecular characterisation of novel viruses based on sequence alone. Disconnect between pre- and post-sequencing eras has led to accidental ‘rediscovery’ of viruses, which has the potential to lead to unnecessary regulation or incorrect conclusions on host ranges and distribution. Three case studies will be presented where HTS has been used to resolve mistaken identity. (1) Resolving the “rediscovery” of

plantain virus X is an ideal example, where sequencing of original isolates of the virus allowed for taxonomic clarification, information on host range, geographic distribution and removal of pending regulation. However, this represents an ideal scenario where the virus was preserved in an accessible collection, after forty years the virus was detected in plants at the same location and the willingness of laboratories to share their data and collaborate to form a comprehensive study on this virus. (2) In Ullucus, Andean potato latent virus and potato leafroll virus have previously been reported, based on serological testing. Contemporary serology testing also suggested the presence of these regulated viruses. Yet, testing by HTS identified novel viruses closely related to these viruses, suggesting a cross reaction with the antisera. In contrast to plantain virus X, lack of original material has so far prevented further work to obtain sequence from the original isolates and do comparison of sequence. (3) In UK pea crops, HTS informed surveillance demonstrated presence, distribution and impact of turnip yellows virus in peas, a virus not previously associated with this host in the UK. As HTS is increasingly used for virus detection and discovery, but the biological relevance and context is lacking behind due it's costly and time-consuming nature, what tools can we use to determine which viruses are of most concern and therefore where we should allocate resources. Increased use of HTS in virome studies will result in new virus findings, but these give little context required for risk assessment. As we continue to use HTS, how can we gather more contextual information to better inform identified viruses?

The evolution of diagnostics for graft-transmissible diseases of citrus from a citrus germplasm quarantine program perspective. G. VIDALAKIS. *Department of Microbiology & Plant Pathology, Citrus Clonal Protection Program, University of California, Riverside, CA 92521, United States of America.* E-MAIL: vidalg@ucr.edu

Over the past century, the diagnostics for graft-transmissible diseases of citrus have evolved from visual symptom identification to advanced molecular and in silico tools. This transformation is embodied in the evolution of the Citrus Clonal Protection Program (CCPP) at the University of California, Riverside, which has served as a global model for germplasm quarantine, diagnostics, and the distribution of pathogen-tested citrus propagative materials. Early diagnostic efforts, dating back to the 1930s, relied on visual inspection of field symptoms such as psorosis bark scaling. The development of bio-

logical indexing in the 1950s marked a major advancement, enabling earlier symptom expression on a range of indicator plants under controlled environmental conditions. However, bioindexing posed inherent challenges, including the need for extensive greenhouse space, highly trained personnel, and high variability in results across different programs and geographic regions. With the introduction of ELISA and sPAGE in the 1970s and PCR-based methods in the 1990s, diagnostics became more specific and scalable. Today, high-throughput sequencing and bioinformatics platforms such as EDNA and MiFi have introduced a paradigm shift in pathogen detection, enabling rapid, sensitive, and comprehensive screening of citrus germplasm. Despite these technological advances, the integration of classical and novel diagnostic tools remains essential to confirm ambiguous results and ensure regulatory compliance. At the CCPP, the diagnostic strategy goes far beyond selecting the “best” test. It implements a multi-layered system that combines bioindexing with molecular tools and continuous innovation, including instruments engineering, qPCR assays updates and validation, synthetic internal standards, bulk sampling, LAMP field diagnostics, controlled environment agriculture, indicator optimization, and light spectrum manipulation to enhance symptom expression. An example of this evolution is the development of a high throughput samples processing system and two universal SYBR® Green RT-qPCR assays for eight citrus viroids, which replaced biological indexing as the official California Department of Food and Agriculture test under the state's mandatory Citrus Nursery Stock Pest Cleanliness Program. More recently, EDNA-MiFi probes were developed for 20 graft-transmissible citrus pathogens. These assays enable simultaneous screening of multiple pathogens without the need for individual tests, reducing costs and increasing efficiency. They were validated according to the MIQE guidelines to ensure reproducibility, sensitivity, and specificity. Assay optimization included melting temperature analysis, degenerate primer design, and compatibility with phenol-chloroform, spin column, and magnetic bead RNA extraction—methods tailored to high-throughput environments. These advancements have supported tens of thousands of viroid tests, achieved a 0% viroid infection rate in commercial nurseries by 2023, led to the discovery of citrus viroid VII, and increased the CCPP's quarantine testing and germplasm introduction capacity by 300%. Lab operations emphasize meticulous sampling, preparation, and workflow design to prevent cross-contamination. These include one-directional sample movement, clean/dirty zones, equipment swab-testing and decontamination, and use of internal synthetic stand-

ards. Sample pooling and targeted tissue selection ensure reliable results, especially for asymptomatic or low-titer infections. This presentation offers a retrospective of citrus diagnostics through the lens of the CCPP's evolution, highlighting how research, standardized protocols, infrastructure, and multidisciplinary collaboration ensure the sustainability of citrus quarantine diagnostics.

Rapid detection of mycotoxigenic *Aspergillus* spp. associated with pistachios using Loop mediated isothermal amplifications (LAMP). W. MELLIKECHE¹, A. RICELLI², M. ABUKHMAISH¹, R. CARACCILO³, M. GALLO¹, C. CASINI³, G. COLELLI⁴, F. VALENTINI¹, A.M. D'ONGHIA¹. ¹International Center for Advanced Mediterranean Agronomic Studies, Bari, Italy. ²National Research Council- Institute of Molecular Biology and Pathology (CNR-IBPM), Rome, Italy. ³Enbiotech SRL, Palermo, Italy. ⁴Department of Agriculture, Food, Natural resources & Engineering, University of Foggia, Foggia, Italy. E-MAIL: mellikeye@iamb.it

Aspergillus species create major postharvest problems due to the food losses caused by their mere presence and the hazardous mycotoxins they produce. The major mycotoxins associated with these species are aflatoxin B1 (AFB1), produced mainly by *A. flavus* and *A. parasiticus*, and ochratoxin A (OTA), produced mainly by *A. carbonarius*. In this study, we developed three rapid detection assays for the aforementioned species based on Loop-mediated isothermal amplification (LAMP). Three assays were developed targeting genes from mycotoxin production clusters, two of them are species-specific targeting genes *pks* and *aflT* for *A. carbonarius* and *A. flavus*, respectively. The third assay is generic to detect aflatoxigenic *Aspergilli* associated with pistachios including *A. flavus*, *A. parasiticus* and *A. nomius*. Result visualization was done in real-time via detection of fluorescent signals. The method developed showed high sensitivity and specificity with detection limits of 0.3 pg/reaction of DNA for the *A. carbonarius* assay and 0.03 pg/reaction for the other two assays. The assays were further implemented on inoculated nuts, including pistachios and almonds, after a one- step crude DNA extraction. These tests revealed a detection level of 0.5 spore/g that shows the effectiveness of LAMP as a rapid method for detecting potentially toxigenic *Aspergillus* spp. directly from food. The validation of the assays included tests on a larger scale that further confirmed their sensitivity and specificity, as well as enabled the production of ready-to-use LAMP kits. These kits are easy to use and aim to

simplify the screening of food samples in order to monitor the presence of specific *Aspergillus* contaminations.

Application of isothermal amplification and high throughput sequencing methods for enhanced virus detection in yam. G. SILVA¹, R. FESTUS^{1,2}, R. PREMPHEH³, M.D. QUAIN³, S.E. SEAL¹. ¹Natural Resources Institute, University of Greenwich, Central Avenue, Chatham Maritime, Kent ME4 4TB, UK; ²current address: Department of Agricultural Science and Plant Protection, Mississippi State University, Mississippi, MS 39762, USA; ³Council for Scientific and Industrial Research-Crops Research Institute, Fumesua, P.O. BOX 3785, Kumasi, Ghana. E-MAIL: g.silva@gre.ac.uk

Yam (*Dioscorea* spp.) productivity is severely compromised by viruses and the lack and high cost of clean planting material. Propagation through 'seed' yam tubers encourages the recycling of infected planting materials, increasing virus incidence and yield losses, a common issue for vegetatively propagated crops. In Ghana, yam farmers often use low-quality, virus-infected planting material from their own or neighbouring farms. The only effective control method is to use virus-free seed yam. The Council for Scientific and Industrial Research-Crops Research Institute (CSIR-CRI) developed an aeroponics and hydroponics system to enhance seed yam production. This system has a high multiplication rate, generating thousands of plantlets from a single plant. However, virus titres in the plantlets are reduced but not eliminated, complicating a reliable virus detection and making it difficult to ensure that planting material is virus-free. This could lead to false virus-negative certification of planting materials, spreading infected materials and threatening crop production and food security. We have developed and evaluated isothermal amplification- and high throughput sequencing (HTS)-based diagnostic tests for virus detection in various crops. The yam tests have been transferred to CSIR-CRI, improving their capacity in diagnostics and seed certification and contributing towards the production and sustainable supply of high-quality seed yams. We will discuss the potential wider application of these tests and the factors influencing their future deployment in support of seed systems of root and tuber crops.

Early diagnostics of *Erysiphe corylacearum* causing emerging powdery mildew on hazelnut. S. MATIĆ^{1,2}, C. D'ERRICO², M. BARONE¹, S. DAVINO¹, E. NORIS², M. MOIZIO³, G. MASOERO⁴. ¹Department of Agricul-

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Erysiphe corylacearum has recently emerged as the causal agent of a new and dangerous powdery mildew affecting hazelnut in northern Italy and other European regions. After shifting to hazelnut as a new host in Asia, the fungus rapidly spread to several European countries within a few years. Compared to the common hazelnut powdery mildew caused by *Phyllactinia guttata*, this pathogen poses a greater threat due to its substantial impact on yield and hazelnut quality. The disease's emergence in Italy revealed distinct *E. corylacearum* sub-groupings, population expansion, and high genetic similarity with recently identified isolates from other regions. Effective containment requires early and reliable detection, ideally before symptom onset. In this study, we evaluated innovative, field-deployable diagnostic methods, including isothermal amplification, NIRS / Raman spectroscopic analyses, and foliar pH measurement. These techniques, integrated with portable devices capable of cloud data storage and mobile phone-based remote monitoring, enabled rapid, non-destructive detection of the disease within 30 minutes directly in the field. Both isothermal amplification and spectroscopy-based approaches showed high accuracy in distinguishing between asymptomatic and symptomatic plants before visible symptoms appeared. Our findings suggest that these methods, through measurements of amplification curves or spectral patterns, offer a promising basis for non-destructive diagnosis of emerging hazelnut powdery mildew and potentially other plant diseases. The portability and ease of use of these systems allow their application directly by farmers, without the need for laboratory expertise, making them highly useful in supporting timely disease control.

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Unmasking the hidden microbiome of *Fagus sylvatica*: Combining PNA clamps and targeted Primers to explore seasonal variation in endophytic diversity. I. GIUBILEI¹, S. TURCO¹, L. MAHAWAR², B.R. ALBRECHTSEN², A. MAZZAGLIA¹. ¹Department of Agriculture and Forest Sciences, University of Tuscia, Vit-

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Climate change is reshaping forest ecosystems, threatening the health and stability of key species like *Fagus sylvatica*. Understanding the dynamics of plant-microbiome interactions is essential, as these microbes influence plant resilience and stress response. This study evaluated the efficiency of Peptide Nucleic Acid (PNA) clamps and selective primers in enhancing bacterial and fungal metagenomic profiling, and explored seasonal dynamics of the endophytic community in *F. sylvatica*. Amplicon sequencing was performed using Illumina technology, targeting the bacterial 16S rRNA gene (V3-V4 region) and the fungal internal transcribed spacer regions (ITS1 and ITS2). The application of PNA clamps significantly improved bacterial detection by reducing host DNA interference and increasing microbial read diversity. Among fungal targets, the ITS1 region amplified with the ITS1F/ITS2 primer set proved most efficient, offering higher taxonomic resolution and greater community richness than other approaches. Seasonal shifts were observed in both bacterial and fungal communities, with notable differences in composition, diversity, and relative abundance between early and late season samples. Certain bacterial (e.g., *Pseudomonas*, *Sphingomonas*) and fungal taxa (e.g., *Stemphylium*, *Epicoccum*) showed clear seasonal enrichment patterns. Interestingly, the increasing abundance and complexity of fungal communities later in the season may contribute to the improved performance of PNA clamps, suggesting the microbial load could influence host DNA suppression efficiency. These findings confirm the effectiveness of PNA clamps and selective primers in refining endophytic profiling and reveal seasonal shifts in the *F. sylvatica* microbiome. These insights deepen our understanding of plant-microbe interactions and support strategies for forest resilience under changing environmental conditions.

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Detection of *Curtobacterium flaccumfaciens* pv. *flaccumfaciens* in bean seeds: Challenges encountered and methodology approaches to address them. M.C. HOLEVA, P.E. GLYNOS, C.I. REPPA, I. DERVISI, C.D. KARAFILA. Benaki Phytopathological Institute, Scientific Directorate of Phytopathology, Laboratory of Bacteriology, 8 Stefanou Delta str., GR 14561, Kifissia, Greece. E-MAIL: m.holeva@bpi.gr

Seed-transmitted bacterial pathogens are of major concern in plant health, as they cause crop diseases with high economic impact, especially when they belong to regulated pathogens and phytosanitary measures are applied. Contaminated or infected seeds can be a major pathway for bacterial pathogens to enter a new area and spread in long distances. Due to the possible variability of treatment efficacy of the commercially produced seed against bacterial pathogens, it is crucial to employ sensitive, specific and robust diagnostic methodologies to detect and exclude contaminated/infected seed lots. Currently, the official seed testing methods are based on standard (validated) protocols relying primarily on the isolation of bacterial pathogens from seeds on semi-selective media. However, this approach can be hampered by factors such as: the outgrowth of the pathogen by other microbes that are present in the tested seeds, the possible viable but nonculturable cell (VBNC) status of the pathogen, the low pathogen population in the seeds and the interference of seed coating in the isolation procedure. In the present study, results of laboratory efforts to address such diagnostic difficulties and assess the presence of live pathogens in seed samples employing a grow-out assay and a PCR-based approach will be presented, focusing on the bean pathogen *Curtobacterium flaccumfaciens* pv. *flaccumfaciens*. The bean crop is of high economic importance for several Regional Units in Greece in which beans constitute a protected designation of origin (PDO) product. Exclusion of contaminated/infected bean seeds is essential to preserve local varieties and achieve increased productivity of enhanced quality.

Monitoring *Erwinia amylovora* spread in apple orchards: development of a detection protocol from corbicular pollen. B. VICELLI^{1,2}, V. GUALANDRI^{1,2}, T. FATTORE¹, C. CAINELLI¹, G. PUOPOLO^{2,3}. ¹Fondazione Edmund Mach, Centro di Trasferimento Tecnologico, Via E. Mach 1, 38098 San Michele all'Adige, Italy. ²Centro Agricoltura Alimenti Ambiente (C3A), Università degli Studi di Trento, Via E. Mach 1, 38098, San Michele all'Adige, Italy. ³Fondazione Edmund Mach, Centro di Ricerca e Innovazione, Via E. Mach 1, 38098 San Michele all'Adige, Italy. E-MAIL: bianca.vicelli@fmach.it

Fire blight, a destructive disease caused by *Erwinia amylovora* (Ea), significantly threatens global apple and pear production. Since honeybees play a key role in Ea transmission within orchards, a qPCR-based protocol has been developed for the early detection of Ea from corbicular pollen. Uninfected pollen was inoculated with

a cell suspension of strain Ea21 (rifampicin-resistant) at different concentrations (from 1×10^8 to 1×10^0 CFU mL⁻¹). Uninoculated pollen was used as an untreated control. One gram of spiked samples was serially diluted and plated on Nutrient Agar, which was amended with rifampicin for the CFU counting. Concurrently, 30 g of spiked samples were homogenised in 120 mL of 0.85% (w/v) NaCl and 0.001% (v/v) TWEEN®80 and incubated on ice under orbital shaking (140 rpm). After 1 h, samples were centrifuged to remove impurities. The supernatants were immediately collected and centrifuged. The resulting pellets were suspended in Tris-HCl (pH 8) buffer and exposed to thermal shock. Total DNA was extracted with DNeasy® mericon® Food kit and amplified in qPCR according to EPPO standard PM 7/20. Following validation, the robustness of this protocol was meticulously tested on corbicular pollen collected in two Trentino valleys. In 2022 and 2023, over 200 corbicular pollen samples from Valsugana and Val di Non were analysed in real-time, ensuring the protocol's reliability and accuracy. The protocol developed in this study has the potential to serve as a crucial predictive tool in the future. It could provide an early warning system for potential Ea infection in apple orchards, offering hope for more effective disease management strategies.

POSTER PRESENTATIONS

Chlorophyll spectroscopy and fluorescence for the racial characterization of yellow rust in wheat. M.T. GARCIA-LOPEZ¹, M.F. RUZ-RUIZ¹, J.R. PORRAS-PEREZ², J.C. SILLERO-SANCHEZ², J.A. JIMENEZ-BERNI¹. ¹AgroPhenoLab, Institute for Sustainable Agriculture-Spanish National Research Council (IAS-CSIC), Cordoba, Spain. ²Breeding and Biotechnology Area, Andalusian Institute of Agricultural Research and Training (IFAPA). Alameda del Obispo Center, Cordoba, Spain. E-MAIL: tgarcialopez@ias.csic.es

Yellow rust (Yr), a fungal disease caused by *Puccinia striiformis* f. sp. *tritici* (*Pst*), affects both durum and bread wheat. Recently, new Asian *Pst* races have caused significant losses in European crops, even in previously resistant varieties. This polycyclic disease progresses rapidly when conditions are favourable, as occurred in spring 2024. Our study characterized different Yr races in 52 near-isogenic lines (NILs) of the susceptible 'Avocet' cultivar, each containing a single Yr resistance gene. We also evaluated bread wheat varieties (*Triticum aestivum* L.) with known resistance genes. Plants were visually assessed four times during spring using the Cobb

severity scale. Simultaneously, we measured flag leaf photosynthetic activity (4 repetitions/NIL) with a PhotosynQ fluorometer and micro-plot spectral reflectance (3 repetitions/NIL) using an SVC HR-1024i spectroradiometer. We observed significant differences in disease severity among NILs: some showed susceptibility, others displayed hypersensitive responses with cellular necrosis, and some remained resistant. Several genotypes with medium severity allowed us to correlate symptom evolution with spectral and photosynthetic parameter changes, demonstrating a direct relationship with visual observations. Developing phenomic tools for early detection of new races of this virulent pathogen is crucial for implementing fast screening, providing objective assessments applicable to large-scale studies and resistance gene identification research in cereals. Moreover, these methodologies could also be applicable in the context of precision agriculture where early detection and disease mapping can enable mitigation measures.

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Development of an isothermal recombinase polymerase amplification assay for the field deployable detection of Begomoviruses. B. DAVENPORT¹, M. AMATO², N. WHITE¹, A. HARNESS¹, H. FALZON¹, K. RAU¹. ¹Research and Development, Agdia, Inc. Elkhart, IN 46514, United States of America. ²Managing Director, Agdia-EMEA, Soisy-sur-Seine 91450, France. E-MAIL: marcos.amato@agdia-emea.com

The Begomovirus genus is the largest of all plant viruses comprising over 450 recognized species infecting several economically important fiber, root, and vegetable dicotyledonous crops. Begomoviruses are persistently transmitted by whiteflies, distributed worldwide with higher prevalence in tropical and subtropical regions, and consist of single stranded circular DNA genomes with both monopartite and bipartite organization. High rates of genomic recombination and population introduction through global trade can lead to issues with established diagnostic methods missing new isolates, variants, or even species. To circumvent both novel speciation detection and rigorous lab-based testing, a novel recombinase polymerase amplification (RPA) assay was developed and validated for the detection of Begomoviruses. The developed RPA reaction utilizes a crude plant extract,

runs at a single operating temperature for 20 minutes, and generates real-time amplification data. The assay has successfully identified 56 isolates from 28 diverse species and is predicted to be able to detect over 350 species based on insilico analysis. The RPA was screened against 42 potential host species to assess performance. The validated test provides a novel tool in screening suspect plants for several Begomoviruses in a single field deployable reaction.

Development and validation of a field-based colorimetric lamp assay for the detection of *Clavibacter michiganensis* in tomato plants. G. MERMIGKA^{1,2}, M. MEGARITI³, M.G. PAGOULATOU¹, E. GIZELI^{3,4}, D.E. GOUMAS^{1,2}. ¹Hellenic Mediterranean University (HMU), School of Agricultural Sciences, Department of Agriculture, Laboratory of Biotechnological Applications and Phytopathology, GR71004, Heraklion, Greece. ²Hellenic Mediterranean University (HMU), University Research Centre, Institute of Agri-Food and Life Sciences, GR71410, Heraklion, Greece. ³Institute of Molecular Biology and Biotechnology, Foundation for Research and Technology-Hellas, Heraklion, Greece. ⁴Department of Biology, University of Crete, Heraklion, Greece. E-MAIL: dgoumas@hmu.gr

Point-of-care (POC) diagnostics are revolutionizing the detection of plant pathogens by enabling rapid, on-site identification without the need for specialized laboratories. One of the tools used for this purpose is Loop-mediated isothermal amplification (LAMP). LAMP is a powerful molecular technique increasingly used in pathogen control for its rapid, sensitive, and specific detection of plant pathogens. Unlike traditional PCR, LAMP operates at a constant temperature, eliminating the need for expensive thermocycling equipment, which makes it ideal for field-based applications. In this study, we have developed and evaluated a portable colorimetric LAMP assay for the detection of *Clavibacter michiganensis* (Cm) in tomato plants. Initially, we designed a set of primers for micA gene that shows high specificity for this subspecies and tested them against genomic DNA (gDNA) isolated from Cm and *C. sepedonicus*. Next, we evaluated a) the detection limit of Cm in plant extracts spiked with bacteria, b) the specificity in lysates spiked with Cm and other bacteria frequently occurring in tomato plants under field conditions, c) the effect of plant lysates from different tomato varieties in Cm detection and d) the detection of Cm in artificially infected tomato plants. The minimum concentration that we could consistently detect Cm was 10⁵ CFU while we were able to detect Cm

as soon as four days post infection in the stems of the infected plants. Altogether, from our findings we can conclude that the developed method for Cm detection can be used as a reliable tool for the early detection of the pathogen.

The study was part of the project "Innovations in Plant Protection for sustainable and environmentally friendly pest control, InnoPP"- TAEDR-0535675 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.

Advanced diagnostic strategies for the detection and identification of seed-borne viruses at ICARDA to enhance seed health security. S.G. KUMARI, A. MOUKAHEL. *Seed Health/Virology Laboratory, International Center for Agricultural Research in the Dry Areas (ICARDA), Terbol Station, Beq'a Valley, Zahle, Lebanon.* E-MAIL: s.kumari@cgiar.org

ICARDA's Seed Health Laboratory (ICARDA-SHL) is responsible for the monitoring, clearance, and documentation of the safe germplasm movement at the Center. The laboratory processes over 50,000 samples annually for seed-borne pathogens/pests, for the purpose of short- and long-term conservation and global distribution. This rigorous screening process is essential for preventing the spread of seed-borne pests across borders, especially viruses that can be transmitted further by insect vectors upon introduction. This has made the management of seed-borne viruses more challenging, as infected seeds can introduce the virus into new areas and lead to infections in subsequent growing seasons. Early detection and accurate diagnosis of viral diseases are critical for implementing effective control measures and strategies. In this context, ICARDA-SHL is actively engaged in the development, validation and dissemination of advanced diagnostic tools and protocols for germplasm health testing, facilitating technology transfer to NPPOs. Accordingly, ICARDA-SHL has developed and applied various molecular tests for the detection and identification of seed-borne legume viruses, including IC-RT-PCR, uniplex RT-PCR and multiplex RT-PCR in a single direct test. These advanced diagnostic techniques allow rapid analysis of large numbers of samples at a relatively low cost, high throughput, short execution times and enhanced accuracy, thus ensuring the availability of certified seeds, which are key strategies to mitigate the spread of seed-borne legume viruses that cannot be controlled by conventional plant treatments. In this presentation, validated molecular tools for the detection and identification of

selected legume viruses (e.g. AMV, CMV, BYMV, PSb-MV) applied by ICARDA-SHL will be described.

Molecular diagnostics for greenhouse populations of *Alternaria* spp. (Ascomycota, Pleosporaceae) on tomatoes. L. DUČKENA^{1,2}, N. BESSADAT^{2,3}, N. BATAILLÉ-SIMONEAU², F. BASTIDE², B. HAMON², M. KOPPEL⁴, K. LOIT⁴, N. RASIUKAVICIUTE⁵, G. BIMSTEINE¹, P. SIMONEAU². ¹*Institute of Soil and Plant Sciences, Latvia University of Life Sciences and Technologies, Liela Street 2, LV- 3001, Jelgava, Latvia.* ²*University of Angers, Institut Agro, INRAE, UMR 1345 IRHS, SFR 4207 QUASAV, Beaucouzé Cedex, 49070, France.* ³*Laboratory of Applied Microbiology, University of Oran1 Ahmed Ben Bella, BP 1524, El M'Naouer, 31000, Oran, Algeria.* ⁴*Institute of Agricultural and Environmental Sciences, Estonian University of Life Sciences, Fr.R. Kreutzwaldi 1, 51006, Tartu, Estonia.* ⁵*Institute of Horticulture, Lithuanian Research Centre for Agriculture and Forestry, Kauno Street 30, LT-54333, Babtai, Lithuania.* E-MAIL: Lilija.Duckena@lbtu.lv

Diseases caused by fungal pathogens, including *Alternaria* spp., are one of the major limiting factors in the production of tomato (*Solanum lycopersicum* L.) due to devastating yield losses. Fungi from the genus *Alternaria* are characterized by overlapping of the morphological characters among several species; therefore, DNA-sequence based identification is required for accurate species identification that is critical for the development of disease management strategies. In our study, a total of 896 plant samples with typical disease symptoms caused by *Alternaria* spp. were collected from greenhouses in Latvia, Lithuania, and Estonia between 2022 and 2024. Recovered *Alternaria* spp. strains were further identified at the species level using a multigene phylogenetic framework. Initial identification of fungal strains based on the internal transcribed spacer regions 1 and 2 and intervening 5.8S nrDNA (ITS) confirmed the occurrence of four sections from the genus *Alternaria*, namely *Alternaria*, *Porri*, *Infectoriae*, and *Ulocladioides*. Small-spored *Alternaria* strains from the section *Alternaria* were further analysed based on RNA polymerase II second largest subunit (*rpb2*), putative F-box-domain- containing protein (*ASA-10*), and putative histone-like transcription factor (*ASA-19*), and *Alternaria arborescens* and *A. alternata* species complexes along with *A. postmessa* were identified on tomato. Further analysis of large-spored *Alternaria* spp. (section *Porri*) based on a multi-locus phylogeny of ITS, *rpb2*, glyceraldehyde-3-phosphate dehydrogenase (*gapdh*), translation elongation factor 1- α

(*tef1*), and *Alternaria* major allergen gene (*alt a 1*) confirmed the occurrence of five *Alternaria* species on tomato in the Baltic region, including novel pathogens that represent two previously undescribed taxa of the genus *Alternaria*.

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A multi-method approach for detection of Tomato brown rugose fruit virus. E. DEL ÁNGEL DE LA CRUZ ARÉVALO¹, J. GARITA-CAMBRONERO², P. OLIVARES PACHECO³, A. DE FRANCESCO³, A.P. FERNÁNDEZ-GETINO GARCÍA¹. ¹*Dirección Técnica de Evaluación de Variedades y Laboratorios, Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria (INIA), Consejo Superior de Investigaciones Científicas (CSIC), Madrid, Spain.* ²*Instituto Tecnológico Agrario de Castilla y León (ITACyL), Subdirección de Investigación y Tecnología Área de Innovación y Optimización de Procesos Laboratorio de Biología Molecular y Secuenciación, Valladolid, Spain.* ³*Asociación Nacional de Obtentores Vegetales (ANOVE), I+D en Protección Vegetal, Madrid, Spain.* E-MAIL: agustina.defrancesco@anove.es

Tomato brown rugose fruit virus (ToBRFV) is an emerging plant virus posing a significant threat to tomato and pepper production worldwide. To enhance early and accurate detection, we developed and evaluated an optimized diagnostic workflow combining multiple methods: LAMP, RT-qPCR, and bioassay, tailored for use across different stages of diagnosis, from greenhouse pre-screening to precise molecular confirmation and direct detection in host plants. The work was carried out with ToBRFV naturally infected seeds. The sensitivity of the bioassay was compared with RT-qPCR, contributing to the bioassay method validation. A LAMP protocol, adapted from a published seed detection method, showed improved performance under our lab conditions starting from seed extracts instead of isolated RNA. A standard curve generated from serial dilutions of crude seed extracts and purified RNA showed consistent RNA detection up to RT-qPCR Ct values of ~26, while LAMP detected infections up to the 10⁻⁵ dilution of crude extracts (Ct ≈ 30.85), surpassing purified RNA in sensitivity. The LAMP method will be further assessed for its viability in greenhouse pre-screening in terms of speed, cost, and diagnostic accuracy. Comparing sensitivities, both bioassay and LAMP exhibited comparable limits of detection, detecting up to one infected seed in a 400

seed lot. The RT-qPCR is the most sensitive method, detecting one ToBRFV-infected seed in a total of 1000 seed lot. These findings underscored the potential of bioassays in discerning virus infectivity, although further validation and standardization are necessary. A final diagnostic series will be proposed for efficient ToBRFV detection.

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Development of a portable real-time PCR and digital droplet PCR assay for *Cercospora beticola* detection on beet. M. CRUDELE, C. LAGUARDIA, P.R. ROTONDO, S. LAERA, T. MASCIA, R.M. DE MICCOLIS ANGELINI, F. FARETRA. *Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, Bari, Italy.* E-MAIL: ritamilvia.demiccolisangelini@uniba.it

Cercospora beticola, the causal agent of Cercospora leaf spot, is one of the most destructive pathogens affecting leaf beet (*Beta vulgaris*). The pathogen spreads rapidly under warm and humid conditions and frequently coexists in mixed infections with *Phoma betae*. Disease management is challenging, particularly due to the emergence and spread of fungicide resistance in the pathogen populations. Rapid and accurate pathogen detection is essential for the development of effective and sustainable strategies of disease control. This study aimed at developing new sensitive and specific TaqMan probe-based assays for rapid detection and quantification of *C. beticola* using portable real-time qPCR systems for in-field diagnostic tests, and digital droplet PCR (ddPCR) for lab tests. The specificity of both qPCR and ddPCR was confirmed against a panel of fungal and bacterial species commonly associated with beet crops. Sensitivity was calculated as the mean of three independent experiments. The limit of detection for the ddPCR assay was 10 pg of DNA, while qPCR was 100-fold more sensitive, quantifying up to 0.1 pg of fungal DNA. To enable in-field detection, different protocols for rapid DNA extraction from fresh leaves, crop residues, and seeds using commercial kits (Biomeme Sample Homogenization and M1 Sample Prep Cartridge kit for DNA; Sigma-Aldrich REDExtract-N-Amp Plant PCR Kit; and BN QuickPick Plant DNA) were tested and compared in qPCR assays. The developed methods for early detection and quanti-

fication of *C. beticola* provide valuable tools for enhancing epidemiological studies that can contribute to design appropriate strategies for the disease management.

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Investigating microbiome shifts in tomato plants under pathogen pressure. E. FRANCOMANO¹, M.M. ACI¹, M. CASUSCELLI¹, N. MOHAMED¹, S. MOSCA¹, L. SCHENA¹, A. MALACRINO^{1,2}. ¹Department of Agriculture, Università degli Studi Mediterranea di Reggio Calabria, Località Feo di Vito, 89124 Reggio Calabria, Italy. ²Department of Biological Sciences, Clemson University, Clemson, SC, USA. E-MAIL: edda.francomano@unirc.it

The plant microbiome plays a critical role in maintaining plant health and productivity, influencing plant tolerance to both biotic and abiotic stressors. Previous research has shown that the plant microbiome composition often changes when plants are exposed to stressors. However, it is still unclear whether this response is stressor-specific and can be used to predict the occurrence of a specific stressor. Here, we set up a microcosm system using tomato as model plant to test the response of the leaf bacterial microbiome to the presence of different belowground pathogens (*Pseudomonas syringae* pv. *tomato*, *Fusarium oxysporum* f. sp. *lycopersici*, and *Alternaria alternata*). Through amplicon sequencing, machine learning, and null-model analysis, we found that different belowground pathogens can indeed leave a signature on the leaf bacterial microbiota, either on its composition or on the ecological relationships within the microbiome. This combined approach is helping us to better understand plant-microbiome interactions under stress, to develop new tools to reliably predict the occurrence of biotic stressors in the field, and to deploy the next generation of microbiome-based plant health management strategies.

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Early and sensitive detection of *Ceratocystis ficiicola* in *Ficus carica*: a new Taqman real-time PCR approach. C. CARBOTTI¹, M. CARLUCCI¹, R. SPANÒ², P.G. LUCCHESI¹, A. PACIFICO¹, F. NIGRO¹. ¹Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, via Amendola 165/A, 70126 Bari, Italy. ²National Research Council, Institute for Sustainable Plant Protection, Via Amendola, 122/D 70126 Bari, Italy. E-MAIL: franco.nigro@uniba.it

Ceratocystis ficiicola is a soil-borne pathogen that infects the roots, the trunk and main stem of susceptible hosts, causing vascular wilt and tree death. It was first identified as the causal agent of *Ceratocystis* canker in fig (*Ficus carica*) in Japan and subsequently reported in Greece and Italy. Diagnostic procedures for *C. ficiicola* are not yet fully standardized. In this study, specific primers and a TaqMan real-time PCR assay were designed and implemented to develop a rapid, sensitive molecular diagnostic tool for *C. ficiicola* detection. Prior to assay validation, the identity of the fungal isolates was confirmed by evaluating key morphological traits, including the size and shape of ascomata and conidia, and by amplifying the internal transcribed spacer (ITS) region using primers ITS5 and ITS4. Assay validation was conducted on a panel of fungal isolates collected from healthy and diseased *F. carica* plants. Additional tests were performed on symptomatic, asymptomatic, and artificially inoculated plant and soil samples, demonstrating the assay's ability to reliably detect the pathogen. The assay exhibited high sensitivity, detecting as little as 0.2 pg μL^{-1} of *Ceratocystis* DNA both in water and in plant DNA extracts. These findings indicate that the molecular method developed, utilizing specific primers and a TaqMan probe, offers a reliable, sensitive, and time-efficient tool for *C. ficiicola* detection. Implementation of this assay will facilitate improved monitoring of the pathogen's distribution, particularly in the Apulian region, and support timely management strategies to mitigate its spread.

The DIFENDO project: Advanced diagnostics for plant health in agriculture. A. TAGLIENTI, S. BERTIN, F. COSTANTINI, N. PUCCI, P.F. ROVERSI. *National*

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The control of plant pests is becoming increasingly challenging due to rising threats posed by food demand and security, global trade, and climate change. Key factors for such control include (i) accurate and rapid detection tests, which are crucial for the appropriate application of phytosanitary measures, and (ii) implementation of natural compounds as eco-friendly biopesticide alternatives to agrochemicals, which are currently subjected to limitations in the European Union (EU Sustainable Use of Pesticides Directive 2009/128/EC). The “DIFENDO project – *Advanced diagnostics for plant health in agriculture*”, coordinated by National Reference Institute for Plant Protection (CREA-DC) and financed by the Italian Ministry of Agriculture (MASAF) aims at achieving advancements in both fields. Tests will be developed for the application of innovative tools to be used on-site (i.e., fields, entry points), based on advanced biosensors and molecular diagnostics. The proposed technologies include Lab-on-Chip systems for pest detection via real-time amplification, nanopore-based high-throughput sequencing, e-nose, and microfluidic based sensing devices. All these tools will be connected to a platform for data transmission and storage, allowing for a rapid and comprehensive setting up of both preventive measures and outbreak response. The project also aims at defining testing protocols and platforms for the evaluation of natural compounds for pest control within the framework of Integrated Pest Management. The effectiveness of natural compounds (i.e., plant biomass extracts, essential oils, hydrosols) will be tested through the quantification of the target pest in the experimental plants, using advanced diagnostic systems such as digital PCR and microfluidic networks.

Phylogenetic relationships of *Alternaria* species infecting apple trees in northeastern Spain. M.VILA DE VARGAS, M. PLA, A. NADAL, I. LLORENTE, C. MORAGREGA. *Institute of Agri-food Technology (INTEA), Carrer Maria Aurèlia Capmany, 61. University of Girona, 17003 Girona, Spain.* E-MAIL: marc.viladevargas@udg.edu

In recent years, an emerging apple tree disease characterized by lesions on leaves and fruits has been reported. This disease is of increasing importance in both Australia and various regions of Europe (Italy, France, and

Spain). The disease symptoms were first detected in apple orchards in the Girona region (Catalonia, north-eastern Spain) in 2013. Within two years, the disease had spread to up to 20% of ‘Golden’ and ‘Gala’ apple orchards, and since then, its impact on production losses has persisted. This disease has been attributed to a species complex within *Alternaria* section *Alternaria*, though the specific species involved in Catalonia outbreak remain unknown. Due to its recent emergence and the limited information available regarding its etiology and epidemiology, there are currently few detection and management methods, posing a potential threat to the major apple-producing regions of Europe. To develop a qPCR-based tool for detecting pathogenic *Alternaria* species affecting apple trees, an analysis of the species diversity associated with the disease in Catalonia was conducted. This analysis was based on four newly developed molecular markers that allow for the distinction of phylogenetic lineages within *Alternaria* sect. *Alternaria*. Sixty *Alternaria* isolates from the INTEA-UdG collection, obtained from different symptomatic apple cultivars from various orchards in Girona, were selected for analysis. Isolates included a range of morphotypes and varying levels of pathogenicity/virulence. This study evaluated the suitability of the molecular markers for identifying the *Alternaria* species present in apple orchards and will provide a basis for the development of pathogen detection techniques.

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Study of some Calabrian olive cultivars for the selection of genotypes resistant to *Verticillium dahliae*. A. MUTO, V. VIZZARRI, M. HUSSAIN, A. IULIANO, A. DE MARCO, I. DE ROSE, F. POLIZZO, E. PERRI, E. SANTILLI. *CREA-Research Centre for Olive, Fruit and Citrus Crops, Via Settimio Severo, 83 87036 Rende (CS), Italy.* E-MAIL: elena.santilli@crea.gov.it

Verticillium dahliae, is one of the most devastating soil-borne fungi of the olive tree (*Olea europaea* L.) worldwide. The olive sector plays a crucial role in Calabria’s agribusiness; in fact, it is Italy’s second-largest olive-growing region, just behind Puglia. However, no information regarding resistant or sensitive Calabrian cultivars affected by *V. dahliae* is available. Yet, our understanding of the defence mechanisms that operate at the root level to explain tolerance to this disease is incom-

plete. This project seeks to explore the molecular networks that contribute to the root defence mechanisms in Calabrian olive cultivars, both tolerant and susceptible, when faced with *Verticillium*. For this study, six Calabrian olive cultivars (two years old), along with non-Calabrian cultivars serving as controls, were selected for inoculation with a strain of *V. dahliae* classified as defoliating (D) at specific time intervals of 0, 2, 7, and 15 days under controlled experimental conditions (three for each time point and conditions). The disease progression of inoculated and control (non-inoculated) plants from each cultivar was first evaluated by scoring the severity and incidence of disease. Next, we quantified *V. dahliae* DNA using real-time PCR to correlate the phenological data obtained. Finally, the expression over time of nine defence-related genes and ten genes associated with the phenylpropanoid pathway was also examined under the conditions reported above. The results obtained showed links between tolerance's level to this pathogen and specific root defence mechanisms at the phenological and genetic levels.

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Loop-mediated isothermal amplification for seed and surface detection of ToBRFV. R. CARACCILO¹, E. TROIANO^{1,2,3}, S. LO CASTRO¹, G. ARCOLEO¹, G. PARRELLA². ¹Enbiotech srl, Via Masuccio Salernitano 28, 84012 Angri, SA, Italy. ²Institute for Sustainable Plant Protection of the National Research Council (IPSP-CNR), P.le Enrico Fermi 1, 80055 Portici, NA, Italy. ³University of Naples "Federico II", Department of Biology, Via Cinthia 26 - 80126 Naples, Italy. E-MAIL: r.caracciolo@enbiotech.eu

Loop-mediated isothermal amplification (LAMP) is a molecular diagnostic method that enables rapid and specific detection of plant pathogens. Tomato brown rugose fruit virus (ToBRFV) is a virus that significantly impacts tomato and pepper production worldwide. The focus of this work was to evaluate the ToBRFV detection by Reverse Transcription-LAMP (RT-LAMP) in tomato and pepper seeds, and on environmental surfaces such as benches, plexiglass, glass, cardboard, and metal. Seeds were artificially contaminated at 1% and 0.1% of infection levels using a homogenate obtained from ToBRFV naturally infected leaves, previously checked for virus infection. Different surfaces were directly contaminated

using homogenates obtained from 1 % of infected seeds. RNA extraction was performed following a rapid protocol based on seeds grindings and surface swab smearing, followed by RT-LAMP analysis conducted on ICGENE system. An RT-qPCR validate protocol was used to compare the efficiency and sensitivity of the proposed RT-LAMP method, using in parallel the total RNA extracted and purified from the same samples. ToBRFV was detected on all contaminated seeds and surfaces with both molecular methods, showing the same efficiency and sensitivity for both methods compared. The results of this study demonstrate the efficiency and sensitivity of LAMP technology in detecting ToBRFV in different matrices, offering a practical and rapid solution for tomato and pepper seed health screening and environmental monitoring. The user-friendly format of the diagnostic tool facilitates rapid on-site testing, thereby enabling timely interventions that minimise the risk of spreading ToBRFV in vulnerable agricultural systems.

Development of Real-time LAMP assay for the rapid detection of *Colletotrichum acutatum* in olive. F.Z. SENNOUN¹, W. MELLIKECHE¹, R. CARACCILO², A.M. D'ONGHIA¹, F. VALENTINI¹, M. GALLO¹. ¹International Centre for Advanced Mediterranean Agronomic Studies, Via Ceglie, 9-70010, Valenzano (BA), Italy. ²Enbiotech SRL, via Del Bersagliere, 45-90143, via Masuccio Salernitano, 28-84012, Angri (SA), Italy. E-MAIL: senounfatmazohra@gmail.com

Olive anthracnose (OA) caused by *Colletotrichum* spp., is a significant disease that affects olive cultivation worldwide, resulting in considerable yield losses and a deterioration in olive oil organoleptic qualities. Effective management of OA requires an integrated approach with a strong focus on the accurate and precise detection of the causal agent, using specific and sensitive diagnostic tools. Among these, loop-mediated isothermal amplification (LAMP) has emerged as one of the simplest, fastest, and most accurate methods. In this study, a characterization of *Colletotrichum* spp., isolated from samples collected in Maruggio area in southern Apulia (Italy), was conducted. Six isolates were obtained from symptomatic olive drupes and identified morphologically and molecularly as *Colletotrichum acutatum sensu stricto* (s.s.). A LAMP assay was subsequently developed for the direct detection of this causal agent. The primers were designed to target the conserved β -tubulin 2 (TUB2) gene, and result visualization was carried out in real time through the detection of fluorescent signals. The assay exhibited high specificity and sensitivity with

no cross-reaction observed and a detection limit of 1 ng DNA/mL. To further validate the assay for direct pathogen detection, it was applied to inoculated olive drupes following a one-step crude DNA extraction protocol. This method successfully detected *C. acutatum* s.s. at concentrations as low as 5 spores per gram, confirming LAMP as an effective, rapid, and sensitive tool for direct pathogen detection, even in complex matrices like olive.

Beyond *Colletotrichum* identification: species diversity, host association patterns and geographical distribution.

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Colletotrichum spp. include important plant pathogens affecting several plant hosts worldwide. Recent re-evaluations in taxonomy, and possible incorrect genomic sequences available in public databases, have led to uncertainties in *Colletotrichum* species identification, raising concerns about the reliability of both past and new records. The CLARITY project, funded by EFSA, aims to resolve these ambiguities by improving species identification, clarifying their real geographical distribution and host associations, and contributing to effective management strategies. To achieve this, *Colletotrichum* isolates from European and global culture collections are being reassessed using molecular and morphological approaches to validate taxonomic identities. Additionally, extensive field surveys across the Mediterranean basin are investigating *Colletotrichum* diversity in various hosts, including tropical and subtropical fruit crops, apples, strawberries, ornamentals and native plants. In parallel, a large-scale analysis of genetic metadata is underway, integrating type specimens, past records and newly obtained isolates. Nearly 80 *Colletotrichum* isolates have been retrieved from European culture col-

lections and over 600 were collected from surveys conducted in Greece, Italy, Portugal and Spain. Robust phylogenetic analyses based on multi-locus sequence typing (MLST) are ongoing to provide accurate species identification, with preliminary data showing a predominance of *C. acutatum* (18.5%), *C. boninense* (26.1%) and *C. gloeosporioides* (51.3%) species complexes. Whole genome sequencing (WGS) will be applied to selected isolates for improved taxonomic resolution and functional genomic insights. The combined results will enhance our knowledge on taxonomy, evolution, geographical distribution and host association patterns of *Colletotrichum* spp., contributing to the development of a comprehensive database for accurate species identification.

This research was financially supported by CLARITY, a project granted from the European Food Safety Authority (EFSA): "GP/EFSA/PLANTS/2023/06 - Improving the knowledge on the European distribution of plant pathogenic species of the genus Colletotrichum, recently subject to taxonomical changes".

CONCURRENT SESSION E2 – Integrated pest management: from plant resistance to chemical control

SESSION KEYNOTES

A key to sustainable management of the grapevine downy mildew pathogen. F. DELMOTTE. INRAE, Bordeaux Sciences Agro, SAVE, ISVV, F-33140, Villenave d'Ornon, France. E-MAIL: francois.delmotte@inrae.fr

Europe is the historical cradle of viticulture, but cultivated grapevine has been increasingly threatened by pathogens of American origin. The invasive oomycete *Plasmopara viticola* causes downy mildew, one of the most devastating grapevine diseases worldwide. Current strategies for controlling *P. viticola* primarily relies on fungicide applications. Genetic analyses revealed that all invasive *P. viticola* populations stem from a single North American lineage associated with *V. aestivalis*. After its introduction into Europe, this lineage spread globally through secondary introductions, underscoring the importance of stricter regulation of international plant material movement to prevent further introductions of downy mildew. As an alternative to fungicide-based control, we are exploring two strategies: deploying disease-resistant grapevine varieties and disrupting the pathogen's sexual reproduction cycle. Our efforts have focused on understanding the dynamics of primary inoculum resulting from sexual reproduction, which play a critical role in initiating disease outbreaks. For the first time in

oomycetes, the locus that determines mating types was described. We then applied ddPCR to describe oospore distribution in vineyard soils. We found higher accumulation of oospores below vines than between rows, consistent with the distribution of grapevine leaf litter which accumulates at the base of the vines in autumn. Finally, we found that removing grapevine leaves containing the pathogen's oospores can significantly delay disease onset in the following season. Taken together, these results confirm that management strategies targeting the disruption of the pathogen's sexual cycle could significantly enhance the control of downy mildew epidemics. The adoption of disease-resistant varieties (DRV) represents another promising innovation to significantly reduce fungicide use. However, while DRV are planted, *P. viticola* populations are demonstrating rapid adaptability, successfully overcoming the grapevine resistances. Promoting sustainable resistance management therefore requires a deep understanding of the genetic interactions between the plant and the pathogen. We explore pathogen adaptation and provided the first description of candidate effectors of *P. viticola* involved in the interaction with 3 resistance factors of grapevine. We discovered that strains overcoming Rpv3 and Rpv12 shared deletions that removed several effector genes. In contrast, the breakdown of Rpv10 appears to be driven by a dominant suppressor locus, likely introduced through a secondary introduction event of the pathogen into Europe. These findings illustrate the evolutionary diversity of virulence strategies used by *P. viticola*, from gene deletions to admixture-driven adaptation. The discovery of these avirulence loci has opened the door to the design of new molecular tools to monitor the evolution of grapevine downy mildew populations adapting to partial resistance of grapevine.

Diversity-Driven strategies to enhance wheat resistance to major fungal diseases in Tunisia. S. BEN M'BAREK^{1,2,3}, M. LARIBI^{2,4}, W. ABDEDAYEM^{2,5}, H. KOUKI², S. ARFAOUI⁶, J. NASRI¹, S. HAMZA⁵, P. KARISTO⁷, A. MIKABERIDZE⁸, M. PATPOUR⁹, M.S. HOVMØLLER⁹, M. RAHMATOV¹⁰, S.E. STRELKOV⁴, C.P. SANSALONI¹¹, S. DREISIGACKER¹¹, P.K. SINGH¹¹, C. SAINT PIERRE¹¹, K. AMMAR¹¹, A. YAHYA-OUI¹². ¹Regional Field Crops Research Center of Beja, Beja, Tunisia. ²IRESA-Wheat Septoria Precision Phenotyping Platform, Tunis, Tunisia. ³Laboratory of 'Appui à la Durabilité des Systèmes de Production Agricole Dans la Région du Nord-Ouest', Higher School of Agriculture of Kef (ESAK). ⁴Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB,

Canada. ⁵Department of agronomy and plant biotechnology, National Agronomic Institute of Tunisia, Tunis, Tunisia. ⁶National Institute of Field Crops, Tunisia. ⁷Plant Health, Natural Resources Institute Finland (Luke), Jokioinen, Finland. ⁸School of Agriculture, Policy and Development, University of Reading, Reading, UK. ⁹Department of Agroecology, Aarhus University, Slagelse, Denmark. ¹⁰Department of Plant Breeding, Swedish University of Agricultural Sciences, P.O. Box 190, SE-23422 Lomma, Sweden. ¹¹International Maize and Wheat Improvement Center (CIMMYT) km, 45 Carretera México-Veracruz El Batán, Texcoco CP56237, Mexico. ¹²Borlaug Training Foundation, Colorado State University, Fort Collins, CO, United States. E-MAIL: sarrah.bm2712@gmail.com

Tunisia, a major durum wheat producer in the Mediterranean, faces recurring threats from foliar fungal diseases such as *Zymoseptoria tritici* (causal agent of Septoria tritici blotch, STB), *Pyrenophora tritici-repentis* (tan spot), and the re-emergent *Puccinia graminis* f. sp. *tritici* (wheat stem rust), although sporadic, is a major concern. In the context of limited resistant cultivars and high reliance on fungicides, a shift toward integrated pest management (IPM) is essential. We present a multi-layered, resistance-driven strategy combining high-throughput field phenotyping, seedling screening under controlled conditions, genotyping, genome-wide association studies (GWAS), and ecological field practices to identify and deploy durable resistance. Since 2015, a Septoria precision phenotyping platform—established by CIMMYT in collaboration with Tunisian research institutions—has screened up to 20,000 wheat accessions from CIMMYT annually and collaborators at two disease hotspot locations under natural and artificial inoculations. This platform accelerates the identification of resistant germplasm to feed breeding pipelines. In parallel, a USDA Mediterranean collection was phenotyped for STB and tan spot, genotyped with the 90K SNP array, and analyzed via GWAS, identifying marker-trait associations (MTAs) and highlighting eight Tunisian landraces as promising donors for tan spot resistance. For stem rust, race phenotyping revealed the dominance of the Sicily race in Tunisia, and seedling screens against Ug99 and related races showed resistance in only 9.4% of Mediterranean accessions, indicating the need for more widespread resistance introgression. To explore sustainable disease management strategies and reduce fungicide inputs, field experiments mixing the susceptible durum wheat variety Karim with 25% of resistant cultivars (Salim or Monastir) significantly reduced STB severity and enhanced both grain yield and thousand-kernel weight, underscoring the potential of varietal mixtures

as an effective approach to disease control. This multi-layered strategy provides a robust IPM framework that reduces dependence on chemical control, harnesses valuable genetic resources, and supports the development of resilient wheat cultivars adapted to Tunisia's evolving disease landscape. During my talk, I will present these key findings and their implications for breeding and disease management.

ORAL PRESENTATIONS

Resistance to fungicides in apple powdery mildew and eco-friendly management alternatives. L. GUR^{1,2}, A. MESHOOLAM^{1,3}, E. KURZBAUM^{1,3}, Y. COHEN⁴, Y. KINRIECH¹, Y. YANOVSKY¹, S. OVADIA⁵, M. REUVENI¹. ¹Shamir Research Institute, University of Haifa, Katzrin 1290000, Israel. ²Institute of Evolution (IOE), University of Haifa, Haifa 3103301, Israel. ³Tel-Hai College, Kiryat Shmona 1220800, Israel. ⁴Bar-Ilan University, Ramat Gan 5290002, Israel. ⁵S.H.F, Agricultural consulting and development services, Karmeit Yosef 9979700. E-MAIL: liorgur@gri.org.il

Apple powdery mildew, caused by *Podosphaera leucotricha*, is a major disease of apple worldwide. It infects leaves, flowers, and fruits, leading to significant yield losses. Disease management relies primarily on fungicides such as demethylation inhibitors (DMIs) and respiration inhibitors (SDHIs, QoIs), which increase resistance risks. Although reduced fungicide efficacy has been documented, no genetic mutations for resistance have been detected in *P. leucotricha*. Israel's climate, with rainy springs and dry summers, favors disease development and necessitates frequent fungicide applications. In this study, we assessed *P. leucotricha* resistance to fungicides in field trials, laboratory assays, and genetic analyses. In orchard trials conducted in the Mediterranean climate of northern Israel, penconazole (DMI) and kresoxim-methyl (QoI) performed poorly (20–30% efficacy) compared to trifloxystrobin (QoI). In leaf disc bioassays, EC90 values were 30 and 75 ppm ai for penconazole and kresoxim-methyl, respectively, compared to 2.1 ppm for trifloxystrobin. No resistance-related mutations were detected, suggesting alternative resistance mechanisms. Preventive foliar sprays in the greenhouse and orchard with macro- and micronutrients reduced disease incidence by 94–97% and severity by 77–83%. Hot air (180°C) applied with the aid of a tractor-towed heat blower reduced disease incidence and severity by 50–70%. Combining reduced fungicide applications with heat treatment resulted in 60–90% and 85–90% reduction

in disease incidence and severity, respectively, suggesting similar or better efficacy compared to conventional treatments. This study provides the first characterization of *P. leucotricha* resistance in Israel and highlights eco-friendly strategies for reducing fungicide reliance.

This research was supported by the Ministry of Agriculture and Food Security, and the Plants Production & Marketing Board, Israel.

Integrated control of *Coniella granati*: fungicide efficacy and the role of cultivar resistance. P. MARINO, A. CARLUCCI, M.L. RAIMONDO, F. LOPS. *Department of Agricultural Sciences, Food, Natural Resources and Engineering (DAFNE), Via Napoli 25, 71121, Foggia, FG, Italy.* E-MAIL: francesco.lops@unifg.it

Pomegranate (*Punica granatum* L.) is gaining increasing importance in Italy, particularly in southern regions such as Apulia, Basilicata, and Sicily, due to favorable climatic conditions. This study aimed to evaluate the effectiveness of different chemical strategies against *Coniella granati*, the causal agent of pomegranate stem and crown canker in the Mediterranean area. The experimental work was divided into two phases, with an initial in vitro screening followed by field trials. The in vitro tests assessed the antifungal activity of 13 commercial formulations applied at three different concentrations. Only a few active ingredients showed effective inhibition of pathogen growth, including Tebuconazole and Dodine. These fungicides were subsequently selected for field applications, with symptom assessments carried out at regular intervals on three commonly cultivated varieties—Jolly Red, Ako, and Wonderful—at two commercial orchards in Apulia. Tebuconazole proved to be the most effective treatment in reducing disease severity, as indicated by the lowest AUDPC values, whereas Dodine showed less consistent results. Additionally, controlled inoculations revealed significant differences in varietal susceptibility: Wonderful exhibited the lowest level of infection, while Jolly Red appeared to be the most sensitive. This study highlights the value of an integrated management approach, combining the targeted selection of the most effective chemical active ingredients with varietal resistance screening as key tools for the sustainable control of *C. granati*, in alignment with the principles of the European Green Deal.

Integrated management of grapevine downy mildew: from disease forecasting to optimized fungicide resist-

ance management. B. LECCHI¹, G. MADDALENA¹, P. BORSA², M. COATTI², L. BORGHI³, S.L. TOFFOLATTI¹. ¹Università degli Studi di Milano, Dipartimento di Scienze Agrarie e Forestali, DiSAA, Via Celoria 2, 20133, Milan, Italy. ²Syngenta Italia, Viale Fulvio Testi 280/6, 20126 Milan, Italy. ³Syngenta Crop Protection AG, Schaffhauserstrasse 101, 4332 Stein, Switzerland. E-MAIL: beatrice.lecchi@unimi.it

Grapevine downy mildew poses one of the main challenges in modern viticulture, emphasizing the need for sustainable and integrated management strategies. While precision agriculture offers valuable tools for optimizing fungicide use and improving disease control efficacy, it often overlooks the critical importance of fungicide selection based on resistance monitoring. This study integrated three key components of disease management: (i) validation of the EPICure forecasting model to optimize treatment timing; (ii) assessment of anti-resistance strategies within two different fungicide application programs, aimed at ensuring effective crop protection and limiting the spread of resistant strains; and (iii) development of monitoring methods to quantify resistant individuals using cell sorting techniques. Field trials were conducted during the 2023 and 2024 seasons in two commercial vineyards located in Northern Italy (Veneto and Friuli-Venezia Giulia). Untreated control plots and plots treated according to anti-resistance strategies were established. Throughout the seasons, disease severity was assessed, and leaf samples were collected for fungicide resistance monitoring through bioassays and molecular analyses. Results demonstrated the forecasting model's reliability in identifying periods of high infection risk and confirmed the effectiveness of anti-resistance strategies, even in the presence of strains exhibiting multiple resistance to different classes of single-site fungicides. Moreover, cell sorting showed promising potential for resistance monitoring, although further methodological optimization is required to enhance its efficiency.

This research is carried out within the framework of an industrial PhD program co-funded by the Ministry of University and Research and by Syngenta Italia.

Beyond the berries: unlocking the biopesticide potential of strawberry tree leaves. F.J. CEBALLOS-BURGOS^{1,2}, J. MARTINS^{1,3}, J. CANHOTO^{1,2}, C. MALEITA^{1,2}. ¹University of Coimbra, Centre for Functional Ecology - Science for People & the Planet (CFE), Department of Life Sciences, Calçada Martim de Freitas, 3000-

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Alongside fruit production, plants synthesize a diverse array of chemical compounds enabling them to adapt to and resist environmental threats. Domestication and selection for consumer-favoured traits (e.g., fruit size, flavour, yield) have often inadvertently compromised the natural defences of cultivated plants against pests, requiring active intervention by farmers. Wild or less domesticated plants, with their retained resistance mechanisms, offer a potential solution. The strawberry tree (*Arbutus unedo*), resilient to challenging environments like marginal lands and drought, is valued for its nutraceutical fruits and its production of phenolic compounds, including arbutin and hydroquinone. While its pharmacological benefits for human health and its antimicrobial properties are known, its efficacy against agricultural biotic stressors has not been extensively explored. Therefore, this research investigated the *in vitro* efficacy of *A. unedo* leaf extracts and/or arbutin/hydroquinone against the root-knot nematode *Meloidogyne incognita* (among the global plant parasitic nematodes with the greatest economic impact), the oomycete pathogen *Phytophthora cinnamomi* (causal agent of crown and root wilting in over 5000 plant species), and the fungal pathogens *Zymoseptoria tritici* (causing septoria tritici blotch in wheat) and *Venturia inaequalis* (causal agent of apple scab), some of the most serious agricultural threats. *M. incognita* mortality and hatching inhibition assays were performed, and growth reduction was measured in solid medium for the fungal and oomycete pathogens. The findings reveal that aqueous and methanolic leaf extracts and arbutin and/or hydroquinone significantly induce nematode mortality and inhibit hatching, as well as inhibit the growth of the tested fungal and oomycete pathogens.

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The prolonged effect of biochar application during the nursery stage on plant diseases. O. FRENKEL¹, T. SAMUCHA¹, G. MORDUKHOVICH¹, A.K. JAISWAL¹,

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Biochar is widely recognized as a soil amendment with potential to suppress plant pathogens. However, its effectiveness in disease suppression is highly dependent on factors such as feedstock type and, particularly, application rate-beneficial effects that are often observed within a narrow dosage range. Given the difficulty of removing biochar once applied to soil, its targeted use in nursery systems and soilless media presents a more controlled and sustainable approach to managing plant diseases. This study aimed to evaluate the duration of induced systemic resistance triggered by nursery-stage biochar application and its potential to suppress both foliar fungal pathogens and soilborne pathogens. Eucalyptus-derived biochar at a 3% concentration was incorporated into the root plugs of commercial tomato trays, which were then transplanted into biochar-free greenhouse soil. The induced resistance effect persisted for over 60 days post-transplanting, resulting in more than a 50% reduction in *Botrytis cinerea* damage of leaves. Comparable suppression was observed in cucumber leaves infected with *Podosphaera xanthii*, the causal agent of powdery mildew, in a field trial. In contrast, suppression of the soilborne pathogen *Pythium aphanidermatum* was effective only after a pre-activation period of over 14 days, during which biochar interacted with the growth substrate and reshaped the microbial community within it. These findings highlight the strategic potential of biochar use in nursery production systems to extend disease resistance beyond early growth stages, offering a precise and sustainable tool for integrated plant disease management.

Development of novel ddPCR assays for detection and quantification of SDHI resistance in *Botrytis cinerea*. G. SOFIANOS¹, K. MAVRIDIS², N. KRASAGAKIS³, E. MARKAKIS³, J. VONTAS², G. KARAOGLANIDIS¹.

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Botrytis cinerea is a high-risk plant pathogen for resistance development and, indeed, fungal populations resistant to fungicides have often been observed in the fields, throughout the world. Since resistance development reduces the control efficacy of botryticides, it is vital that quick and efficient resistance monitoring programs be developed and implemented in the control strategies. Unfortunately, typical approaches of fungicide resistance detection take time and often lack the ability of sensitive and accurate quantification. Digital droplet PCR (ddPCR) could constitute the ideal diagnostic tool, since it can be used both to identify and quantify alterations in genomic sequences, even in large pooled samples, while it may provide high specificity and sensitivity. In this study, we tested the applicability of ddPCR in analyzing the most common, and functionally validated, SDHIs resistance mutations in *B. cinerea*, namely H272R, H272Y N230I, P225F and I274V. Method validation was performed using control samples possessing the mutations at known concentrations, and DNA extracted from pooled conidia samples with various mutations' ratio, mimicking environmental sample collections. Consequently, a new ddPCR detection tool was established that could effectively detect, even at very low frequencies (reaching a detection limit of 0.5%), widespread SDHI-resistance conferring mutations in *B. cinerea* environmental samples.

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POSTER PRESENTATIONS

From field to gene: insights into zoxamide resistance management in *Plasmopara viticola*. M. PERACCHI, B. LECCHI, G. PIGNI, G. MADDALENA, S.L. TOFFOLATTI. Università degli Studi di Milano, Dipartimento di Scienze Agrarie e Ambientali – DISAA, Via Celoria 2, 20133 Milan, Italy. E-MAIL: mattia.peracchi@unimi.it

Fungicides are essential tools in the management of grapevine downy mildew, a disease caused by the oomycete *Plasmopara viticola*, a biotrophic and polycyclic pathogen with a high risk of developing resistance. Zoxamide, a fungicide with a low to moderate risk of selecting resistant strains, acts by inhibiting tubulin polymerization, with resistant strains characterized by a missense mutation at amino acid position 239 of the β -tubulin

gene. This six-year study (2017–2022) aimed to investigate the emergence and distribution of zoxamide-resistant *P. viticola* strains under different field conditions and fungicide use histories. Sensitivity was assessed through biological assays on the oospores and complemented by molecular screening for resistance-associated mutations. A total of 126 samples were collected from 57 vineyards, mostly located in North-Eastern Italy. The majority (90%) showed full sensitivity to zoxamide ($EC_{50} < 0.2 \text{ mg L}^{-1}$; EC_{95} and $MIC < 10 \text{ mg L}^{-1}$). Thirteen samples exhibited reduced sensitivity ($MIC > 100 \text{ mg L}^{-1}$), but only two showed a consistent presence (24–33%) of resistant oospores at 100 mg L^{-1} . Resistance was mainly observed in vineyards where zoxamide had been applied four or more times per season. Multiple polymorphisms at codon 239 of the β -tubulin gene were identified through sequencing, including the known C239S and C239G mutations and a potentially novel C239T variant. These findings highlight the role of selection pressure in shaping resistance in *P. viticola* populations and emphasize the need to limit the number of zoxamide applications per season. The observed genetic variability also calls for further research into the fitness and epidemiology of resistant strains to inform sustainable resistance management strategies.

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Elucidating the key determinants affecting Italian *Pyricularia oryzae* genetic diversity. S. BOSCO¹, F. SILLO², P. RUFFA¹, D. TENNI³, P. ABBRUSCATO⁴, D. TORELLO MARINONI¹, M. ROMANI³, M. MEZZALAMA^{1,5}, V. GUARNACCIA^{1,5}, D. SPADARO^{1,5}.

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Rice blast, caused by *Pyricularia oryzae* Cavara, is the most damaging disease in Italy. The use of resistant cultivars represents a sustainable method to minimize yield

loss, however high genomic plasticity of the pathogen promotes rapid adaptation to host resistance. Understanding *P. oryzae* population structure is fundamental to assess its genetic variability and to devise durable management practices. While the first population study in Italy dates to 2005, updated knowledge of local *P. oryzae* diversity is lacking. In this study, we genotyped the population structure of 242 isolates collected from rice panicles in five Italian regions (Piedmont, Lombardy, Veneto, Emilia-Romagna, and Sardinia) in 2011–2012 and 2020–2022 using SSR markers. The potential occurrence of sexual reproduction was determined by linkage disequilibrium and mating type assessment, displaying the exclusive presence of the MAT1-1 idiomorph, thus confirming the clonal structure of the Italian *P. oryzae* population. Cluster analysis of 200 distinct multilocus genotypes (MLGs) via DAPC, UPGMA, and STRUCTURE allowed to identify five genetic clusters. Molecular variance and genetic divergence analyses revealed that geographic origin and sampling year significantly affected population structure. Correlation analysis in isolates from Piedmont highlighted combined effects of climatic factors and geographic origin on local allelic diversity. Virulence testing of cluster-representative strains suggested the disappearance of a less virulent cluster over time. These findings provide novel insights into the temporal and spatial dynamics of the Italian *P. oryzae* population in Italy.

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The potassium phosphite-based fungicide CANON® controls basil downy mildew under greenhouse conditions in Israel. N. NITZAN, H. GOREN. *The Valley of Springs Research & Extension Center, Israel*, M. P. Beit She'an. E-MAIL: nitzan_nadav@yahoo.com

Sweet basil downy mildew (BDM), caused by *Peronospora belbahrii*, is the most destructive disease of commercially produced sweet basil in Israel, and unless properly managed, may lead to 100% yield reduction. Four experiments were conducted over three growing seasons (fall 2021, fall 2022 and spring 2023) under commercial-like conditions to determine the efficacy of the phosphonic acid-based fungicide Canon® following a conservative, seven-day protective spraying protocol.

While Canon did not eradicate the pathogen, treated plants had 41 to 99.9% less ($P < 0.05$) disease incidence and / or severity than the untreated control plants. The disease scores on plants treated with Canon were similar ($P > 0.05$) to an alternation of Revus^{*} / Cabrio^{*} in fall 2021; less ($P < 0.05$) than Revus in fall 2022 and higher ($P < 0.05$) than an alternation of Acrobat^{*} / Cabrio at the end of the spring 2023 experiment. The inhibitory effect of Canon, its short pre-harvest application interval and low cost compared to other fungicides make it an excellent addition and a potential replacement for controlling BDM under greenhouse conditions that should be further explored.

GrapeBreed4IPM: Developing sustainable solutions for viticulture through multi-actor innovation targeting breeding for integrated pest management. S.E. LEGLER¹, V. MANSTRETTA¹, K. AVIA², M. DRESSLER³, L. HAUSMANN⁴, G. DI GASPERO⁵, B. LAURENT⁶, M. DE LA FUENTE⁷. ¹Horta srl, Piacenza, IT 29122, Italy. ²INRAE, Université de Strasbourg, UMR SVQV, 68000 Colmar, France. ³University of Ludwigshafen, Winecampus, D-67059 Ludwigshafen, Germany. ⁴Julius Kuhn Institute (JKI), Institute for Grapevine Breeding Geilweilerhof, 76833 Siebeldingen, Germany. ⁵Istituto di Genomica Applicata, 33100, Udine, Italy. ⁶IFV, UMT SEVEN, Blanquefort, France. ⁷Spanish Wine Technology Platform – CEIGRAM - Universidad Politécnica de Madrid, Madrid, Spain. E-MAIL: s.legler@horta-srl.com

Viticulture is an economically and culturally important in the EU, highly dependent on the use of Plant Protection Products, in order to protect plants from diseases. The sector faces significant environmental pressures, aiming for a transformation towards sustainability and ecosystem preservation in line with the goals set by the European Union. Advances in grapevine breeding gave origin to Disease Resistant Varieties (DRV), also known as PIWI, which open the possibility to drastically reduce the use of fungicides in vine management. However, the reduced need for fungicides cannot be easily implemented and requires holistic pest control strategies and varieties adapted to future climate conditions are also needed. To address these issues, the GrapeBreed4IPM project focus on developing grapevine varieties with enhanced resistance to fungal diseases, adapted to local environmental and pedoclimatic conditions, aiming to provide tools to reduce the reliance on the application of fungicides. Methods like for example genomic selection are used to improve and accelerate the breeding process. In addition, the project is tailoring existing digital tools and

developing guidelines and best practices for Integrated Pest Management (IPM) specifically for the management of DRV, thus equipping farmers and advisors with the tools needed to optimize pest control while minimizing chemical usage. Within the project, Horta customized its Decision Support System for sustainable management of grapevine specifically for PIWI varieties by calibrating the plant growth model and the downy and powdery mildew models and validating them in a three-year vineyard trial in the Colli Piacentini viticultural area.

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Foundation Plant Services: member of US national clean plant network. M. AL RWAHNIH. *Department of Plant Pathology, Foundation Plant Services, University of California, Davis, USA.* E-MAIL: malrwahnih@ucdavis.edu

Located in Davis, California, USA, Foundation Plant Services is one of thirty-three US clean plant centers that make up the US National Clean Plant Network (NCPN). The NCPN can be used as a model of successful implementation of a coordinated clean plant strategy that supports producers and improves food security. The seven specialty crops within NCPN are berries, citrus, fruit trees, grapes, hops, roses, and sweet potatoes. These crops are clonally propagated, which assures identical genetic horticultural characteristics in propagated progeny, but can easily spread pathogens including viruses, viroids, phytoplasma, and bacteria if the mother plant is infected. These pathogens can have detrimental effects on performance and yield of infected plants, and the only cure is complete removal of the plant. It is important to limit pathogen spread and therefore reduce economic impact due to crop loss by screening for pathogens in plant stocks before using them for propagation. NCPN centers provide pathogen diagnostic and elimination services and maintain and distribute virus-tested plant material. In total, NCPN centers distribute on average over 37 million propagative units each year, allowing producers around the US to plant fields, orchards, and vineyards with clean material. Estimates of economic impact for grape and fruit tree centers supplying virus-tested plant material show a return on public investment dollars (\$) ranging from 10:1 to 150:1.

Evaluation of *Pisum sativum* resistance to *Didymella pinodes* after gamma irradiation in ex- planta bioassays. E. SARRI¹, A. KATSILEROS¹, S. MIGARDOU², P. VILIOTIS¹, I. SIDIROPOULOS¹, D. SIFNAIOS¹, P. DIAMANTIS¹, N. SKLAVOUNOS², P. BEBELI¹, D. KIZIS², E. TANI¹. ¹Laboratory of Plant Breeding and Biometry, Department of Crop Science, Agricultural University of Athens, Athens, Greece. ²Laboratory of Mycology, Scientific Directorate of Phytopathology, Benaki Phytopathological Institute (BPI), Athens, Greece. E-MAIL: d.kizis@bpi.gr, etani@aua.gr

Ascochyta blight caused by one or more pathogenic fungi including *Didymella pinodes*, *Ascochyta pisi*, and *Phoma pinodella*, is a devastating field pea disease, characterized by leaf, stem, and pod lesions. Plant breeding focuses on developing new crop varieties that fulfill nutritional requirements for humans and livestock while adapting to environmental challenges. A key tool in modern genetic improvement is random mutagenesis via gamma- irradiation, a non-GMO method that generates genetic variability and accelerates the identification of genotypes with desirable traits. This study examined the resistance of the M2 generation of forage pea variety 'Dodoni' (*Pisum sativum* L. var. *arvense* 'Dodoni') to *Didymella pinodes* CBS 251.47 strain. The M2 plants originated from M0 seeds treated with 100 Gy gamma radiation and grown in the greenhouse during 2023-2024. Resistance to the fungus was evaluated through a detached-leaf bioassay using mycelium agar slabs for artificial infection, in Petri dishes, under controlled conditions. Leaf photos were captured on the 3rd and 5th days post-infection. Image analysis was conducted to assess the extent of leaf lesions and calculate the Diseased Area and Disease Severity Index. After extensive screening in both greenhouse-grown (16 families x 24 plants) and field conditions (100 families x 24 plants), three M2 families showed statistically significant differences in resistance compared to non- irradiated controls. Additionally, within these families, plants exhibited statistically significant differences in yield, with increased pod number and higher fresh and dry weight. Further evaluation of selected genotypes will enable the development of varieties towards enhanced productivity and pathogen resistance.

Different susceptibility of local Apulian and Calabrian grapevines to downy mildew and characterization of *Plasmopara viticola* populations. R. CORONELLI¹, D. GERIN¹, F. SPATARO¹, S. TOFFOLATTI², M. PERACCHI², B. LECCHI², R.M. DE MICCOLIS ANGELINI¹,

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Downy mildew, a severe grapevine disease worldwide, is caused by different cryptic species of *Plasmopara viticola*. Several approaches are under study to reduce the impact of chemicals in the disease management, including the selection of resistant genotypes for grapevine breeding. In this study, the susceptibility of six different local wine grape cultivars was assessed on leaves and bunches, in 2023, a year particularly favourable to the downy mildew epidemic. Based on McKinney's index (MKI: 37.3 to 52.6%), Marchione, Moscato reale, Maresco and Moscato di Terracina were more susceptible than Moscato di Trani, Trebbiano and Verdeca, classified as resistant ones (MKI: never exceeding 16.6%). Additionally, the genetic variability of *P. viticola* populations infecting grapevine in Apulia, Calabria and Lombardia was evaluated. Sporangia were collected from oil-spots in 26 vineyards (15 Apulia, 11 Calabria), during 2023 and 2024 years. The CAPS analysis was carried on restriction enzyme *AseI* profiles of the amplified DNA using the ITS1-O/ITS2, according to Rouxell *et al.* (2013). A total of 130 isolates and 20 DNA samples from oil spots sampled in Lombardia were analysed. All belong to the clade *aestivalis*, as also confirmed by BLASTn analysis of the ITS partial sequence of few representative isolate of Apulia and Calabria regions. These preliminary results suggest that the resistant behaviour of analysed grapevine genotypes is limited to *P. viticola* clade *aestivalis* and highlight the opportunity to preserve the local biodiversity for possible introduction of new *P. viticola* clades, currently absent.

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Evaluation of an organic biostimulant on ZYMV infection and behaviour of its aphid vector in *Cucurbita pepo* L. C.L. CORRADO^{1,3}, T. TUNGADI², L. DONATI¹, T. BRUCE², A. TAGLIENTI¹, S. BERTIN¹.

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Biostimulants are substances that enhance plant growth and nutrient uptake by improving plant or rhizosphere characteristics (EU Regulation 1009/2019). This study assessed the effect of a commercial biostimulant containing alfalfa, brown algae, and molasses extracts on the interaction among zucchini (*Cucurbita pepo*), zucchini yellow mosaic virus (ZYMV), and one of its aphid vectors *Myzus persicae*. Biostimulant-treated plants showed enhanced vegetative growth (e.g., leaf number, biomass) in healthy conditions and improved reproductive traits (e.g., flowers, fruits) in infected conditions. Repeated biostimulant applications reduced ZYMV titre over time and slowed symptoms progression. The pathogenesis-related gene 1 (PR1), associated with systemic acquired resistance, and the peroxidase gene (POD), involved in the oxidative stress pathway, was up-regulated in biostimulant-treated ZYMV-infected plants, suggesting a role of biostimulants in priming the plant defence mechanisms. Aphid choice tests using detached leaves showed fewer *M. persicae* settling on treated plants, regardless of infection status. When caged on potted plants, aphids exhibited lower survival and fecundity on treated plants. Volatile organic compounds (VOCs) were collected from the plants, and GC-MS analysis revealed differences in volatiles blend composition between treated and untreated plants. The same volatile samples were used in olfactometer assays, whose results indicated that aphids preferred untreated plants, suggesting a possible repellent effect due to biostimulant-induced VOC production. Overall, the obtained results suggest that biostimulant application may reduce ZYMV impact by enhancing plant defence and limiting vector performance, offering a promising tool for IPM strategies for cucurbit crops.

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Ecological approaches to grapevine protection: preliminary considerations on the potential of stilbenes.

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The grapevine has developed various adaptative mechanisms in response to environmental fluctuations and growth-threatening factors, with phytopathogenic infections being among the most significant. Their occurrence and severity are primarily influenced by climatic conditions and the plant's natural resistance. In this context, secondary metabolism serves as a key mechanism to mitigate or delay damage. Secondary metabolites synthesized in response to phytopathogenic infections are classified as phytoalexins. During interactions with plant pathogens, phytoalexins exhibit inhibitory effects, and their synthesis is triggered by the plant-microorganism interaction. This suggests that their presence is directly linked to resistance. Among these compounds, resveratrol, a stilbenoid organic compound, plays a crucial role in the grapevine's immune system. Its presence has been detected in most grape varieties at varying concentrations, with each cultivar differing in its ability to synthesize and accumulate this compound. The initial resveratrol concentrations are typically low, but synthesis increases significantly upon pathogen infection. Whether the infection is halted at an early stage or will continue to spread depends on subsequent weather conditions, physical barriers such as the berry skin thickness, leaf surface, etc., along with the synthesized amount of resveratrol. This study investigates the potential fungitoxic effect of resveratrol on *Botrytis cinerea* spores in laboratory conditions. The aim is to determine whether and to what extent the resveratrol application affects spore viability and to what extent the plant's natural defense mechanism can be efficient.

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Food safety assessment of strobilurin and carboxamide fungicides in some fruits and vegetables.

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The cultivation of fruits and vegetables requires intensive protection against harmful organisms. Crops, such as strawberries, raspberries, lettuce and spinach are particularly vulnerable to diseases, primarily caused by various fungal infections. Due to persistent presence of pathogenic fungi throughout the growing season, frequent applications of plant protection products are often necessary. At the same time, food safety has become an increasingly critical global concern, with pesticide residues posing a major potential risk to human health. This risk is further exacerbated by the short growing cycles of these crops and the timing of pesticide applications, which often coincide with the ripening stage. Consequently, assessing the potential health risks of pesticide residues is essential, particularly for fresh produce consumed directly. In this study, we evaluated the health risks associated with residues of pesticides belonging to the chemical groups of strobilurins and carboxamide in strawberries, raspberries, lettuce and spinach. Field experiments were conducted following EPPO standards, with a plant protection product (PPP) containing boscalid and pyraclostrobin applied at recommended rates. Sampling was performed according to pre-harvest intervals. Fungicide residues were analyzed using a validated QuEChERS-based method followed with HPLC. The Hazard Quotient (HQ) was calculated using the Estimated Daily Intake (EDI) and the relevant Acceptable Daily Intake (ADI) value. In order to evaluate risk assessment, as the worst-case scenario, the maximum residue levels of pesticides obtained one hour after the application, were used. The results suggest that the consumption of these fruits and vegetables does not pose a significant health risk to humans.

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Differential molecular responses of pear and apple cultivars in response to *Erwinia amylovora* causal agent of fire blight disease. Y. GHARBI, E. BOUAZIZI, M.A. TRIKI. *Laboratory of Olive Genetic Resources: Characterization, Valorization and Phytosanitary Protection, Institute of the Olive Tree, University of Sfax, B.P 1087, Sfax, Tunisia.* E-MAIL: yaakoub.gharbi@yahoo.com

A collection of pear and apple cultivars adapted to Tunisia's growing conditions were evaluated for resistance to fire blight disease (FBD) caused by *E. amylovora*. In

this context, the molecular response of local and foreign pears and apple cultivars to FBD was investigated using an artificial infection model and monitoring of defense-related genes using Q-RT-PCR. Based on the evaluation of 8 cultivars, 6 were classified as susceptible (Meski, Anna, Williams, Bouguedma, Alexandrine, Meski Ahrech), and 2 as resistant (Lurka and Ambri) to FBD. The expression levels of genes coding for chalcone synthase (CHS), phenylalanine ammonia-lyase (PAL), and coronatine-insensitive protein (COI) were higher in the resistant cultivars, indicating that early induction of the phenylpropanoid pathway plays a crucial role in resistance against *E. amylovora* and FBD symptom mitigation. The resistant cultivar exhibited early upregulation of catalase (CAT) alongside enhanced superoxide dismutase (SOD) activity, triggering a hypersensitive response (HR) at infection sites that effectively limited pathogen spread. In addition, simultaneous early up-regulation of chitinase and β -1,3-glucanase genes correlated with reduced host susceptibility and FBD symptom severity in the resistant cultivars Lurka and Ambri. These findings suggest that the resistant cultivars Lurka and Ambri represent valuable genetic material for replanting programs or potential use as rootstocks for susceptible varieties. Future studies should evaluate grafted pear and apple hybrids of these resistant cultivars to assess the heritability of this resistance trait.

Improvement of olive anthracnose management in prefecture of Chania, Crete. A. PAPAGEORGIOU¹, A. KARAGIANNI², C. STEPPAS³, M.M. MATHIOUDAKIS², D.I. TSITSIGIANNIS¹, A.L. LAGOPODI³. ¹*Department of Crop Science, Laboratory of Plant Pathology, Iera Odos 75, Agricultural University of Athens, Athens, 11855, Greece.* ²*Institute of Olive Tree, Subtropical Crops & Viticulture (ELGO-DIMITRA), Plant Pathology Laboratory, Karamanlis Ave. 167, Chania, Crete, 73134, Greece.* ³*Aristotle University of Thessaloniki, School of Agriculture, Plant Pathology Laboratory, P.O. Box 269, Thessaloniki, 54124, Greece.* E-MAIL: lagopodi@agro.auth.gr

In previous years, temperatures rise combined with high relative humidity, led to serious outbreaks of the olive anthracnose disease in Greece. In the frame of the GleoliveTreat project, an integrated olive anthracnose management strategy was implemented in Chania prefecture of Crete. It included an integrated management model adapted to the local conditions indicated by meteorological data, the pathogen's population profile in the area of action, and specific phenological olive stages in pilot groves (conventional, organic). Sprays were applied from

the beginning of blooming and during autumn, using selected pesticides that were evaluated in previous studies. The weather conditions during 2023–2024 did not favor the appearance of olive anthracnose, but in combination with entomological infestations, the development of rots from other fungi was not successfully prevented with most predominant species in genera of *Fusarium* and *Alternaria*. The causative species of anthracnose, endemic to the region is *Colletotrichum acutatum*. However, this pathogen participated in the complex of olive fruit rots by only 3% of the total isolates tested. The sprays did not significantly reduce the rates of rotting, which is possibly due to the use of formulations that targeted *Colletotrichum* species, while other pathogens prevailed during the application period. The implementation of an integrated pest management system is difficult to be applied due to the natural topography of the area making it imperative to investigate additional means of treatment. Precision agriculture approaches seem to be the most appropriate tools to manage anthracnose and other olive fruit rots in mountainous areas' olive groves.

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UCB-1 Pistachio: a resilient choice for orchard rootstock or landscape planting. M. AL RWAHNIH, R. ABOU KUBAA. *Department of Plant Pathology, Foundation Plant Services, University of California, Davis, USA.* E-MAIL: malrwahnih@ucdavis.edu

UCB-1 pistachio rootstock is a hybrid from the controlled cross of the *Pistacia atlantica* by *Pistacia integerrima*, selected at University of California, Berkeley, USA. Seeds have been produced in California since the 1960s, with current UC production at Foundation Plant Services in Davis, California. The UCB-1 hybrid is used as a rootstock for *Pistacia vera* for commercial nut production. Individually, *P. atlantica* and *P. integerrima* were investigated as pistachio rootstocks and compared to the UCB-1 seedlings. Each of these *Pistacia* species exhibit useful traits for pistachio production: *P. integerrima* is tolerant of *Verticillium*, and is the more vigorous of the two, while *P. atlantica* is more tolerant of cold and salinity. The UCB-1 hybrid exhibits a combination of these beneficial traits, having tolerance to *Verticillium*, salinity, cold, and drought. The highest yields of 'Kerman' *P. vera* scion were from trees on UCB-1 rootstock. For decades, UCB-1 has been the most used rootstock in

California pistachio orchards because of these attributes. Its resilience factors have also led to the use of ungrafted UCB-1 as an ornamental tree in the US, where it is planted in yards and along roadsides in arid environments. When allowed to grow as a landscape tree instead of grafting UCB-1 is deciduous, with a beautiful red hue to new growth in the spring, and often dazzling red, orange, and yellow colours in the fall.

Application of phage display for the selection of antibodies for the identification of defence-related proteins in plants. L. ZAGORCHEV, I. ALBANOVA, D. TEOFANOVA. *Department of Biochemistry, Faculty of Biology, Sofia University "St. Kliment Ohridski", 8 Dragan Tsankov blvd., 1164, Sofia, Bulgaria.* E-MAIL: lzagorchev@biofac.uni-sofia.bg

Proteins play a pivotal role in plant disease resistance, serving key functions in pathogen recognition, intracellular signalling, and defense activation. Resistant genotypes typically exhibit distinct protein expression profiles compared to susceptible varieties, particularly during or following pathogen challenge. These differential expression patterns enable resistant plants to more effectively detect, respond to, and combat pathogenic threats. Therefore, the identification and characterization of specific resistance-associated proteins is crucial in understanding the molecular basis of disease resistance and screening diverse genotypes for resistance. Antibodies represent a powerful tool for studying protein expression and localization. While phage display technology offers a valuable alternative to traditional immunization methods, it still requires highly purified antigen preparations. In the present study, a novel approach was developed for rapidly selecting antibodies against putative disease-related proteins using phage display libraries. Our strategy employed alternating rounds of positive and negative selection to select antibodies targeting antigens present in the treated samples (e.g., infected plants), while excluding those present in the control. To validate this methodology, we utilized a *Solanum lycopersicum* cultivar, resistant to *Cuscuta campestris*, a holoparasitic plant with significant economic impact. Following four selection rounds, we successfully selected a polyclonal phage-conjugated antibody suspension, specific to a protein antigen, exclusively present only in infected plants.

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Sustainable pest and weed management strategies for enhancing olive production and quality. ABDEL KADER EL HAJJ. *Lebaa station, Lebanese Agricultural Research Institute*. E-MAIL: ak.hajj@lari.gov.lb

The olive oil industry has a long-standing history and remains a crucial agricultural sector, particularly in Mediterranean regions. However, olive production faces significant challenges due to pest infestations and weed competition, which can negatively impact yield and quality. This review highlights challenges from pests and weeds affecting yield and quality and explores sustainable pest and weed management strategies aimed at enhancing olive production while maintaining environmental and economic sustainability. Integrated pest and weed management approaches and smart farming technologies are discussed. Emphasis on minimizing chemical herbicides and pesticides reliance through the use biological, cultural, the sterile insect techniques, and biopesticides and inert materials. Future research should optimize strategies for diverse olive-growing regions. By integrating modern agricultural techniques with traditional practices, olive growers can achieve higher productivity while ensuring sustainable land use.

Screening of Tunisian cucurbit accessions for resistance to virus and fungal diseases. M. ELBEKKAY, H. HAMZA, C. DESBIEZ, N. DJEBALI. *Institut of Arid Region, Medenine, Tunisia*. E-MAIL: mokhtra123ira@gmail.com

The evaluation of resistance of local melon, watermelon and squash cultivars (cv.) to seven viruses and two fungal pathogens *Fusarium oxysporum* f. sp. *melonis* and *Podosphaera xanthii* was made. The results showed that 26 out of 44 cultivars evaluated, exhibited resistance for at least for one of the studied pathogens. The local cucurbit cultivars were generally resistant to MNSV and ToLCNDV in comparison to commercial cultivars. Specifically, melon cultivars M5 and M18, and squash cv. C46, C60 and C65 were resistant to WMV. Watermelon cultivars P15 and P24 along with squash cultivar C60 exhibited resistance to CMV. Partial resistance to MWMV was observed in melon cultivars M35 and M129, watermelon cultivars P35 and P55, and squash cultivar C65. Only C65 squash cv. was resistant to PRSV. For fungal pathogens, the local melon cultivars M6, M26, M58, M120, M131 were resistant to race 1 of FOM, and all studied cultivars were susceptible to race 2 of this fungus. Two local melon cultivars M120 and M122 were resistant to *P. xanthii* races 1, 2 and 3. The wild

desert bitter watermelon was resistant to WMV, MNSV, CMV, MWMV and ToLCNDV, but not to PRSV. This study demonstrated that the Tunisian cucurbit genetic resources present valuable material for improving resistance to viral and fungal diseases in these crops.

CONCURRENT SESSION F1 – Biocontrol of plant pathogens: recent advances and future challenges

SESSION KEYNOTES

Developing microbial biocontrol agents: a long road full of question and exclamation marks. G. PUOPOLO^{1,2}.

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Our journey as researchers, from the isolation of microbial biocontrol agents (mBCAs) to their final registration and application in agriculture, is a rollercoaster of uncertainty and excitement. The first significant challenge we face is selecting the target plant pathogen and the crops it infects. This decision is crucial, as it determines whether the chosen plant pathogen aligns with the interests of companies. Once this hurdle is crossed, we encounter the question of where to search for new mBCAs. Typically, we isolate potential mBCAs based on the environmental niche and epidemiological parameters of the target plant pathogen. However, evaluating their efficacy in plant protection can yield surprises, and none of the isolated microorganisms could be effective in controlling the target plant pathogens. Recently, coupling classical isolation procedures with metagenomic analyses is a practical approach for focusing attention on specific microbial taxa associated with controlling plant pathogens. Screening for effective mBCAs may also lead to disappointment; indeed, even if they prove to be effective, some microorganisms must be discarded due to concerns primarily related to their impact on human health, their ability to grow cost-effectively, and their resistance to environmental stressors. Thus, incorporating these aspects into the design of the screening strategies could reduce the number of microorganisms that need to be evaluated for efficacy in controlling the target plant pathogens. Once we identify the right mBCA, we enter the phase we enjoy most: characterising its mode of action. We extensively use *in vitro* methodologies to assess the mBCA's capacity to produce antibiotics, lytic enzymes, phytohormones, plant resistance elicitors and

siderophores that can be involved in the direct and/or indirect control of plant pathogens. Recently, particular attention has been given to the capacity of mBCAs to release Volatile Organic Compounds (VOCs) that can have a toxic effect on plant pathogens. Moreover, microbial VOCs can also prevent plant pathogen infections by triggering resistance mechanisms in plants. Nowadays, we combine *in vitro* data with findings generated through omics technologies. Metabolomics and transcriptomics can provide a clearer understanding of how our selected mBCA can effectively control plant pathogens. These omics technologies could provide indications of genes and secondary metabolites, specifically expressed and/or produced during the microbe-microbe and plant-microbe interactions. Subsequently, we take our chosen mBCAs into the field, designing and formulating them appropriately. To do this, we will continue characterising our mBCAs by assessing their ability to resist desiccation, wash-off, and UV light. Based on this, we will select the most effective additives to protect our mBCAs against these factors. Finally, we could dedicate the last part of our efforts to providing all the data related to the ecotoxicity of the selected mBCAs. This phase is crucial as it ensures that our mBCAs are not only effective in controlling plant pathogens but also safe for the environment. All this information will be helpful for their registration and introduction of our mBCAs into the global market.

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Biological induced resistance in plants against pathogens: recent advances and future trends. A. SIAH. JUNIA-BioEcoAgro, Lille, France. E-MAIL: ali.siah@junia.com

In the current context promoting agroecology and sustainable agriculture, looking for eco-friendly control strategies of plant diseases is strongly encouraged. Plant resistance inducers from biological origin are considered as promising biocontrol tools that can fit with these challenges. These compounds, also referred to as bio-elicitors, are agents that confer improved protection to pathogen or pest attacks by inducing host defense mechanisms. They are effective against a wide range of plant pathogens, including viruses, bacteria, fungi, oomycetes, and nematodes. The mode of action of these compounds relies on the activation of the plant immune system rather than direct antimicrobial activity against

phytopathogens. Plant resistance inducers of natural origin include living microorganisms, plant extracts, microbial cell-wall extracts, microbial metabolites, minerals, and ions. Plant immunity mechanisms involve at the biochemical level the activation of several defense responses, including reactive oxygen species metabolism, pathogenesis-related protein synthesis, as well as phenylpropanoid and octadecanoid pathways. However, plant defense pathways triggered and the level of observed protection can vary depending on the considered compound, the targeted pathosystem, and the environmental conditions. Plant immunity can occur at two distinct scales, either locally (local acquired resistance, LAR) or systemic (systemic acquired resistance, SAR). LAR is expressed by cells, tissues, or even organs that have been treated by an elicitor, whereas SAR occurs in parts of the plants spatially distant from the treated tissues or organs. LAR is often a prerequisite to the expression of SAR, and SAR has been characterized as broader and more long-lasting than LAR. Other characteristic features of SAR are that it depends on the perception of salicylic acid (SA) and results on the expression of PR-protein encoding genes. Beside SAR, the systemic expression of induced resistance can also consist of induced systemic resistance (ISR), triggered at the root level by certain beneficial microorganisms occurring in the rhizosphere (such as plant growth-promoting bacteria, PGPR; plant growth-promoting fungi, PGPF; and arbuscular filamentous fungi, AMF). Although ISR also leads to a broad and long-lasting induced resistance, it relies on jasmonic acid (JA) perception and generally does not result in PR-protein biosynthesis. The compounds inducing the plant immune defenses may be distinguished into two categories, including elicitors, that directly activate host defense responses after their application, and priming agents, that require additional signals, such as pathogen recognition, to trigger full defense responses. Primed plants may develop a stronger and/or faster response pattern than so-called naïve (unprimed) plants. They can also detect the pathogen invasion at a lower threshold and hence react in a more sensitized way. Finally, primed plants can exhibit other response networks, involving specific defense pathways. An overview of the recent advances on plant resistance inducers as well as future challenges to promote their development and wide use in disease management programs will be discussed.

ORAL PRESENTATIONS

Root colonization by the oomycete biocontrol agent *Pythium oligandrum* in various French and world

vineyards and protection of grapevines exposed to biotic and abiotic stresses. L. CHABOUSSIE^{1,2}, A. YACOUB¹, J-F. GUISE², E. ATTARD¹, P. TROTEL-AZIZ², F. FONTAINE², P. REY¹. ¹Université de Pau et des Pays de l'Adour, CNRS, IPREM UMR 5254, 64000 Pau, France. ²Université de Reims Champagne-Ardenne, INRAE, RIBP USC 1488, 51100 Reims, France. E-MAIL: lisa.chaboussie@univ-pau.fr

Grapevine trunk diseases (GTDs) are complex and pose a significant threat to viticulture worldwide. This study investigates the potential of the oomycete *Pythium oligandrum* as a biocontrol agent to protect grapevines. Colonisation of grapevine roots by *P. oligandrum* was assessed in 11 cultivars from diverse wine-growing regions across France, Austria, Hungary, Iran, Israel, Italy, South Africa and Spain. Results showed that, with the exception of South Africa, most grapevine roots were colonised by *Pythium* spp. with echinulated oospores. ITS-region sequencing identified 90% of these strains as *P. oligandrum*. Amplification of the elicitor-like genes encoding the proteins, oligandrin and the cell wall protein (POD1), which are essential for the induction of systemic plant resistance, showed that the majority (80%) of these strains possessed both genes. Associated rhizospheric microbial communities (i.e. bacteria, fungi and oomycetes) have also been studied. In parallel, one Po strain was applied to grapevine cuttings later infected by *Neofusicoccum parvum*, a GTD- pathogen, with or without additional abiotic stresses (e.g. water deficit and heat shock). Grapevines colonised with *P. oligandrum* showed a 41% reduction in internal necrosis caused by *N. parvum* compared to control plants. However, under abiotic stresses, the protection provided by the biocontrol agent was no greater than under normal conditions. This research highlights the potential of *P. oligandrum* as an effective biocontrol agent and provides insights into its mechanism of action and its role in improving grapevine resistance to fungal diseases and abiotic stresses.

Environmental isolates of *Pseudomonas* spp. inhibit *Armillaria mellea* and promote microbiome- induced growth in olive plants. M.M. ACI¹, G.E. AGOSTEO¹, G. PELLE¹, V. CIANCI¹, M.G. LIDESTRI NICOSIA¹, A. MALACRINO^{1,2}, L. SCHENA¹. ¹Department of Agriculture, Università degli Studi Mediterranea di Reggio Calabria, Località Feo di Vito, 89124 Reggio Calabria, Italy. ²Department of Biological Sciences, Clemson University, Clemson, SC, USA. E-MAIL: miyassa.aci@unirc.it

Modern agriculture faces significant challenges in managing soil-borne pathogens such as *Armillaria mellea*, a destructive pathogenic fungus that causes root rot in several plant species. Harnessing the soil microbiomes offers a promising alternative to overcome the limitations of chemical control methods. In this study, four *Pseudomonas* isolates, selected from a wider panel of 155 strains isolated from soil, demonstrated a strong *in vitro* efficacy against *A. mellea*; three isolates inhibited fungal growth, and one prevented rhizomorph formation. Whole genome sequencing revealed gene clusters related to the biosynthesis of antifungal compounds and siderophores, implicating these as likely mechanisms for pathogen suppression and plant growth promotion. Microcosms experiments and amplicon metagenomic analysis revealed that the presence of the biocontrol agents increased plant biomass, while exerting only limited effects on the bacterial communities associated with olive plant roots and rhizosphere. Through structural equation modelling, we found that biocontrol agents influence plant biomass both directly and indirectly through the modulation of root and rhizosphere microbiota. These bacterial isolates offer an alternative approach to manage soilborne pathogens like *A. mellea*, promoting plant health and growth with minimal impact on the ecosystem.

This research was financially supported by i) PSR Calabria 2014/2020 Misura 16.2. Prevenzione e contrasto alla diffusione del marciume radicale fibroso da Armillaria in olivicoltura (ARMISTOP) and ii) Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

Biocontrol potential of *Bacillus halotolerans* against *Fusarium oxysporum* f. sp. *radicis-cucumerinum* under normal and saline soil conditions. F. SEVDALI¹, A. MONKA¹, M. MARINAKOU¹, E. POULAKI¹, P.C. TSALGATIDOU², E. PAPLOMATAS¹, A. VENIERAKI¹. ¹Laboratory of Plant Pathology, Department of Crop Science, Agricultural University of Athens, Iera Odos 75, 11855 Athens, Greece. ²Department of Agriculture, University of Peloponnese, 24100 Kalamata, Greece. E-MAIL: venieraki@aia.gr

Root and stem rot caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum* (FORC) is a serious threat to

cucumber cultivation worldwide, often leading to substantial yield losses. With the limited efficacy of chemical control methods, biological control strategies offer promising alternatives. This study evaluated the biocontrol potential of two endophytic *Bacillus halotolerans* strains (Cal.l.30 and Cal.f.4) against FORC under greenhouse conditions. Cucumber plants were treated with foliar applications of Cal.l.30, Cal.f.4, and their combination, with repeated treatments every 10 days over a 30-day period. Plants treated with the bacterial strains exhibited a delayed onset of disease symptoms and achieved approximately 67% protection compared to untreated controls. Additionally, the experiments were repeated under increasing soil salinity levels up to 200 mM NaCl. Under salinity stress, treatments with Cal.f.4 and the Cal.l.30 + Cal.f.4 mixture conferred enhanced protection against FORC, even under combined abiotic (salinity) and biotic (pathogen) stress conditions. The bacterial strain Cal.f.4 demonstrated a protective effect on FORC-infected plants grown under saline conditions, reducing overall disease and abiotic stress symptomatology by approximately 20%, and enhancing the production of healthy fruits by 30% compared to the negative control. Molecular analyses indicated the upregulation of defense-related gene expression in treated plants. These findings highlight the potential of *B. halotolerans* strains not only as effective biocontrol agents against FORC but also as enhancers of plant resilience under salinity stress. The results support their integration into sustainable disease management programs, especially in agricultural systems increasingly affected by soil salinization.

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Biological control of *Podosphaera xanthii* in cucumber by *Bacillus halotolerans* Cal.l.30: activation of plant defense pathways and role of secondary metabolites. T. MARGARITOPOULOU¹, S. NASTOS¹, K. KOT-SARIDIS², M. MARATOS¹, P.C. TSALGATIDOU³, A. VENIERAKI⁴, E. MARKELLOU¹. ¹Laboratory of Mycology, Scientific Directorate of Phytopathology, Benaki Phytopathological Institute, 14561 Athens, Greece. ^{2,1}Laboratory of Virology, Scientific Directorate of Phytopathology, Benaki Phytopathological Institute, 14561 Athens, Greece. ³Department of Agriculture, University of Peloponnese, 24100 Kalamata, Greece. ⁴Laboratory of Plant Pathol-

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Powdery mildew (PM), caused by *Podosphaera xanthii*, is a major constraint in cucumber (*Cucumis sativus* L.) cultivation worldwide. The extensive use of fungicides underscores the need for sustainable biological control strategies. This study evaluated three endophytic *Bacillus* strains — Cal.r.19 (*B. subtilis*), Cal.r.29 (*B. velezensis*), and Cal.l.30 (*B. halotolerans*) — isolated from *Calendula officinalis*, for their efficacy against PM. Greenhouse assays showed that Cal.l.30 consistently achieved a reduction of over 90% in disease severity without phytotoxic effects. In *in vivo* assays performed on cucumber cotyledons, the infection process and morphological development of *P. xanthii* were microscopically monitored in the presence of the biological control agents. Observations focused on pathogen structures such as spore germination, mycelial growth, and conidiophore formation, allowing for the assessment of the inhibitory effects exerted by the beneficial *Bacillus* strains at the early stages of infection. Additionally, gene expression analyses revealed that Cal.l.30-treated plants exhibited significantly higher expression of defense-related genes (PR-1, PR-4, WRKY, MAPK), indicating the activation of induced systemic resistance (ISR). Moreover, Cal.l.30 produces secondary metabolites with fungicidal activity. Protein-ligand docking studies suggest that bacillaene analogues interact with fungal ribosomal subunits, leading to the disruption of protein expression. This interference affects the functional activity of the *P. xanthii* effector protein RPS27A, thereby inhibiting fungal pathogenicity. These findings highlight a dual-action mechanism involving both direct antagonism and host defense priming. *Bacillus halotolerans* Cal.l.30 emerges as a strong candidate for the biocontrol of obligate pathogens such as *P. xanthii*, supporting the integration of endophytic beneficial bacteria into cucurbit disease management programs.

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Harnessing nava rhizosphere soil microbial communities to support traditional almond cultivation in Morocco. Z. BOUABIDI¹, A. MOSCA², D. NICOTRA², G. DIMARIA², F. MODICA², M.E. MASSIMINO², L.E.

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Almond (*Prunus dulcis* Mill.) cultivation in Morocco, particularly of traditional varieties such as Beldi, plays a vital role in supporting rural livelihoods and preserving agrobiodiversity. In many regions of the country, almonds are still propagated by seed, a practice that not only sustains genetic diversity but also reflects deep-rooted cultural traditions. However, climate variability, soil degradation, and plant diseases threaten the resilience and productivity of these systems. Native Mediterranean plants, known for their adaptability, may harbor beneficial microbiota with the potential to support almond growth and health. To investigate this potential, rhizospheric soils from five plant species were sampled, including almond trees and four wild Mediterranean species commonly associated with almond cultivation areas in Morocco. Total genomic DNA was extracted and subjected to amplicon-based metagenomic sequencing, targeting bacterial 16S rRNA and fungal ITS regions. Significant differences in bacterial richness and fungal diversity were detected using the Observed and Shannon α -diversity metrics, respectively. Similarly, β -diversity analysis revealed a clear differentiation of samples based on the plant species from which the soils were collected. A collection of cultivable microorganisms was established from all soils using multiple culture media. Bacterial isolates were screened for functional traits related to plant growth promotion, including salinity tolerance, phosphate solubilization, and siderophore production. Both bacterial and fungal isolates were assessed for antagonistic activity against phytopathogens. The results are expected to provide insights into plant-specific microbiomes in Moroccan soils and facilitate the selection of new microbial inoculants to enhance seed germination, as well as seedling growth and health.

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Isolation and characterization of Psa-bacteriophages from infected kiwifruit orchards in Portugal: potential for biocontrol of *Pseudomonas syringae* pv. *actinidia*. E. GIMRANOV^{1,2,3}, J. AZEREDO², C. SANTOS¹, L. MOURA³. ¹Biology Department, Faculty of Science, University of Porto (FCUP), 4169-007 Porto, Portugal. ²Centre of Biological Engineering, University of Minho, 4710-057 Braga, Portugal. ³CISAS—Centre for Research and Development in Agrifood Systems and Sustainability, Instituto Politécnico de Viana do Castelo, 4900-347 Viana do Castelo, Portugal. E-MAIL: up201804355@edu.fc.up.pt

Bacterial canker is a severe disease of *Actinidia* species. This disease is caused by *Pseudomonas syringae* pv. *actinidiae* (Psa), which is an extremely virulent pathogen compromising kiwi production worldwide. Efficient treatments are unavailable, and the recurrent use of copper-based compounds led to Psa-resistant populations. The bacteriophages represent a safer and sustainable solution for Psa control. The aim of this study is to isolate and characterize a population of Psa-bacteriophages from infected kiwifruit orchards in North and Central Portugal and evaluate their potential as biocontrol agents against Psa. To achieve this goal, bacteriophages were isolated from different samples. Bacteriophages lytic activity was characterized against representative Psa-lineages through efficiency of plating and inhibition curves. The most promising phages were selected for transmission-electron-microscopy and genome sequencing. Finally, *in vitro* assay to assess the impact of bacteriophages on Psa concentration in surface of kiwifruit leaves, as well as stability under different pH levels (5–8), temperatures (5–35°C), and UVA radiation exposure (180 min). In total, 41 bacteriophages were isolated, among which two were lytic and other lysogenic. Three types of tail morphologies were observed (*Podovirus*, *Myovirus* and *Siphovirus*). Only Brt_Psa3 demonstrated strong activity and specificity (91 % of the bacterial collection). Genomic analysis supports its safe application as it does not possess any bacterial virulence genes. *In vitro* experiments demonstrated significant reduction of Psa levels on kiwifruit surfaces and tolerance to different environmental conditions, highlighting the bacteriophage's therapeutic potential. Future work should focus on optimizing bacteriophage formulations and conducting extensive field trials.

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From Lab to plant phenotyping platform: a stepwise pipeline for the selection of high-performing antagonistic bacteria against tomato soil-borne pathogens.

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Biological control is a valuable tool for the sustainable management of soil-borne diseases in tomato. Digital technologies can play a key role in accelerating the screening of new, high-performing microbial biocontrol agents. In this study, a collection of eleven endophytic bacterial strains, isolated from tomato and compatible with *Trichoderma* spp. (with a potential for future consortium formulation), were evaluated against the tomato fungal pathogens *Fusarium oxysporum* f. sp. *lycopersici* and *Sclerotium rolfsii*. The assessment included both *in vitro* and *in vivo* assays, and biochemical characterization. The final stage of the evaluation was conducted using the PlantEye 500 multispectral dual scanner embedded within a plant phenotyping platform to acquire the plant's phenomic response to the microbial treatments. The experimental design included potted tomato plants 'Crovarese' (n=234) distributed among 11 microbial treatments along with non-treated and healthy controls, each with six replicates under three different conditions: infection with *F. oxysporum* f. sp. *lycopersici*, infection with *S. rolfsii*, and no infection. Multivariate analysis of 20 digitally computed traits identified *Peribacillus* spp. C5NA and *Neobacillus* spp. TR12 as the most effective against *F. oxysporum* f. sp. *lycopersici*, while *Peribacillus* spp. TR2 and C6 showed significant efficacy against *S. rolfsii*. Additionally, *Microbacterium* sp. TR9 was found to enhance digital biomass, plant height and NDVI, and to increase biofertilizing properties (ammonia production, nitrogen fixation, phosphate solubilization) and hormone-like activity (indoleacetic acid production), highlighting its potential as a promising biostimulant. Overall, the use of phenomics supported the selection of specialized and effective microbial strains for biocontrol applications.

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Evaluation of potential biological control agents against *Colletotrichum godetiae* and *C. nymphaeae*, the main causal agents of olive anthracnose in southern Spain. L. SÁNCHEZ-PEREIRA¹, B.I. ANTÓN-DOMÍNGUEZ¹, A. TRAPERO¹, C. AGUSTÍ-BRISACH¹.

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The use of beneficial microorganisms as biological control agents (BCAs) has been proposed as an alternative to control olive anthracnose, caused by *Colletotrichum* species. The antagonistic effects of 21 BCAs against *C. godetiae* (Col-511) and *C. nymphaeae* (Col-151) were evaluated. Copper sulfate (2000 mg/L) and Serenade ASO[®] (*Bacillus subtilis*, 8 mL L⁻¹) were included for comparison. Their inhibitory effect on mycelial growth and conidial production of both pathogens were assessed using dual culture assays *in vitro*. Bioassays on detached fruits and potted plants of cv. Arbequina were conducted to determine their effects on disease progression, severity, and incidence. Six BCAs suppressed Col-511 growth between 31 and 43%, while 12 inhibited Col-151 growth between 32 and 56%. Sixteen BCAs reduced Col-511 conidial production by >50%, but only five achieved >80% inhibition for Col-151. Copper sulfate and Serenade ASO[®] were the most effective *in vitro*. Five BCAs reduced disease progression by >30% in detached olives inoculated with Col-511; however, reductions did not exceed 23% in fruits inoculated with Col-151 compared to the untreated control. The three most effective BCAs (*Bacillus* sp. PV-700, *Bacillus* sp. PV-1394 and *Myrothecium inundatum* PV-329) from the detached fruits assay were further evaluated in potted plants. *In planta*, they reduced disease incidence between 32.9-57.8% for both *Colletotrichum* species compared to the untreated control, whereas copper sulfate reduced disease incidence by 60% and 50% for Col-511 and Col-151, respectively. These results will allow the selection of potential BCAs for further investigation of their mechanisms of action against the disease.

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POSTER PRESENTATIONS

Integrating biostimulation and plant pathogen control: endophytic *Trichoderma* strains boost yield and disease resistance in field crops. A. CSÓTÓ¹, P. RIZU², A. ZABIÁK³, L. KARAFFA⁴, E. SÁNDOR³. ¹University of Debrecen, Faculty of Agricultural and Food Science and Environmental Management, Institute of Plant Protection, H-4032 Debrecen, Böszörményi út 138., Hungary. ²KITE Zrt., Innovation Directorate, H- 4181 Nádudvar, Bem J. u. 1., Hungary. ³University of Debrecen, Faculty of Agricultural and Food Science and Environmental Management, Institute of Food Science, H-4032 Debrecen, Böszörményi út 138., Hungary. ⁴University of Debrecen, Faculty of Science and Technology, Department of Biochemical Engineering, H-4032, Debrecen, Egyetem tér 1, Hungary. E-MAIL: karaffa@agr.unideb.hu

The biostimulant potential of endophytic *Trichoderma* fungi in field conditions presents a viable option for sustainable agriculture. This research examined the impact of foliar spraying with a formulated consortium of *Trichoderma afroharzianum* TR04 and *T. simmonsii* TR05 on maize and sunflower fields in Hungary. The treatment significantly enhanced plant photosynthetic activity, evidenced by an increase in the number of viable leaves by up to 6.7% along with a rise in the SPAD index, indicating chlorophyll content, by up to 19.1% relative to the control. Under extreme drought conditions, maize yield doubled, increasing from 0.587 t/ha to 1.62 t/ha ($P < 0.001$). Additionally, consistent increases in harvested seed moisture content and sunflower plant height were observed. *In vitro* studies revealed that these *Trichoderma* strains demonstrated effective mycoparasitism against *Fusarium* and *Aspergillus* species, suggesting a significant potential to reduce mycotoxin contamination in field crops. Field applications significantly reduced the incidence of *Fusarium* head blight in wheat. This study supports the use of foliar applications of well-selected endophytic *Trichoderma* biostimulants to enhance crop yield, quality, and abiotic stress tolerance in both monocot maize and dicot sunflower crops, offering a sustainable and effective strategy for modern agriculture.

Biocontrol of tomato diseases by alfalfa extracts. A. TAGLIENTI¹, A. TAVA², S. BERTIN¹, E. BIAZZI², C. VINCENZO^{3,4}, C.L. CORRADO¹, S. SIMONI¹, C. PANE⁴. ¹CREA Research Centre for Plant Protection and Certification, Via C.G. Bertero 22, 00156 Rome, and Via Lanciola 12A, 50125 Florence, Italy. ²CREA Research Centre for Animal Production and Aquaculture, Viale Piacenza 29, 26900, Lodi (LO), Italy. ³Department of Agricultural Sciences, University of Naples Federico II, Piazza Carlo di Borbone 1, 80055 Portici (NA), Italy. ⁴CREA Research Centre for Vegetal and Ornamental Crops, Via Cavalligieri 51, 84098 Pontecagnano Faiano (SA), Italy. E-MAIL: anna.taglienti@crea.gov.it

The control of plant pathogens by compounds extracted from plants is increasingly popular as a sustainable alternative to agrochemicals, which are currently subjected to limitations in the European Union; in fact, regulatory frameworks aim at reducing the risks and impacts of synthetic pesticides in terms of toxicity, environmental pollution, and the development of resistance. In this context, plant extracts are gaining interest because of their effects on plant fitness and tolerance to biotic stressors. Flavonoids and saponins extracted from alfalfa, and prosapogenins obtained by alkaline hydrolysis of saponins, were the subjects of this study. The flavonoid extract was mainly composed of apigenin and chrysoeriol glycosides, while saponins contained bidesmosides of medicagenic and zanhic acids, and prosapogenins were made up only of the corresponding monodesmosidic compounds. An evaluation of their biopesticidal potential towards two fungi and a virus affecting tomato, as well as their possible undesired effects, was performed. Prosapogenins inhibited both the mycelial growth and conidia germination of *Alternaria alternata* and *Botrytis cinerea*, and showed lesion- containment efficacy against alternariosis and grey mould. The flavonoid mixture decreased the tomato spotted wilt orthotospovirus titer by 10⁻³-fold one-month post-inoculation, with a concurrent regression of symptoms. The assessment of non-target effects towards two beneficial mites, *Phytoseiulus persimilis* and *Amblyseius swirskii*, indicated that the treatments were not toxic, while some impact on fertility was observed especially with flavonoid extract on *A. swirskii*. The biopesticide activity against fungi and viruses in tomato, and their safe toxicity profile, support the application of these extracts in Integrated Pest Management programs.

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INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

Assessment of a *Streptomyces* strain and its metabolites against fungal pathogens on wheat and tomato. T. CONTE¹, M.G. MOREA¹, G. RICCIARDI¹, J. G. ZORRILLA², R. M. VARELA², F. A. MACIAS², M. MASI³, A. CIMMINO³, A. CARLUCCI¹. ¹Department of Agricultural Sciences, Food, Natural Resources and Engineering, University of Foggia, Foggia, FG 70122, Italy. ²Grupo de Alelopatía (GAC), Department of Organic Chemistry, University of Cadiz, Puerto Real, 11510, Spain. ³Department of Chemical Sciences, University of Napoli Federico II, University Complex of Monte Sant'Angelo, Naples, NA 80126, Italy. E-MAIL: thomas.conte@unifg.it

One of the most difficult challenges for modern agriculture is providing sustainable production capable of meeting the demands of a growing global population, which is expected to reach 9.8 billion people by 2050. In the past, chemical management helped achieve high productivity levels, but nowadays the environmental, economic and ethical issues associated with this approach underscore the need to find sustainable alternatives for plant protection against fungal diseases. In particular, natural products derived from microorganisms are potential tools against plant pathogens. Among these microorganisms, the genus *Streptomyces* is known for its capability to produce many largely unexplored bioactive compounds with potential antifungal activity. Through research activities carried out under both *in vitro* and *in vivo* conditions, four *Streptomyces* strains were screened for their potential use as Biocontrol Agents (BCAs). Among them, the strain *Strep_22* significantly reduced the growth of ten fungal pathogens (*Athelia rolfsii*, *Fusarium graminearum*, *F. oxysporum*, *F. solani*, *Rhizoctonia solani*, *Sclerotinia sclerotiorum*, and *Verticillium dahliae*). Moreover, the culture filtrate of the same *Streptomyces* strain was able to protect the roots of wheat and tomato and, at the same time, to promote plant growth. As a subsequent step, the biological activities of metabolites isolated from this same strain following chromatographic methods have also been explored.

Use of agricultural waste to control *Phytophthora cinnamomi* root disease on *Quercus ilex*. R. LÓPEZ¹, M.T. HIDALGO¹, M. GARCÍA¹, R. RODRÍGUEZ-ARCOS², A. JIMÉNEZ-ARAUJO², M.S. SERRANO¹. ¹Department of Agronomy, Campus de Rabanales Edif. C-4, University

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The circular economy could turn agricultural waste into innovative biocontrol tools. We studied the effect of two organic extracts enriched in bioactive compounds obtained from by-products of asparagus (spears) and date (seeds), as well as the residues of both species, on *Phytophthora cinnamomi* (*Pc*) root disease development *in planta*. Extracts were chosen by their known effectiveness on inhibition of *Pc* sporangial production. Residues previously shredded and extracts diluted in water were added separately at 0.05% (w/v) to a substrate (sand-peat 1:1 vol) infested with 250 chlamydospores of *Pc* per gram of dry soil and 10 pots per treatment were planted with 1-year-old *Quercus ilex* seedlings. Pots with infested and uninfested substrate were also prepared as controls. All pots were placed in a shade house and subjected to waterlogging 2 days per week. Fourteen weeks later, seedlings growing in infested and untreated substrate showed high levels of root symptoms, whereas plants growing in substrate treated with both extracts and residues showed a significant reduction of symptom severity, being the organic extract of date seeds the most effective treatment. It is highlighted that there are not significant differences between extract and residues for each crop although they were tested at the same dose regardless of their phytochemical concentration. Results suggest that soil application of asparagus spears and date seeds extract, and especially residues, should be considered for the reduction of the incidence of *Pc* root disease affecting *Quercus*.

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Potential antagonism and plant growth promoting capacities of two endophytic actinobacteria strains isolated from Moroccan aromatic and medicinal plants. EZ. OUBASSOU¹, L. AZLAY¹, Y. OUAHMANE¹, M. BAZ¹, A. JAMJARI¹, A. BERR³, M. BARAKATE^{1,2}. ¹Laboratory of Water Sciences, Microbial Biotechnologies and Natural Resource Sustainability; Microbial Biotechnology, AgroSciences and Environment & CNRST Labeled Research Unit N° 4, Faculty of Sciences Semlalia, Cadi Ayyad University, 40000 Marrakesh, Morocco. ²Biodiversity and Plant Sciences Program, AgroBioScience Department, College of Agriculture and Environmental Sciences, Mohammed VI Polytechnic University (UM6P), 43150 Benguerir, Morocco. ³Institut de Biologie Moléculaire des

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Since the discovery of actinomycin, *Actinobacteria* have provided many important compounds of high commercial value and continue to be routinely screened for new bioactive substances that are investigated for their agricultural applications. However, the intensive application of pesticides for crop protection can affect the environment and the health of humans and animals. Thus, the present work aims to assess the effectiveness of endophytic actinobacteria isolated from some interesting medicinal plants as biocontrol agents against *Pectobacterium* spp. and *Streptomyces scabies* which are the agents of soft rot and common scab, respectively and constitute a severe problem for potato crops. From a total of forty-four endophytic actinobacteria isolated from Aromatic and Medicinal plants, which were screened for their ability to inhibit *in vitro* different strains of *Pectobacterium* sp. and *S. scabies*, as well as for their PGPR traits, two *Streptomyces* isolates (A28 and A36) were selected for their *in vitro* antimicrobial activity against both pathogens and their ability to produce hydroxamate-type siderophores. *In vivo*, they significantly reduced ($P < 0.05$) the severity of soft rot on potato slices of cv. Desiree caused by *P. carotovorum* and *P. atrosepticum*. Furthermore, culture filtrate application of *Streptomyces* sp. A28 on potato tubers reduced the severity and incidence of the disease, as a preventive measure under storage conditions. This prevention was accompanied by the induction of defense enzymes related to oxidative metabolism and the phenylpropanoid pathway. Our data suggests that *Streptomyces* sp. A28 could be a promising natural biocontrol agent against both *Pectobacterium* spp. and *S. scabies*, and act as a biostimulant, to partially substitute the use of synthetic chemicals.

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Biocontrol potential of bacterial strains against root and stem rot of cucumber. S.K. SOULTATOS^{1,2}, N.E. KRASAGAKIS^{1,2}, M. PAPADAKI², V. MICHALOPOULOU^{3,4}, P.F. SARRIS^{3,4,5}, E.A. MARKAKIS^{1,2}. ¹Institute of Olive Tree, Subtropical Crops and Viticulture, Hellenic Agricultural Organization "DIMITRA", Kastorias 32A, 71307, Heraklion, Crete, Greece. ²School of Agricultural Sciences, Hellenic Mediterranean University, Stavromenos 71004, Heraklion, Crete, Greece. ³Institute of

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Root and stem rot of cucumber (*Cucumis sativus* L.) caused by the soil-borne fungus *Fusarium oxysporum* f. sp. *radicis-cucumerinum* (F.o.r.c.) is one of the most severe diseases of cucumber, with no effective chemical control available. In this study, we evaluated the suppressive effect of 31 bacterial strains isolated from the rhizosphere of halophytes in Crete (Greece) against root and stem rot of cucumber. Preliminary evaluation of bacterial strains *in-vitro* revealed that 17 strains inhibited fungal growth significantly ($\geq 20\%$). These most effective bacterial strains were further evaluated thoroughly in dual-culture (confrontation tests) and dual-plate (volatile tests) assays *in vitro*. Fungal parameters such as growth rate, hyphal width and sporulation of the pathogen were assessed. Confrontation tests revealed that 13 bacterial strains inhibited fungal growth rate and 6 decreased hyphal width significantly, whereas none of the tested strains inhibited sporulation of the pathogen. Volatile tests revealed that 5 strains were capable of inhibiting fungal growth, 8 strains decreased hyphal width and 5 strains decreased F.o.r.c. conidial production significantly. Based on *in vitro* assays, the 10 most effective bacterial strains were selected and evaluated further for their efficiency to protect cucumber against root and stem rot, by conducting *in planta* bioassays. The results indicated that one bacterial strain (code SRL871) reduced all disease parameters significantly compared to the positive control plants. Decreased, symptom severity was associated with lower plant colonization levels by the fungus and increased plant fresh weight.

The present study was conducted within the framework of the project entitled: «Innovations in Plant Protection for sustainable and environmentally friendly pest control» (Acronym InnoPP, Code Act TAA: TAEDR-0535675) «Funded by the European Union- Next Generation EU, Greece 2.0 National Recovery and Resilience plan»

Potential of indigenous biological agents for the control of grapevine trunk diseases in montenegrin viticulture. N. LATINOVIĆ¹, J. LATINOVIĆ¹, B. KANDIĆ¹, A. JELUŠIĆ², T. POPOVIĆ MILOVANOVIĆ³. ¹University of Montenegro, Biotechnical Faculty, Bulevar Mihaila Lalića 1, 81000 Podgorica, Montenegro. ²Institute for Multidisciplinary Research, University of Belgrade, Kneza Višeslava 1, 11030 Belgrade, Serbia. ³Institute for Plant

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Grapevine trunk diseases (GTDs) are a group of diseases caused by a large number of fungal phytopathogens that infect and colonize the woody tissue of grapevine, causing chronic or apoplectic decline of plants, and as such posing a serious threat to viticulture. Among the most virulent pathogens are *Neofusicoccum parvum*, the causal agent of Botryosphaeria dieback, and *Fomitiporia mediterranea* considered as the main causal agent of grapevine rot within the Esca disease complex. This study aimed to isolate and select potent indigenous bacteria collected from Montenegrin vineyards to serve in the control of GTDs. Isolations were performed from soil samples taken in a zone of grapevine root (Montenegro) on NA after soil was dissolved in water and heating at 80°C for 20 min. A total of 262 isolates were tested for antagonistic activity in a direct *in vitro* test on PDA. The appearance of a clear zone was checked after fungi development. Results showed that 32 Montenegrin isolates inhibited the growth of *N. parvum* by 34.0–65.0%, and *F. mediterranea* by 34.8–71.0%. Neighbour-joining phylogenetic analysis based on the 16S rRNA (primers P0/P6) sequences differentiated those isolates into six clusters but lacked the resolution to differentiate between several species. Six groups were as follows: I (21 isolates: *Bacillus amyloliquefaciens*/*B. velezensis*/*B. siamensis*/*B. subtilis*), II (2: *B. subtilis*/*B. velezensis*/*B. tequilensis*), III (5: *B. halotolerans*/*B. mojavensis*/*B. subtilis*/*B. velezensis*/*B. tequilensis*/*B. spizizenii*), IV (1: *B. licheniformis*/*B. paralicheniformis*), V (2: *B. cereus*/*B. thuringiensis*/*B. albus*/*B. anthracis*), VI (1: *Paenibacillus peoriae*/*P. polymyxa*). Sequencing of housekeeping genes would provide more accurate identification.

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Effectiveness of Tunisian Bacteria B8, B9, and B15 in controlling *Diplodia seriata* in vitro and in planta in grapevine. H. TRABELSI^{1,2}, L. KALAI GRAMI², Y. ZAOUALI¹, C. MESSOUAD¹, F. FONTAINE³, A. BEN GHNAYA-CHAKROUN¹. ¹University of Carthage, INSAT, Laboratory of nanobiotechnologies LR17ES22, Department of Biology, North Urbain Center BP 676, Tunis 1080, Tunisia. ²Laboratory of Biotechnology Applied to Agriculture, National Institute of Agricultural Research

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Grapevine trunk diseases (GTDs), such as Esca and Botryosphaeria dieback, pose a major threat to viticulture worldwide. In Tunisia, little is known about these diseases, but their incidence is increasing, and no effective treatment currently exists. *Diplodia seriata* is a fungal pathogen and one of the etiological agents of Botryosphaeria dieback in grapevines. This study evaluates the effectiveness of three bacteria (B8, B9, B15), isolated from Tunisian grapevines, in controlling *Diplodia seriata* strain *Ds type18*. The bacteria were tested for their ability to colonize grapevine vitroplants, inhibit fungal growth *in vitro*, and reduce disease severity *in planta* (detached canes assay). The results show that B8, B9, and B15: i) successfully colonized grapevine vitroplants from roots to leaves; ii) Inhibited *Ds type18* *in vitro* (64.69 %, 51.25 %, and 56.3 % inhibition, respectively); iii) significantly reduced disease severity *in planta* (54 %, 47 %, and 44 % protection, respectively). These findings highlight the potential of these bacteria as promising biocontrol agents for *Diplodia seriata*. Future studies should focus on field applications, bioformulation and ecological assessments.

Development of sustainable activity of some plant extracts against the fire blight. S. LAALA¹, D. BOUAI-CHA^{1,3}, S. CHAMAT², H. DJABALI¹, A. KERROUM¹, F. VALENTINI³. ¹Laboratory of Phytopathology and Molecular Biology, Botany Department, Higher National Agronomic School (ENSA), EL-Harrach, Algiers, Algeria. ²Laboratory of chemistry, Research Center Scientific and Technical in Analyzes Physico-chemical (CRAPC), Tipaza, Algeria. ³CIHEAM Bari, Centre International de Hautes Etudes Agronomiques Méditerranéennes, via Ceglie 9, 70010 Valenzano (BA), Italy. E-MAIL: samia.laala@edu.ensa.dz

Fire blight is a serious disease in many countries, and even a small outbreak could have a significant economic impact on a newly affected country, due to potential restrictions on international plant trade. The development of sustainable, effective, and eco-friendly compounds to contain and counteract pathogen development is a key goal of phytopathological research, which focuses on identifying natural molecules or compounds effective against phytopathogenic bacteria. There are several

research studies in literature on extracts and/or compounds of plant origin that are effective in counteracting pathogens and pests that have subsequently been used in the open field in integrated pest management programs. The present study investigates the potential efficacy of four plant extracts (pomegranate, onion, eucalyptus, and garlic) for the control of *Erwinia amylovora*. The selected plants were processed using the maceration technique to obtain aqueous and organic extracts. These extracts were evaluated for their antibacterial activity *in vitro* against *E. amylovora* through disc diffusion, well diffusion, and agar paper tests. Additionally, *vivo* assays were conducted on immature pear fruits and detached pear leaves. From the results obtained, it was found that pomegranate ethanolic extract showed increased and remarkable antimicrobial activity against the pathogen during the biological test (disc diffusion test, well diffusion test and agar paper test) and bioassay (immature pear fruit and detached pear leaves) on reference strains. Pomegranate extract contains a high concentration of bioactive compounds.

Comparative evaluation of biological control agents against *Colletotrichum* species causing olive anthracnose in Italy and Spain. L. SÁNCHEZ-PEREIRA¹, M. RIOLO², C. AGUSTÍ-BRISACH¹, S.O. CACCIOLA².

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Olive (*Olea europaea*) anthracnose, caused by *Colletotrichum* species, is a major disease of this crop worldwide, particularly across the Mediterranean Basin. This study evaluated six potential biological control agents (BCAs; *Aureobasidium pullulans* PV-1033; *Bacillus amyloliquefaciens* PV-700; *Epicoccum nigrum* RB4C; *Phoma* sp. PV-375; *Trichoderma asperellum* IMI393899; *T. atroviride* TRICH-S) against representative isolates of the main *Colletotrichum* species causing the disease in Italy and Spain: *Colletotrichum acutatum* C9D2C and UWS149, *C. godetiae* OLP12, OLP16, Col-508, Col-511 and *C. nymphaeae* Col-151 and Col-466. Copper sulfate (2 g L⁻¹) and *Bacillus subtilis* (Serenade ASO[®], 8 mL L⁻¹) were used as reference treatments. *In vitro* assays assessed the BCAs' inhibitory effects on mycelial growth and their production of volatile organic compounds (VOCs). All BCAs significantly inhibited mycelial growth in dual cultures and produced VOCs that suppressed the growth of all *Colletotrichum* species. TRICH-S and IMI393899

showed the strongest inhibition in both tests. The pathogenicity of all the *Colletotrichum* isolates was previously assessed by inoculating detached 'Arbequina' fruits. All isolates developed symptoms, with disease incidence ranging from 41.67% to 98.33%. The most virulent *Colletotrichum* isolates (C9D2C, OLP12, Col-511, and Col-151) were used to test the effect of the BCAs on disease development in detached fruits. To this end, BCA suspensions were applied by spraying 4 days and 24 hours prior to pathogen inoculation. IMI393899 was the most effective against C9D2C and OLP12; PV-700 against Col-511; and RB4C and PV-700 against Col-151. These findings identify promising BCAs for further *in planta* evaluation and investigation of their mechanisms of action.

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Multifunctional bio-based strategies for sustainable control of postharvest fungal pathogens in citrus: yeasts, fermentates and edible coatings. M. RIOLO¹, F. LA SPADA¹, S. CONTI TAGUALI¹, R. PARLASCINO, C. BUA, M.C. TAMBE¹, A. PANE¹, G. MECA², S.O.C. CACCIOLA¹.

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Postharvest rots significantly impact the shelf life and marketability of citrus fruit in the Mediterranean region. In this study, different methods based on biocontrol agents (BCAs) and bio-formulations to control key postharvest pathogens were evaluated. The yeast *Candida oleophila* strain O was tested against *Penicillium digitatum*, the causal agent of green mold in citrus, using in

vivo assays simulating all stages of the postharvest supply chain. To optimize the treatment, the yeast applied to various citrus varieties before, during, and after pathogen inoculation. *Candida oleophila* showed an efficacy comparable to imazalil, the most popular fungicide used for post-harvest treatments of citrus fruit. Transcriptomic analyses of fruits treated with the yeast revealed activation of defense genes, including PAL, β -1,3-glucanases, and peroxidase, indicating multiple mechanisms of action, competition with or antagonism against the pathogen and induction of plant resistance. Cell-free supernatants (CFSs) from fermentates of *Lactiplantibacillus plantarum* and the cyanobacterium *Arthrospira platensis* inhibited toxigenic fungi, such as *Fusarium graminearum* and *Penicillium expansum*, both *in vitro* and *in vivo* on various agricultural products including lemons. The antifungal properties of CFSs were mainly due to acidic metabolites, such as lactic acid, benzoic acid, and DL-3-phenyllactic acid. Finally, chitosan-based, biodegradable-edible fruit coatings activated by nesting the chitosan film with antifungal agents derived from citrus fruit peel were developed. Bioactive coatings effectively prevented *Penicillium* rots and improved UV shielding. BCAs, bioactive fermentates and biodegradable-edible coatings can be exploited as part of integrated and sustainable management strategies of post-harvest decay of citrus fruits.

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Exploring the potential of mycoviruses as new biological control agents (BCAs) against the grey mould fungus *Botrytis cinerea*. S. LAERA, M. CRUDELE, C. ROTOLO, M. MARASHI, L. VACCARO, P.R. ROTONDO, D. DI COSMO, T. MASCIA, F. FARETRA, R.M. DE MICCOLIS ANGELINI. *Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, Bari, Italy.* E-MAIL: ritamilvia.demiccolisangelini@uniba.it

Botrytis cinerea is the fungus that causes grey mould, a major disease affecting a wide range of host plants, both in the field and postharvest, leading to significant economic losses. Several mycoviruses are known to infect fungal plant pathogens, offering a potential new approach for biocontrol. In this study, we investigated the phylogenetic relationships of mycoviruses previously identified through a metagenomic analysis in *B. cinerea*, the presence of viruses putatively associated to

host hypovirulence in individual isolates of the fungus, and their possible influence on the host phenotype. The analysis was performed on 11 (+) ssRNA viruses and revealed a great diversity and the phylogenetic relationships of the identified mycoviruses with known viruses within the *Hypoviridae*, *Fusariviridae*, and *Mitoviridae* families. The presence of eight viruses was checked by RT-PCR in 28 *B. cinerea* isolates, revealing single or multiple viral infections. Among the tested viruses, *Botrytis cinerea mymonavirus 1* (BcMyV1) was selected for further analysis. Virus-free isogenic fungal lines were generated by antibiotic treatments combined with hyphal tipping. *In vitro* assays in virus-infected and virus-free isogenic lines revealed that BcMyV1 did not affect colony growth and spore germination. On artificially inoculated grape berries, a slightly reduced virulence of the virus-infected isolate compared to the virus-free isogenic lines was observed but this difference was not statistically significant. The results obtained in this study contribute to the understanding of mycovirus diversity in *B. cinerea* and highlight the potential use of mycoviruses as new candidate biological control agents against grey mould.

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A new approach for the obtention of *Ascophyllum nodosum* stimulants for plant defense and improved seedling development. G. OJEDA^{1,2}, J. ARAUJO², J. COTAS³, L. PEREIRA³, F.J. CEBALLOS-BURGOS^{4,5}, C. MALEITA^{4,5}, K. BAHCEVANDZIEV⁶, A. SOBRAL². ¹*Escuela de Ciencias Agrícolas, Pecuarias y del Medio Ambiente ECAPMA, Universidad Nacional Abierta y a Distancia UNAD, Cl. 14 Sur # 14-23, Bogotá, DC, Colombia.* ²*Coimbra Chemistry Centre, Department of Chemistry, University of Coimbra, Coimbra, Portugal.* ³*CFE—Centre for Functional Ecology: Science for People & Planet, Marine Resources, Conservation and Technology—Marine Algae Lab, Department of Life Sciences, University of Coimbra, 3000-456 Coimbra, Portugal.* ⁴*University of Coimbra, Centre for Functional Ecology - Science for People & the Planet (CFE), Department of Life Sciences, Calçada Martim de Freitas, 3000-456 Coimbra, Portugal.* ⁵*University of Coimbra, Chemical Engineering and Renewable Resources for Sustainability (CERES), Department of Chemical Engineering, Rua Sílvio Lima, Pólo II – Pinhal de Marrocos, 3030-790 Coimbra, Portugal.* ⁶*Research Centre for Natural Resources, Environment and Society (CERNAS), Coimbra Agriculture School, Polytech-*

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Due to its bioactive compounds, extracts from the brown algae *Ascophyllum nodosum* (ASn) can be exploited as plant nutritional supplements, acting as biostimulants for plant defence induction. Even though abundant literature reports the effects of *A. nodosum*-based products on plant growth and protection, studies involving simple, low-temperature extraction methods are scarce. We compared two extraction methods to obtain a plant biostimulant to promote plant growth and to induce resistance against the root-knot nematode *Meloidogyne incognita*, an important pest for agriculture worldwide. The protocol we followed was: (a) Biomass: 10 g of *A. nodosum* biomass dried at 60°C. (b) Extraction method: (1) Soxhlet (water, 100°C) – AsnH2500, (2) drip column (water, 20°C) – AsnG2500. (c) Experimental design: 3 treatments (control, AsnH2500, AsnG2500). (d) Seed growth: *Brassica rapa* cv. cymosa (25/dish, 10 days at 21.6°C). (e) *M. incognita* mobility: 50 nematodes*4 replicates incubated at 24°C for 72 h. (f) Chemical characterization: F-TIR and GC/MS analysis. As a final result we could see that the ASnH2500 (high temperature) and ASnG2500 (low temperature) extracts increased the roots, hypocotyl, and total dry weight of *B. rapa* compared to the control treatment. In addition, several compounds responsible for promoting growth, modulating defense, and stress responses were detected. Therefore, it is possible to obtain multipurpose biofertilizers useful for improving plant growth and inducing plant resistance against pests and environmental stress at low temperatures. Although the nematostatic effect of extracts on *M. incognita* was not significant, it opens a way to obtain new valuable extracts under different temperatures.

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Evaluation of biostimulants and beneficial bacteria for controlling Pistachio decline. W. ROUISSI¹, I. OUEGHRI^{1,2}, I. HEMISSI¹, A. CHELLI CHAABOUNI¹. ¹National Institute of Agronomic Research of Tunisia, University of Carthage, Tunis, Tunisia. ²Faculty of science,

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In recent years, the experimental pistachio orchards at INRAT's Agricultural experimentation Unit in Mor-nag have faced phytosanitary problems, leading to tree decline and reduced productivity. Isolation, Microscopic and molecular identification showed that damages are mostly due to *Alternaria alternata* and *Neofusicoccum mediterraneum*. To address these challenges sustainably, biostimulants were tested as a potential solution to improve tree resilience and enhance yield and quality. *In vivo* trial was conducted on a 33-year-old orchard on Pistachio trees cv. Mateur grafted onto *Pistacia vera* and *Pistacia atlantica* rootstocks, to evaluate the combined effect of two biostimulant products, PLANTA and EPSOMIT, under three treatments: rainfed control (T0), irrigated (T1), and PLANTA-EPSOMIT combined treatment (T2) with regular phytosanitary monitoring. A pre-treatment with Methyl- thiophanate was applied on all the orchard. Observations conducted after treatments during filed inspections showed no new cases of plant dieback and a significant reduction in necrotic leaves previously caused by *Alternaria*. Absence of infected fruits on bunches of trees treated with Planta-Epsomit comparing to the control T0 was also recorded. Additionally, PLANTA and EPSOMIT effectiveness against *Neofusicoccum mediterraneum* was assessed *in vitro*, comparing to plant growth-promoting bacteria (B1, B2, and B3). The results revealed that both PLANTA and B3 exhibited the highest antifungal activity, by reducing significantly the fungal mycelium growth by 65% and 50% respectively. This highlights B3 as a promising biological control agent, suggesting that beneficial bacteria and biostimulants, could play a key role in mitigating pistachio decline while reducing reliance on chemical treatments.

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Kinetics of lipopeptide production from the supernatant of *Bacillus methyltrophicus* strain TEB1 and dose-response effect of these lipopeptides on the fungus *Pleurotus tracheiphilus*. L. KALAI-GRAMI^{1,2}, I. BEN SLIMENE², O. NAILI¹, F. LIMAM², M. MNARI HATTAB¹, M. RABEH HAJLAOUI¹. ¹Laboratoire de Biotechnologie Appliquée à l'Agriculture, INRA, Hedi Karray, Tunis, Tunisia. ²Laboratoire des Substances Bioactives, CBBC, Hammam-Lif, Tunisia. E-MAIL: Kalai_leila@yahoo.fr

Citrus mal secco is a highly destructive vascular disease caused by the fungus *Pleurothidium tracheiphilus* which has a relevant economic impact on the lemon (Citrus lemon) industry in the mediterranean region. A lipopeptide-producing endophytic strain *Bacillus methyltrophicus* TEB1, obtained from citrus leaves, exhibited a good *in vitro* activity against *P. tracheiphilus* in dual cultures as well as with the well diffusion method. In this study, we compared the Pt antifungal activity of the supernatant of TEB1 spore harvested on Difco sporulation media, with that of vegetative cell harvested on LB medium. We also investigated the optimal time course of lipopeptide production from vegetative cell supernatant as well as the minimum inhibitory concentration (MIC) of these lipopeptides against the fungus *P. tracheiphilus* and their dose-response effect on the germination and morphology of conidia. Results showed that TEB1 vegetative cells exhibited higher inhibition than spores. Lipopeptide production from vegetative cell supernatant started at the early growth phase and reached a plateau after 36 h of culture reducing the mycelium growth by 80%. The lipopeptide extract harvested at the stationary phase efficiently inhibited *P. tracheiphilus* mycelial growth and the MIC values displaying 50 and 90% inhibition of conidial germination were approximately 47.5 and 100 mg mL⁻¹, respectively. Furthermore, morphological analysis showed that an increase in the concentration of the lipopeptide extract till 3 mg mL⁻¹ induced 10% swelling and 3% crumbling of fungal conidia while an increase to 5 mg mL⁻¹ resulted in 40% swelling and 20% crumbling.

Identification and characterization of autochthonous microbial communities' inhabiting Apulian vineyards soil for natural improving grapevine health. A. AGNUSDEI, E. CHIAROMONTE, M. MOSCHETTA, D. SALAMONE, V. MONTILON, S. POLLASTRO, D. GERIN, F. FARETRA. *Department of Soil, Plant and Food Sciences, University of Bari, Via G. Amendola, 165A/70126 Bari, Italy.* E-MAIL: donato.gerin@uniba.it

Microorganisms can benefit plant health by enhancing plant nutrient-use efficiency and protecting plants against biotic and abiotic stresses. This study aimed to isolate and characterize as plant growth promotion and biocontrol potential autochthonous microbial communities from soil samples of two vineyards located in Rutigliano (BA; vineyard 1) and Ginosa marina (TA; vineyard 2). A total of No. 16 bacterial isolates, selected as the most abundant morphologies (8 in both vineyards representing 5.3×10⁵ CFU g⁻¹ of dry soil in vine-

yard 1 and 4.8×10⁵ CFU g⁻¹ in vineyard 2) on nutrient agar (NA) and starch casein agar (SCA) were identified by morphotypes and 16SrDNA region sequencing, and characterized for indole 3-acetyl acid (IAA) production, phosphate solubilisation, copper sensitivity and salt tolerance. *Pseudomonas* sp., *Raoultella* sp. and *Enterobacter* produced the higher amount of IAA (1.5, 0.9 and 2.2 mg L⁻¹), while *Advenella* sp., *Raoultella* sp. and *Enterobacter* were more able to solubilize phosphate (solubilization index≥3). Finally, almost all isolates were inhibited by CuSO₄·5H₂O at the concentrations over 300 µg L⁻¹ and by >2% NaCl concentration, while two *Pseudomonas* sp. isolates, *Raoultella* sp., *Pseudarthrobacter* sp. and *Cupriavidus* sp. were able to grow also in presence of 4% of NaCl. Their ability to inhibit *Dematophora necatrix* was also assessed in dual culture assay. Additionally, a beneficial like *Pseudomonas* sp. reduced the colony growth of the fungus for ~ 40% by both direct- and VOCs mediated-antibiosis. This work proposed a high-throughput approach to screening soil microorganisms with beneficial potential.

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Effect of LED UV-C rays on ToBRFV infected tomato seeds. L. DONATI¹, S. BOLLANTI², D. MURRA², P. DI LAZZARO², L. FRY³, A. MANGILLI¹, D. LUISON¹, A. TIBERINI¹. ¹CREA Research Centre for Plant Protection and Certification, Via C.G. Bertero 22, 00156 Rome, Italy; ²ENEA, Nuclear Department, Fusion and Technology for Nuclear Safety and Security, Frascati Research Center, via E. Fermi 45, 00044 Frascati, Italy. E-MAIL: livia.donati@crea.gov.it

Tomato brown rugose fruit virus-ToBRFV (*tobamovirus fructirugosum*) (genus *tobamovirus*, family: Virgaviridae) is an important crop pathogen for tomato industries, with several outbreaks worldwide. ToBRFV is mechanically transmitted, also through contaminated seeds, promoting further dissemination and spread to new growing areas. ToBRFV is located only on the seed coat and infect emerging seedlings. Several papers describe physical methods such as UV radiation to stimulate the hormetic effect in plants, but little is known regarding their use in the treatment of seeds and plant material to inac-

tivate any pathogens present on the surface. In tomato, it has already been demonstrated that hormetic UV-C treatments on seeds can control diseases caused by different fungi, but no positive results have been obtained against viral pathogens. In this work, the effects of UV-C radiation at doses 12 kJ/m² emitted by light-emitting diode (LED) arrays on ToBRFV-contaminated seeds were evaluated, to assess both the virucide and inactivation effect. After the irradiations, the viral titre was assessed by TaqMan qPCR assay and the virus infectivity by bioassay on both tobacco indicator plants through mechanical inoculation. Biostimulation parameters (% germination, root and hypocotyl length, DW, FW and free water of seedlings) were evaluated during germination. The hormetic effect was evaluated using the MPM-100 which measures the quantities of chlorophylls, flavonols and anthocyanins in irradiated plants compared to control plants. Although UV-C treatment had no effect in reducing the viral titre of seeds, ToBRFV inactivation was observed for the highest dose (12kJ m²) preventing their transmission to the indicator plants.

Selection of *Bacillus* spp. for the control of *Sclerotinia sclerotiorum* and growth promotion in common bean. A.C.M. SANTOS¹, E.A. LOPES¹, W.V. CUNHA², Z. BOUABIDI³, L.E. VISÔTTO¹. ¹Federal University of Viçosa - Campus Rio Paranaíba. Road MG 230 Km 7, Zip code 38810-000, Rio Paranaíba - MG, Brazil. ²University of Patos de Minas, Via Maj. Gote, 808 - Caiçaras, Zip code 38700-207, Patos de Minas - MG, Brazil. ³Natural Resources Engineering and Environmental Impacts Team, Multidisciplinary Research and Innovation Laboratory, Polydisciplinary Faculty of Khouribga (FPK), Sultan Moulay Slimane University, BP 145, 25000 Khouribga, Morocco. E-MAIL: zinebbouabidi3@gmail.com

Some bacteria suppress the development of phytopathogenic fungi and can contribute to integrated pest management. The aim of this study was to select isolates of *Bacillus* spp. with antagonistic potential for the control of *Sclerotinia sclerotiorum*. The suppression of mycelial growth and inhibition of the germination of sclerotia of *S. sclerotiorum*, by diffusible and volatile substances produced by 12 isolates of *Bacillus* spp., were evaluated. Among the isolates, GB01, GB05, GB13, GB14 and GB16 produced diffusible substances capable of inhibiting mycelial growth by 39.0; 23.7; 19.6; 26.5 and 15.3%, respectively. Isolates GB10, GB01, GB14 and GB16 inhibited by 47.1; 60.0; 69.0 and 100% the germination of sclerotia at an incubation temperature of 20°C and 47.1; 60.0; 69.0 and 100.0% at 28°C, respectively. Volatile

organic compounds produced by bacterial isolates did not reduce the development of *S. sclerotiorum* and the germination of sclerotia. The isolates GB01, GB14 and GB16, selected in the in vitro tests, were evaluated for their ability to promote the growth of beans and reduce the incidence of white mold. The percentage of diseased plants varied between 0% (isolated GB14) and 12.5% (negative control). No bacterial isolate promoted bean growth. The isolates GB01, GB14 and GB16 from *Bacillus* spp. have potential for use in the biocontrol of *S. sclerotiorum*, however, it is necessary to establish standards that suit the particularities regarding the adaptation of these microorganisms to the environment.

Science-based innovative solutions transfer for chestnut blight biocontrol by hypovirulence in Portugal. E. GOUVEIA^{1,2}, J. ARAÚJO², V. COELHO². ¹CIMO, LA SusTEC, Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300- 253 Bragança, Portugal. ²Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300- 253 Bragança, Portugal. E-MAIL: egouveia@ipb.pt

Chestnut blight (CB) is associated with *Cryphonectria parasitica* (Murill) Barr. is introduced and established in Europe since 1938. In Portugal was introduced only in 1989, one of the last European countries where the disease was established as a rapid and devastating epidemic. Twelve years were enough to the disease be present in all chestnut areas of Portugal that cover more than 50 thousand hectares. Eradication, sanitary, physical and cultural measures didn't stop the spread and didn't control the disease. The imminent catastrophic situation of the chestnut ecosystem mobilizes regional entities and farmers for more efficient control means be applied. Biological control of CB by hypovirulence is associated with RNA virus (*Cryphonectria Hypovirus 1* - CHV1) that infect *C. parasitica* which reduces sporulation and virulence. This method is considered very efficient and recommended by EFSA to be applied when the natural hypovirulence is rare or not present. In this work we present the long, hard and persistent work, of which we are proud and pleased for having made possible the treatment of CB based on the principles of biological control by hypovirulence. After being officially authorized in 2015, and still in practice, more than 150 thousand chestnuts trees are treated in more than 7 thousand plots' and have participated in this program more than 3 thousand chestnut producers. In addition to healing cankers and tree recovery and the sustainability of the method a second level of outcomes was the successful transfer technology and large application in the field by

the farmers, the end users who can put hypovirulence in practice.

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Evaluation of *Dunaliella salina* extracts as potential antifungal and antioxidant agents *in vitro*. S. AOUZAL^{1,2}, FZ. IBN MOKHTAR^{1,3}, A. KHTIRA^{1,4}, D. PLISCHKE², S. KRIMI BENCHEQROUN¹. ¹Plant Protection Laboratory, Regional Center of Agricultural Research of Settlat, National Institute of Agricultural Research, Avenue Ennasr, BP 415 Rabat Principal, 10090 Rabat, Morocco. ²Viride Maroc SARL, Av. El Aargoub 01-7724, 73000 Dakhla, Morocco. ³Laboratory of Agri-Food and Health, Faculty of Sciences and Techniques, Hassan First University, B.P. 539, 26000 Settlat, Morocco. ⁴Laboratory of Functional Ecology and Environmental Engineering, Faculty of Sciences and Technology, Sidi Mohamed Ben Abdallah University, Fes, Morocco. E-MAIL: sanae.krimibenchecqroun@inra.ma

Dunaliella salina is a microalga known for its wide range of bioactive compounds. In this study, the antioxidant and antifungal properties of various *D. salina* extracts were evaluated *in vitro*. Antifungal activity was assessed against *Fusarium oxysporum* (FOC) and *Ascochyta rabiei* (AB) using the broth microdilution technique to determine the Minimum Inhibitory Concentration (MIC) and Minimum Fungicidal Concentration (MFC). Antioxidant activity was measured through DPPH, ABTS, and total phenolic content (TPC) assays. Statistical analysis revealed significant differences among the extracts in both antifungal and antioxidant activities. Among the three tested extracts, extract 1 was the only one exhibiting fungicidal effect against FOC with a MIC of 1.25 mg mL⁻¹ and an MFC of 5 mg mL⁻¹. Extracts 2 and 3 demonstrated fungistatic activity against both pathogens. In antioxidant assays, extract 1 also demonstrated the highest activity across DPPH, ABTS, and TPC tests with the values of 37.77%, 0.42 mg TE mg⁻¹, and 35.37 µg GAE mg⁻¹, respectively. These findings highlight the promising antifungal and antioxidant potential of *D. salina* extracts, suggesting their possible use in agricultural and pharmaceutical applications. Further studies are recommended to isolate and characterize the specific bioactive compounds responsible for these activities.

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In planta evaluation of bacterial isolates as biological agents against *Clavibacter michiganensis* and assessment of their plant growth promotion properties. G. MERMIGKA^{1,2}, M. PANIS¹, M.G. PAGOULATOU¹, V. MICHALOPOULOU³, P. SARRIS^{3,4}, D.E. GOUMAS^{1,2}. ¹Hellenic Mediterranean University (HMU), School of Agricultural Sciences, Department of Agriculture, Laboratory of Biotechnological Applications and Phytopathology, GR71004, Heraklion, Greece, ²Hellenic Mediterranean University (HMU), University Research Centre, Institute of Agri-Food and Life Sciences, GR71410, Heraklion, Greece, ³Institute of Molecular Biology and Biotechnology, Foundation for Research and Technology-Hellas, Heraklion, Greece, ⁴Department of Biology, University of Crete, Heraklion, Greece. E-MAIL: dgoumas@hmu.gr

The use of beneficial microbes has emerged as a promising, sustainable alternative to traditional chemical inputs, harnessing the power of naturally occurring microorganisms to enhance crop growth and resilience. These microbes work symbiotically with plants to improve nutrient uptake but also to bolster plant defenses by suppressing soil-borne pathogens and pests, thus decreasing reliance on chemical pesticides. In our laboratory, we have isolated various bacteria belonging to the *Bacillus* spp. and evaluated their ability to inhibit *in vitro* the growth of the phytopathogen *Clavibacter michiganensis* (Cm). Among these bacteria, eight showed a more profound effect against Cm. The aim of this study was to evaluate these bacteria on Cm-infected plants. For this purpose, tomato seedling plants were root-drenched with bacterial suspensions of each one of the eight bacterial species. Five days later, the plants were inoculated with Cm, placed in a growth chamber, and the symptoms were monitored for a period of 30 days. In parallel with the above experiments, healthy tomato plants were root-drenched with each of the bacterial suspensions and used to evaluate the plant growth promoting features of these treatments. From the latter experiments, we found that the bacterial strains used had no effect on the phenotypical characteristics of the plants. Concerning the effect of the bacteria on the Cm infection, we found that one strain had a statistically significant delay in the onset of the infection symptoms. This strain will be used in future experiments under greenhouse conditions to further evaluate its effect against Cm.

The study was part of the project "Innovations in Plant Protection for sustainable and environmentally friendly pest control, InnoPP"- TAEDR-0535675 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.

Biocontrol over chemicals: native microorganisms combat soilborne pathogens in arid agriculture.

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Soilborne pathogens, particularly *Pythium aphanidermatum*, cause severe damping-off in cucumber and tomato crops and are a major constraint to vegetable production in the GCC countries. This study evaluated the biocontrol potential of indigenous endophytic and rhizospheric microorganisms isolated from local environments, including desert plants, and vegetable rhizospheres. Several fungal isolates, including *Talaromyces variabilis*, *T. pinophilus*, *Aspergillus terreus*, *Cladosporium omanense* and several *Trichoderma* species exhibited strong antagonistic activity against *P. aphanidermatum* in vitro and significantly improved seedling survival under greenhouse conditions. These fungi inhibited pathogen growth through multiple mechanisms, including production of cellulase and glucanase enzymes, suppression of oospore formation, and induction of morphological abnormalities in pathogen hyphae. Seedling survival rates improved by up to 70% when treated with these biocontrol agents. In addition, the endophytic bacterium *Enterobacter cloacae*, isolated from different hosts, reduced disease incidence significantly and caused structural damage to pathogen hyphae, while maintaining cucumber growth and vigor. Overall, this research demonstrates the promising potential of native microbial isolates as sustainable alternatives to chemical fungicides for managing soilborne diseases. The findings support the development of integrated, environmentally friendly disease management strategies suited to arid agricultural systems in the GCC countries.

Harnessing *Aeromonas* FONT1B for sustainable tomato cultivation: growth promotion and biocontrol under abiotic and biotic stress.

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Global climate change and the intensification of international plant trade have significantly increased crop exposure to abiotic and biotic stressors. Conventional management practices rely heavily on chemical fertilizers and pesticides, which contribute to environmental

degradation, human health risks, and the emergence of resistant pathogen strains. In this context, Plant Growth-Promoting Bacteria (PGPB) offer a sustainable alternative for enhancing crop resilience and productivity. This study evaluated the multifunctional role of the *Aeromonas* strain FONT1B as both a PGPB and a biological control agent (BCA) in tomato plants subjected to drought stress and *P. infestans* infection. FONT1B demonstrated key plant-beneficial traits, including indoleacetic acid (IAA) production, phosphate and potassium solubilization, and tolerance to salinity, drought, and pH fluctuations. Whole-genome sequencing revealed genes involved in stress adaptation, phytohormone biosynthesis, and the production of antimicrobial compounds. *In vivo* assays confirmed the efficacy of this strain in promoting plant growth and reducing disease severity under both abiotic and biotic stress conditions. These findings highlight the potential of FONT1B as a bioinoculant for integrated disease and stress management in sustainable tomato cultivation.

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CONCURRENT SESSION F2 – Postharvest - new control tools

SESSION KEYNOTES

Biological control of post-harvest diseases: from single microorganisms to synthetic microbial communities (SynComs).

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The idea of managing postharvest diseases of fresh fruits and vegetables using beneficial microorganisms dates back to the mid-1980s, stemming from concerns on the risk of using synthetic fungicides on postharvest products. Starting from the early 90s, the expected ban of several synthetic chemicals used in crop protection pushed the interest toward biological control in postharvest products, with hundreds of scientific papers published each year. Many potential biocontrol agents (BCAs) including bacteria, filamentous fungi, and yeasts

were enthusiastically proposed as the one-size-fits-all solution to safely manage postharvest diseases. Due to concerns about the production of antibiotics by bacterial strains, yeasts were considered more acceptable by consumers and were largely the most investigated. After more than 40 years, this huge scientific effort yielded several strains of BCAs with commercial potential, but only few commercial products have been developed, and none of them has seen a widespread commercial use. The reasons for this limited success can be identified in difficulties in managing products based on living organisms, the high registration costs, the restricted range of application of most BCAs (both in terms of target host and pathogens), and, more importantly, the lack of a consistent efficacy. Over the years, it has become increasingly clear that the simplistic model where a single antagonist is used to control a single or multiple pathogens is ineffective. Indeed, this approach does not consider the complexity of the postharvest systems and the interactions between host, its microbiome, pathogens, and the surrounding environment. Recent research is therefore gradually shifting from analyzing individual microbes to examining entire microbial communities, viewing microorganisms as a complex network of interacting organisms playing a critical role in mitigating plant diseases, including postharvest rots of fruits and vegetables. In this study, instead of focusing on individual BCAs, we engineered microbial communities capable of effectively contrast postharvest diseases. Microbial communities from different environmental sources were used to inoculate wounds on apple fruits. Microbial communities were then selected for their ability to quickly colonize wounds without causing disease through 10-cycle successive passaging. Microbial communities from the first and the last cycle were then tested to evaluate their *in vivo* efficacy in preventing the development of the fungal pathogens *Botrytis cinerea* and *Penicillium expansum*. Amplicon sequencing analyses revealed shifts in the structure and diversity of the microbiome across the cycles, resulting in a reduction in disease incidence by 90% (*B. cinerea*) and 70% (*P. expansum*). To further validate the robustness of our engineered communities, we conducted additional tests using three SynComs designed to mimic the microbiome we recovered in wounds after ten re-inoculation cycles. The efficacy of a SynCom proved comparable to that of the sourcing selected microbiomes, thereby reinforcing the validity of our approach for designing functional SynComs and developing sustainable and effective solutions for postharvest disease management.

ORAL PRESENTATIONS

Control efficacy of a new SIGS-based biofungicide against *Penicillium digitatum* on citrus fruits. S. TESTEMPASIS^{1,4}, A. DALAKOURAS², V. KOIDOU², K.K. PAPADOPOULOU³, G.S. KARAOGLANIDIS¹.

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Penicillium digitatum (Pd) is considered as one of the most significant post-harvest pathogens of citrus fruits worldwide. This study aimed to develop a new biofungicide using exogenous application (Spray Induced Gene Silencing, SIGS) to address post-harvest infections caused by the fungus Pd in orange fruits. Specifically, the effectiveness of pathogen control was tested using exogenous applications of dsRNA targeting the silencing of various important genes involving in the RNA silencing mechanisms (*Pd_ds1*, *Pd_ds2*, *Pd_ds3*, *Pd_ds4*) and pathogens development (*Pd_ds5*). The effectiveness of dsRNA applications was tested on artificially inoculated orange fruits (cv. Salustiana) with the pathogen Pd, where the dsRNA was applied at the wound site prior to the infection. The evaluation of effectiveness was conducted by measuring the severity and incidence of the disease in the infected fruits, seven (7) days post-inoculation. Among the dsRNAs tested, the silencing of the *Pd_ds1*, *Pd_ds2*, and *Pd_ds3* genes, significantly reduced ($P < 0.05$) the severity and incidence of disease compared to the control treatment. Specifically, the silencing of genes *Pd_ds1*, *Pd_ds2*, and *Pd_ds3* significantly inhibited the disease development by 84.2%, 89.8%, and 84.2%, respectively. The innovation of this study lies in the use of SIGS technology to develop an environmentally friendly new biofungicide, offering a new approach to addressing post-harvest diseases of citrus fruits.

This research titled “Innovative Solutions for Sustainable and Environmentally Friendly Crop Protection of Greece’s Horticultural Crops in the Europe of the Future” (TAEDR-0535675), was implemented within the framework of the National Recovery and Resilience Plan “Greece 2.0”, with funding from the European Union – NextGenerationEU.

Electrolyzed salt solutions application as sustainable control tool against major postharvest diseases of fresh fruit and vegetables. O. INCERTI, M. ROBBE, M. PASQUALICCHIO, G. CELANO, M. DE ANGELIS, A. IPPOLITO, S.M. SANZANI. *Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, Via Amendola 165/A, 70126 Bari, Italy.* E-MAIL: ornella.incerti@uniba.it; simonamarianna.sanzani@uniba.it

Over the last decades, the postharvest management of fresh fruits and vegetables (FFVs) has been dependent on chemicals. This resulted in the rise of resistant pathogen populations and the formation of unhealthy products, causing a progressive restriction in the allowed active substances. As such, the sustainable management of phytopathogens, microbial contaminants, and FFVs' quality is a major concern for the horticultural industry. The electrolysis of water has gained considerable interest as sanitizer and cleaning agent, due to nonthermal, non-toxic, and broad-spectrum microbial inactivation, as well as for its low cost, portability, and preservation of FFVs quality. In the present study, we tested the electrolysis in presence of different electrolytes, as sanitization process of washing water of a citrus packinghouse, and thus against *Penicillium digitatum*. A consistent reduction of the microbial population of the water was recorded after 1 h of electrolysis in presence of NaHCO₃. Hints on the putative mode of action are also provided. Furthermore, electrolyzed water obtained in the absence (eW) and presence of NaCl and NaHCO₃ (eNaCl and eNaHCO₃) was tested against some of the most relevant postharvest pathogens and diseases of fresh fruit and on a range of commodities. Overall, eNaCl resulted the most efficient treatment for preventing spore germination, as well as for minimizing fruit rots, followed by eW and eNaHCO₃. As such, electrolysed solutions seem promising as dipping treatments for preventing postharvest decay of FFVs.

Project funded under the National Recovery and Resilience Plan (NRRP), Mission 4 Component 2 Investment 1.3 - Call for tender No. 341 of 15 March 2022 of Italian Ministry of University and Research funded by the European Union - NextGenerationEU; Project code PE00000003, Concession Decree No. 1550 of 11 October 2022 adopted by the Italian Ministry of University and Research, CUP D93C22000890001, Project title "ON Foods - Research and innovation network on food and nutrition Sustainability, Safety and Security - Working ON Foods".

Isolation and evaluation of endophytes as biological control agents to control wound and latent apple pathogens. G. REMOLIF¹, V. GUARNACCIA^{1,2}, M.

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Apple fruit diseases, caused by both wound and latent pathogens, cause significant production losses during cold storage, transportation, and marketing. Biological control using microbial antagonists is a promising alternative to synthetic fungicides for disease management. Endophytes are gaining increasing attention as biocontrol agents (BCAs), due to their symbiotic relationship with the host plant, which may enhance their competitiveness and efficacy in disease control. The aim of this work was to isolate endophytes from healthy apples and to evaluate their potential as BCAs against several apple pathogens. A protocol was developed to isolate endophytic yeasts and bacteria from mature apples of various cultivars and geographical origins. A total of 137 strains were obtained. *In vivo* assays were performed to evaluate their potential antagonistic activity, using *Botrytis cinerea* as the target pathogen. The most effective strains were molecularly identified and selected for further analyses. Yeasts belonged to the genera *Aureobasidium*, *Bullera*, *Metschnikowia*, and *Wickerhamomyces*, while bacteria to the genera *Pantoea*, *Pseudomonas*, *Rahnella*, *Orchobactrum*, and *Stenotrophomonas*. The growth of the selected strains at different temperatures was also tested to assess their ability to survive in field and storage conditions. Additionally, their efficacy against latent apple pathogens, including *Phlyctema vagabunda* and two *Colletotrichum* species, was evaluated. Most strains showed a significant reduction in disease severity compared with the inoculated control, showing promising potential as biocontrol agents. The most effective ones will be further studied to elucidate their mechanisms of action and evaluate their potential for field and postharvest applications.

This research was financially supported by Next Generation EU - National Recovery and Resilience Plan (PNRR) - Mission 4, Component 2, Investment 1.4, National Research Centre for Agricultural Technologies - AGRITECH, code CN00000022, CUP D13C22001330005.

Flow cytometry: a rapid and low-cost tool for detection and quantification of feared fungal plant pathogens. E. CHIAROMONTE¹, A. AGNUSDEI¹, E. ALTAMURA², D. GERIN¹, F. MAVELLI², S. POLLASTRO¹,

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The demand for rapid, reproducible and low-cost tools for the detection and quantification of plant pathogens has increased over the last years. Flow cytometry (FCM) allows to rapid provide a multiparametric analysis of single cells in a complex matrix, through two morphological parameters, forward scattering (FCS) and side scattering (SSC) respectively, combined with fluorescence signals. *Aspergillus* spp. and *Botrytis cinerea* are responsible for mold, affecting worldwide several crops both in field and in postharvest. Furthermore, some species belonging to *Aspergillus* genera produce dangerous mycotoxins such as ochratoxin A (OTA). Here, FCM was successfully applied for a rapid identification of *A. carbonarius*, *A. niger* and *B. cinerea* conidia both in pure conidia suspension and in strawberry wash water. Conidia of each species, stained with calcofluor white, were properly characterized based on, both, morphological parameters and fluorescence signals. The availability of an albino mutant of *A. carbonarius* allowed to observe in conidia water suspension the autofluorescence of OTA. The mycotoxin was not detectable in the wild type, probably due to a quenching effect caused by the melanin. In mixed suspensions, conidia of the two *Aspergillus* species were easily distinguishable based on their morphology, which affected light scattering. Even in more complex matrices, such as strawberry wash water spiked with single-species conidial suspensions, detection was still achievable, opening new possibilities for multiplex analysis. These findings confirm the applicability of flow cytometry for the rapid detection of plant pathogens, representing a pioneering step in the agricultural field.

This research was partially financially supported by: Agritech National Research Center and received funding from the European Union Next-Generation EU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) –MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

POSTER PRESENTATIONS

Microbiome dynamics of sooty fungal epiphytes on postharvest apples. F. REY^{1,2}, S. OETTL¹, H. SCHULER². ¹Laimburg Research Centre, Laimburg 6-Pfatten/

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During postharvest storage, certain fungi outcompete other microorganisms on the surface of apples and produce dark, sooty blemishes. These visual imperfections reduce the marketability of the fruit and require fungicide treatments and controlled storage conditions. To assess the dynamics of the epiphytic fungal community of the cultivar Rosy Glow/Pink Lady® during storage, a metabarcoding analysis was performed: apples were harvested from two experimental orchards, both treated in a randomized block design with Merpan® 80 WDG (Captan), Geoxe® (Fludioxonil), or Ulmasud (rock powder, bentonite and lignosulfonates). Sampling occurred at harvest and after six months of storage at 1.5°C and 96% relative humidity. Metabarcoding analysis reveals that alpha diversity (shannon and pielou) significantly decreases from healthy to symptomatic apples. Furthermore, ordination of the Bray-Curtis metric produces distinct clusters, with significant percentage of the variation explained by field, treatment, symptoms and storage. Clusters derive from universal changes from healthy to symptomatic apples, which consist in a reduction of the relative abundance of yeasts, known to have biocontrol properties (e.g. *Kalmanozyma*, *Vishniacozyma*) and an increase in wood decaying fungi (e.g. *Hypoxylon*, *Diaporthe*, *Diplodia*). To identify potential causal agents, fungal thalli from symptomatic peel plugs will be isolated using a needle under a dissecting microscope, inoculated serially on PDA and, when pure, the ITS 3-4 region sequenced. These isolates will be cross-referenced with Illumina data from peel plugs with matching symptoms. This research provides key insights into postharvest microbiome dynamics and contributes to the optimization of epiphyte management during postharvest storage.

A non-invasive approach to targeted lenticel and core inoculation of apple fruit with fungal phytopathogens. S. BHARTI¹, H. QAYYUM¹, S. BARIC^{1,2}. ¹Faculty of Agricultural, Environmental and Food Sciences, Free University of Bozen-Bolzano, Piazza Università 5, 39100 Bozen-Bolzano (BZ), Italy. ²Competence Centre for Plant Health, Free University of Bozen-Bolzano, Piazza Università 5, 39100 Bozen-Bolzano (BZ), Italy. E-MAIL: shbharti@unibz.it

Apple (*Malus × domestica*) is the third most widely cultivated pome fruit tree worldwide. However, postharvest

fungal pathogens can significantly threaten the apple quality during storage. Early detection of latent post-harvest pathogens would greatly aid the sorting process before sending the produce to the market. Hyperspectral imaging has potential for the early detection of pathogens, but to develop accurate diagnostic protocols, it is crucial to use inoculation methods closer to natural infection routes. Nonetheless, injury-based inoculation techniques have been extensively studied; there remains a need for noninvasive, targeted methods of inoculation. In this study, we used *Colletotrichum godetiae* and *C. fioriniae*, both responsible for bitter rot of apple, for targeted lenticel inoculation, and *Fusarium avenaceum* for inducing core rot. Spot plasters soaked with 50 μL of conidial suspension (10^8 spores mL^{-1}) were attached to the fruit of 'Golden Delicious' and 'Gala' at 90-degree intervals across the equatorial plane to induce lenticel rot. Plasters were removed after 14 days post-inoculation (dpi), and the fruits were monitored for visual infection symptoms until 40 dpi. For core rot, 100 μL of spore suspension was injected through the calyx of 'Red Delicious' fruit using needles. Fruits were evaluated at 7, 14, and 21 dpi. The infection rates for targeted lenticel inoculation were 69% for *C. fioriniae* and 60% for *C. godetiae*, while 98% of the fruits showed infection in the core rot inoculation. Further optimization is ongoing to establish a reliable noninvasive protocol for inducing targeted lenticel and core rot infections in apple fruit.

This study is part of the research project HIPPA ("Hyperspectral imaging for the detection of physiologically and parasitically induced damages on apple fruit at harvest and during post-harvest"; CUP I53C23001650007; EFRE1044) that was funded within the EFRE-FESR 2021-2027 Programme of the European Union.

Ligninolytic fungi as biocontrol agents against post-harvest phytopathogens. N. SCHLOSSEROVA^{1,2}, M. ROBBE³, A. MOSTACCI³, O. INCERTI³, A. IPPOLITO³, S.M. SANZANI^{3*}. ¹Department of Chemistry and Biochemistry, Mendel University in Brno, 613 00 Brno, Czech Republic; ²Department of Biosciences, Biotechnologies and Environment, University of Bari Aldo Moro, Via Orabona 4, 70126 Bari, Italy; ³Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, Via Amendola 165/A, 70126 Bari, Italy. E-MAIL: simonamarianna.sanzani@uniba.it

Postharvest losses of fruits and vegetables caused by phytopathogens and inadequate handling remain a significant issue in many countries. Common fungal pathogens such as *Alternaria alternata*, *Botrytis cinerea*,

Fusarium avenaceum are known to lead to rapid fruit decay during both pre- and postharvest stages. Some of those pathogens are known to produce toxic metabolites known as mycotoxins. Control of diseases has relied heavily on the use of fungicides, both during crop cultivation and after harvest. However, in recent years, there has been an increasing interest in developing more environmentally friendly and safer alternatives to extend the shelf-life of fruits and vegetables. In the present investigation, basidiomycetes, belonging to the genera *Ganoderma*, *Laetiporus* and *Fomitopsis*, already used in traditional medicine, were morphologically and molecularly identified and used as biocontrol agents against selected postharvest fungi both as live cells and cultural filtrates on different growth media. The effect was assessed both *in vitro* and *in vivo*. The selected basidiomycetes, free from any detrimental effect on the target hosts, controlled the phytopathogen growth. In particular, the extracellular fraction of the basidiomycete culture proved to be efficient in controlling fungal viability. Moreover, *in vivo*, it reduced the ability of phytopathogens to colonise fruit tissues. Although further large-scale trials are needed results seem promising.

This study was partially carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022).

Mycovirus-based biocontrol potential of post-harvest fungal pathogens in Citrus fruits: current insights and future prospects. H. BENZAHRA^{1,2}, I. MRABTI^{1,2}, H. GRIJJA¹, K. SELMAOUI², M. AFECHTAL¹. ¹Laboratory of Virology, Regional Agricultural Research Center of Kenitra, National Institute of Agricultural Research, 14000 Kenitra, Morocco. ²Faculty of Sciences, Laboratory of Plant, Animal, and Agro-Industry Productions, University Ibn Tofail, Kenitra, Morocco. E-MAIL: mohamedafechtal.inra@gmail.com

Postharvest losses in citrus fruits are largely attributed to fungal pathogens, particularly *Penicillium digitatum* and *P. italicum*, the main causative agents of green and blue mold, respectively. In recent years, mycoviruses have emerged as potential modulators of fungal virulence and promising tools for biocontrol. This review explores the diversity, characteristics, and potential biocontrol applications of mycoviruses infecting *Penicillium* species associated with citrus postharvest diseases. A comprehensive literature review was conducted using databases such as PubMed, Scopus, and Web of Science,

focusing on studies published between 2000 and 2024. We compiled and analyzed reported mycovirus infections in *P. digitatum* and *P. italicum*, assessing virus families, genome characteristics, and their effects on fungal phenotype. Key findings revealed that mycoviruses from families such as *Partitiviridae*, *Chrysoviridae*, *Hypoviridae*, and *Totiviridae* have been identified in *Penicillium* species. Notable examples include *Penicillium digitatum* partitivirus-1 (PdPV-1) and *Penicillium chrysogenum* virus (PcV). Certain mycoviruses, such as those in the *Hypoviridae* family, have been linked to hypovirulence, reduced spore production, and decreased mycotoxin biosynthesis. Both in vitro and in vivo studies suggested that these mycoviruses can significantly attenuate pathogenicity, highlighting their potential as alternative biocontrol agents for managing citrus postharvest decay. While the prospects are promising, the practical application of mycoviruses in integrated postharvest disease management remains limited by gaps in our understanding of host-virus interactions, viral stability, and effective delivery systems. Future advances in RNA sequencing technologies and reverse genetics will be crucial in identifying cryptic infections and developing mycovirus-based strategies as sustainable alternatives to chemical fungicides.

Evaluation of biocontrol agent alternatives for winter management of fungi associated with canker disease in hazelnut. E. MOYA-ELIZONDO¹, J. SAN MARTÍN¹, B. RUIZ¹, K. ROJAS¹, Y. VEGA², M.J. LISPERGUER³. ¹*Departamento de Producción Vegetal, Facultad de Agronomía, Universidad de Concepción, Campus Chillán 3812189, Chile.* ²*Department of Research and Development, Bioprotegens Innovation SpA., Arauco 360, Chillán, Chile.* ³*Departamento Técnico, Frutícola Agrichile S.A., Lote A, Higuera 1, La Florida del Alto, Curicó, Chile.* E-MAIL: emoya@udec.cl

Hazelnut (*Corylus avellana* L.) is the second most important nut tree in Chile, currently occupying 46,000 hectares. Various fungal species cause cankers and branch dieback, necessitating effective management options. In this study, biocontrol agents (BCAs) and BCA-based paints were evaluated to protect pruning cuts in 'Tonda Di Giffoni' hazelnut. In late winter, ten branches per tree were cut with pruning shears and treated on the same day with *Trichoderma atroviride* MUCL 45632 (Ta), a *Pseudomonas protegens* Ca6-based paint (PPp), an organic acid-based paint (POA), and a tebuconazole-chlorothalonil based paint (5 and 50 g L⁻¹ [PFTC]), plus an untreated control (UTC). Five branches were inocu-

lated with 5 mm APDA mycelial plugs of *Diaporthe ambigua* (Da), *Fusarium culmorum* (Fc), and *Diplodia mutila* (Dm). The remaining five branches were treated, covered with aluminium foil, and inoculated 7 days later with the mentioned pathogens. After eight months, branches were collected, the canker length was measured, and fungi from the necrotic tissues were reisolated. Results indicated that Ta and PPp reduced significantly the length of necrosis caused by Da. PPp and PFTC treatments had the lowest re-isolation frequency of fungal pathogens. In branches inoculated 7 days later, treatments did not differ from UTC, except for PPp, which reduced Fc necrosis by 40%. Ta, PPp, and PFTC reduced Fc re-isolation. In both inoculation scenarios, POA significantly increased canker length compared to UTC, but had the lowest re-isolation for all pathogenic fungi. Winter treatments with BCAs could be an alternative for managing canker fungi in hazelnut.

This research was financially supported by the Phytopathology Laboratory of the Facultad de Agronomía at the Universidad de Concepción.

Sweet cherry canker diseases: understanding their ecology, epidemiology and control in Chile. D. GRINBERGS, J. CHILIAN, R. ORREGO, M. ISLA. *Instituto de Investigaciones Agropecuarias INIA Quilamapu, Av. Vicente Méndez 515, Chillán, Chile.* E-MAIL: dgrinbergs@inia.cl

Sweet cherry cultivation has increased in Chile becoming the first exporter worldwide, reaching 413.979 t and USD 2.048MM FOB in 23/24, and turning into the most profitable fruit crop. Fungal diseases are the main causes of limiting productivity and fungal trunk diseases have become, especially in recent years, highly relevant affecting both productivity and longevity of sweet cherry. Determining the etiology and understanding the epidemiology of these diseases are highly relevant to design prevention and control strategies. Symptomatic woody samples (n=830) have been collected since 2019 from the Metropolitana to Los Lagos Regions. Fungi were isolated and pathogenicity tests were performed for representative isolates from most isolated species, in detached twigs and nursery cherry plants var. Lapins. The most isolated and pathogenic genera were *Cytospora* (43%), followed by *Calosphaeria* (20%), *Chondrostereum* (10%), *Nectria* (3%), and *Eutypa* (2%). To study the epidemiology of the most relevant pathogens, four commercial orchards with different climatic conditions were weekly monitored for 24 months collecting the aerial inoculum

on glass spore traps. Inoculum of each pathogen was quantified by qPCR and correlated with temperature and precipitation, which were the most significant factors affecting spore release. Knowing the periods of highest risk, it was possible to design a prevention and control strategy. Chemical (n=15), biological (n=5) and immune response inducers (n=2) treatments were evaluated *in vitro* and in orchard trials, obtaining that all the products had an effect on fungal growth, wood discoloration, canker formation, and number of dead spurs, with benomyl, tebuconazole, chlorothalonil, and two biologicals showing the most significant results.

This research was financially supported by National Agency for Research and Development (ANID), Foundation for Agricultural Innovation (FIA) and O'Higgins Regional Government.

Metabolic profiles of Crisp A and D isolates of *Erwinia amylovora*. G. SOUSA-GOMES¹, Á. QUEIROZ¹, L. MOURA². ¹Instituto Politécnico de Viana do Castelo, Escola Superior Agrária (IPVC/ESA), Refóios, 4990-706 Ponte de Lima, Portugal. ²CISAS - Centre for Research and Development in Agrifood Systems and Sustainability, Instituto Politécnico de Viana do Castelo 4900-367, Viana do Castelo, Portugal. E-MAIL: guilherme.g@ipvc.pt

Fire blight, a disease caused by *Erwinia amylovora*, poses a significant threat to plants in the Rosaceae family, particularly pome fruits. In Portugal, the bacterium is widely spread across the main pear and apple production areas, especially in the Oeste Region. The 'Rocha' pear sector has experienced significant yield losses recently, resulting in substantial economic losses for Portuguese producers. This study aims to isolate and genetically and phenotypically characterize the *Erwinia amylovora* population obtained from symptomatic plant material collected in 2024, from pear and apple orchards in the Oeste Region of Portugal. Ten *Erwinia amylovora* isolates were characterized using the CRISPR-PCR technique and the Gen III Biolog MicroPlates for phenotypic analysis. CRISPR genotyping distinguished the isolates into two genotypes: genotype A and genotype D, depending on the presence or absence of the duplication of spacer 1029, respectively. Phenotypic characterization revealed heterogeneity within the population, regarding carbon source use and chemical sensitivity. The presence of both genotypes A and D, along with the observed diversity in metabolic carbon profiles and chemical sensitivity in the recently isolated *Erwinia amylovora* population from the main Portuguese pear and apple production region, could

suggests different adaptive processes of this pathogen. These processes are possibly driven by the disease control methods applied and its adaptation to environmental and climatic conditions over the time. This study demonstrated that despite this species's previously known genetic homogeneity, there is phenotypic diversity within the *E. amylovora* population in Portugal. This diversity provides a better understanding of the pathogen's epidemiological behaviour under Portuguese production conditions.

This research was financially supported by the project BioFago - New strategies to control Fire Blight (PRR- C05-i03-I-000179) financed by national funds through the Agriculture and Fisheries Financing Institute, Public Institute - I.F.A.P.

Fungal diversity and pathogenicity in olive wood decline: Insights from the North Aegean Region. E.A. MARKAKIS^{1,2}, M.M. MATHIOUDAKIS³, S. SOULTATOS^{1,2}, N. KRASAGAKIS^{1,2}, M. KISSANDRAKI³, A. KARAGIANNI³, G. LAGOUTARIS⁴, E. KOUKOULI⁴, G. KATSIKOIANNIS⁴, S. NTOUGIAS⁵, N. KAVROULAKIS³. ¹Institute of Olive Tree, Subtropical Crops and Viticulture, ELGO-DIMITRA, 71307, Heraklion, Greece. ²School of Agricultural Sciences, Hellenic Mediterranean University, 71004, Heraklion, Greece. ³Institute for Olive Tree, Subtropical Crops and Viticulture, ELGO-DIMITRA, 73134 Chania, Greece. ⁴Directorates of Agricultural Economy (DAOK) of the North Aegean Region. ⁵Department of Environmental Engineering, Democritus University of Thrace, 67100, Xanthi, Greece. E-MAIL: kavroulakis@elgo.gr

An extensive survey was conducted in olive groves across the islands of Lesbos, Chios, Samos, and Ikaria (North Aegean Region, Greece) during 2023–2024 to investigate fungal pathogens associated with wood diseases. Wood samples were collected from symptomatic olive trees exhibiting leaf chlorosis, wilting, scorching, twig and branch dieback, wood discoloration and overall tree decline. Fungal isolations were performed on acidified PDA, and isolates were identified based on morphological characteristics and sequencing of the ITS region and additional genes as needed. Results revealed the presence of several phytopathogenic fungi associated with wood diseases, primarily from the genera *Pseudophaeomoniella*, *Pleurostoma*, *Phaeoacremonium* and *Comoclathris*. Among these, *Pseudophaeomoniella* species appeared to be the most prevalent. Additionally, non-primary pathogens such as *Cladosporium* spp., commonly linked to wood-feeding insects, were also identified. The study is ongoing, with further evaluation of the pathogenicity

of these fungi on the major regional olive cultivars, i.e., Koroneiki, Kolovi, Adramytini and Throumbolia.

The study was funded by the North Aegean Region under the programming contract "Research and information actions to enhance the preparedness of the North Aegean Region regarding the immediate eradication of the pathogen Xylella fastidiosa in case of detection"-XyLeVA.

At the heart of decay: Investigating the ecology and epidemiology of *Phellinus pomaceus* in California prune orchards. L. HOFFMAN. *Department of Plant Pathology, University of California, Davis, One Shields Ave, Davis, CA 95616. E-MAIL: hoffman@ucdavis.edu*

Surveys have confirmed the widespread presence of the wood decay fungus, *Phellinus pomaceus*, in California prune orchards. This pathogen is associated with a significant decline in orchard lifespans. Infection by *P. pomaceus* leads to heart rot decay, associated with the loss of tree scaffolds and the need for tree replacement and more rapid orchard turnover. Despite its increasing prevalence, little is known about the biology, life cycle, and epidemiology of this fungal pathogen—particularly in the Western United States. This study investigated the seasonal development, sporulation dynamics, and spatial distribution of *P. pomaceus* across orchards in California through field surveys, spore trapping, whole-tree dissections, and laboratory culturing. Using monitoring of airborne spores, we are specifically tracking the timing of sporulation events to identify key environmental conditions associated with spore release. Our findings thus far indicate that sporulation is highly seasonal. Entry into healthy prune trees by *P. pomaceus* was thought to be associated with pruning wounds due to the position of fruiting bodies emerging at branch stubs. Further work in this project now supports this hypothesis, highlighting the importance of understanding pruning practices and seasonal timing considerations in disease management.

This research was financially supported by the California Prune Board, Roseville, CA 95661.

***Diaporthe* complex is responsible of *Phomopsis* canker on *Prunus dulcis* in Apulia.** D. SALAMONE, V. MONTILON, G. INCAMPO, D. GERIN, S. POLLASTRO, F. FARETRA. *Department of Soil, Plant and Food Sciences, University of Bari, Via G. Amendola, 165A/ 70126 Bari, Italy. E-MAIL: giuseppe.incampo@uniba.it*

The almond industry in Italy represents a high degree of structural variability where old traditional orchards coexist with new plantations, very different in age and orchard systems. Apulia and Sicily remain the most important production areas where 90% of the almonds are grown profitably under suitable climatic conditions. Self-fertile cultivars, clonal rootstocks and new planting designs optimized for mechanization were the most important technical features revealing a new way to set up almond orchards. Several wood pathogens damage almond tree and frequently bud necrosis and severe twig dieback can be observed. The disease symptoms occur widely and are cultivar-dependent with 'Ferragnes' and 'Guara/Tuono' more being more susceptible than 'Filippo Ceo'. Brown, sunken, elongated, necrotic lesions were observed around the buds and the new shoots usually wilted and died. A putative pathogenic fungus was isolated from the necrotic tissues on potato dextrose agar (PDA) producing pycnidia mainly with one-celled, hyaline, fusoid α -conidia and rarely with β -conidia. For molecular identification, DNA was extracted from the mycelium of the isolates, subjected to PCR using ITS primers and custom-sequenced. The obtained sequences let to ascertain the presence of a complex of *Diaporthe* species with *Diaporthe amygdali* (anamorph: *Phomopsis amygdali*) being the most common. Additionally, a co-culture assay was used to evaluate the intra-species variability in terms of vegetative compatibility and a high intra-species variability, suggesting the need for further detailed study. This work builds the foundation for further studies to help develop appropriate plant-pathogen disease management strategies.

This research was financially supported by Agritech National Research Center and received funding from the European Union Next-Generation EU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) –MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022) and Almond Management Innovations: Approcci per una Mandorlicoltura biologica Innovativa (AMI) PSR 2014-2020 avviso pubblico approvato con D.A.G. n. 194 del 12/09/2018, pubblicata nel B.U.R.P. n. 121 del 20/09/2018. This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

CONCURRENT SESSION F3 – EUPHRESCO III: strengthening the plant health research community

SESSION KEYNOTE

Strengthening plant health in the Mediterranean region through coordinated research and international collaboration. M. D'ONGHIA¹, A. ALAWAMLEH², M. CARA³, L. CRUZ⁴, A. NAJAR⁵, G. BALDISSERA⁶.

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Plant health risks—primarily driven by global trade and climate change—demand a rethinking of current research strategies to enable rapid and effective responses. Strengthening research in this field is a key challenge that countries across the Mediterranean region must confront. However, the wide variation in national priorities—regarding pests, infrastructure, and expertise—has weakened the overall impact of the research efforts. Coordinated action is therefore essential to enhance the efficiency and effectiveness of national research initiatives. The Euphresco self-sustained network provides a platform for cooperation and coordination, aiming to optimise strategies to meet these challenges and ensure a stronger link between research and policy. The network's success has laid the foundation for global discussions on research priorities, as demonstrated in the EUPHRESCO III project. Two main factors are crucial for aligning national research programmes: (i) bottom-up alignment through the involvement of research stakeholders, and (ii) top-down alignment through the engagement of research funding organisations. Building trust and achieving consensus at all levels through regular dialogue and consultation is equally important. In line with these principles, the EUPHRESCO III project has launched consultations with stakeholders active in plant health from diverse regions and disciplines relevant to the network. The main role of regional and disciplinary champions is to create or strengthen mechanisms for

engaging and communicating with their respective communities on plant health research issues. For the Mediterranean region, a dedicated survey was conducted by CIHEAM-Bari—as regional champion—together with the Euphresco Network Office, to identify plant health priorities in the Mediterranean basin. Feedback was collected through a questionnaire distributed to national stakeholders. The analysis focused on key areas such as the geographical distribution of respondents, stakeholder types, priority pests and crops, capacity-building needs, research demands, and critical gaps. This initial assessment provides a valuable foundation for shaping future research and regional cooperation in plant health. The results of national surveys will support the upcoming face-to-face consultation with national and regional decision-makers. This event aims to secure formal commitments on topics such as collaborative research agendas, priorities for transnational cooperation, and institutional engagement (e.g. participation in regional and global networks). Outcomes from four participating countries—Jordan (Near East), Portugal (Southern Europe), Morocco (North Africa), and Albania (Balkans)—will be presented as case studies. Each represents a different sub-region and illustrates the outcome of the national survey within the regional consultation mechanism that have been developed and tested through the EUPHRESCO III project.

POSTER PRESENTATION

CIHEAM Bari - Former Trainees Network (FTN): an institutional tool to evaluate the impact of plant health educational programs. N. DRIQUECH, N. AL GNDI, O. ANTONELLI. *International Centre for Advanced Mediterranean Agronomic Studies (CIHEAM Bari), Via Ceglie, 9- 70010, Valenzano (BA), Italy.* E-MAIL: driouech@iamb.it

Since 1962, CIHEAM Bari has been a leader in advanced agricultural education and training for young agronomists and experts from the Mediterranean region. In 1985, it performed a Master's program curricula in Integrated Pest Management, contributing significantly to the development of a professional community in plant health. The FTN was therefore established for strengthening such a community and linking to several CIHEAM Bari activities. To support this professional network, the CIHEAM Bari Alumni Follow-up Research Study was initiated in 2010. Conducted every five years using Ex-Ante and Ex-Post methods, the study evaluates the long-term impact of CIHEAM Bari's educational

programs while updating its alumni database. The latest study surveyed 1300 alumni from 1999 to 2021 using an online, self-administered questionnaire (available in English and French), with 366 IPM graduates answers. Respondents were predominantly male (66%) and originated mainly from the Mediterranean, followed by the Balkans, Central Africa, and the Near and Middle East. Approximately 34% pursued PhD studies, often in collaboration with Italian universities and CIHEAM Bari. Job placement data show 62% work in the public sector, 20% in the private sector, and 18% in semi-public or international institutions. Alumni hold positions such as researchers (38%), academics (14%), project managers (10%), and consultants (5%). Notably, 82% of respondents reported continued collaboration with CIHEAM Bari through joint research, scientific publications, and technical and Institutional partnerships. These findings highlight the effectiveness of CIHEAM Bari's education/training programs in advancing and promoting sustainable agriculture and regional cooperation, particularly within countries engaged in the CIHEAM Bari education mission and within FTN Network.

CONCURRENT SESSION G1 – Advances in understanding and managing canker diseases of fruit, nut, and forest trees

SESSION KEYNOTES

***Neoscytalidium dimidiatum*, a last century's fungal tree pathogen has become this century's menace to fruit and nut tree crops in California.** T. MICHAILEDIS¹, G. MAKRIS¹, V. GABRI¹, G. GUSELLA². ¹University of California, Department of Plant Pathology, Kearney Agricultural Research and Extension Center, Parlier, CA 93648, United States of America. ²University of Catania, Department of Agriculture, Food & Environment, Plant Pathology, Via Santa Sofia 100, 95123 Catania, Italy. E-MAIL: tjmichailides@ucanr.edu

The tree pathogen known as *Hendersonula toruloidea* or *Nattrassia mangiferae* and now as *Neoscytalidium dimidiatum* has caused major diseases on fruit and nut crops in California in the last couple of decades. Walnut trees have had a chronic, but sporadic, branch wilt caused by *H. toruloidea* (reported in 1947) and the same pathogen along with *Botryosphaeria dothidea* were reported causing band canker of almond back in 1974. Similarly, a branch wilt and killing initially reported in 1964 on old fig orchards in central San Joaquin Valley (SJV), was again caused an epidemic on figs about two

decades ago. The disease killed major branches and limbs of fig trees, resulting in significant yield reductions. Infection courts included wounds created by mallets used during harvest, sunburn tissues, and pruning wounds. The same pathogen was also found in causing severe hull rot of almond from infection by airborne inoculum coming off neighboring walnut and/or fig orchards where the pathogen had killed major tree branches. In one case, an old walnut orchard was removed, and the branches were ground to make the field available to a construction company. To the south of this orchard, there was a Nonpareil almond orchard at the hull split-stage, which is a susceptible stage to infection by hull rot fungi, *Rhizopus stolonifer*, *Monilinia fructicola*, and *Aspergillus niger*. However, in this case, *N. dimidiatum* caused high levels of hull rot, long cankers internally and unusually large, black gum galls on the surface of infected branches. In a second case, a grower shredded the prunings of his old fig orchard, severely infected by *Neoscytalidium* canker, which resulted in a severe case of hull rot on his nearby almonds. In another case, a severe band canker of young almond trees developed in Kern County in 2022. It is known that up to eight different species in the *Botryosphaeriaceae* fungal family can cause band canker of almond; however, *N. dimidiatum* was the solo pathogen isolated from the band canker of these almond trees in Kern Co., CA. In 2013, a nursery producing walnut trees lost approximately 5,000 trees due to infection at the graft union by *N. dimidiatum* (90%) and *Lasiodiplodia citricola* (10%). In 2017, orchards of 'Star Ruby' grapefruit suffered a major limb killing caused by *N. dimidiatum* at the UC Lindcove Research and Extension Center in Exeter, CA. Following two major heat waves in August 2024, a severe epidemic of a branch killing (citrus gummosis) disease appeared in a plethora of Star Ruby grapefruit orchards in central and southern SJV. A total of 15 orchards surveyed and the recovery of the pathogen in samples ranged from 90-100% in 'Star Ruby' grapefruit, 86% in a lemon, and 86% in each of two Pomelo orchards. In several of these orchards, the canker entered the trunk, resulting in tree death. The dry bark of infected branches and trunks was naturally peeled and revealed underneath a black, powdery mass of arthrospores of *N. dimidiatum*. Disease management methods will be discussed.

This research was financially supported by the California Walnut Board, the California Fig Institute, the Almond Board of California, and the Citrus Research Board.

Fungal Trunk Diseases of fruit crops in the Mediterranean Basin: advancing knowledge to tackle a huge diversity of pathogens. V. GUARNACCIA^{1,2}. ¹*Department of Agricultural, Forest and Food Sciences (DISAFA), University of Turin, Largo Braccini 2, 10095 Grugliasco (TO), Italy.* ²*Interdepartmental Centre for Innovation in the Agro-Environmental Sector AGROINNOVA, University of Turin, Largo Braccini 2, 10095 Grugliasco (TO), Italy.* E-MAIL: vladimiro.guarnaccia@unito.it

Fungal trunk diseases (FTDs) are increasingly recognized as a major threat to fruit and nut crop production across the Mediterranean Basin. These diseases, caused primarily by Ascomycota fungi, compromise tree health and yield, reduce orchard longevity, and challenge sustainable agriculture. The complexity and impact of FTDs are emphasized by a wide diversity of fungal taxa involved, including species in the families *Botryosphaeriaceae*, *Cytosporaceae*, *Diaporthaceae*, *Diatrypidae*, *Nectriaceae*, *Togniniaceae*, and others. Those species often co-occur, showing latent pathogenic behavior, and exhibit wide host ranges, contributing to diagnostic and management challenges. A comprehensive overview of the diversity of FTD-associated fungal species reported on fruit and nut crops such as apple, pear, pistachio, hazelnut, citrus and avocado in Mediterranean countries is provided. Recent advances in fungal taxonomy were highlighted, which, supported by multi-locus phylogenetic analyses, allowed the identification of numerous novel or cryptic species, as well as better delineation of known taxa. For example, genera like *Neofusicoccum*, *Lasiodiplodia*, *Diaporthe*, and *Cytospora* have been frequently associated with dieback, canker, gummosis, and wood decay symptoms across multiple hosts. In detail, case studies from Italy, Spain, Greece, Turkey, and North African countries demonstrate the regional variation in species prevalence and disease symptoms, as well as the role of environmental and agronomic factors, such as climate, cultivar susceptibility, orchard age, and stress conditions, in disease outbreaks. Molecular detection tools, including real-time PCR, are discussed as critical methods for early diagnosis and epidemiological studies. Moreover, imaging-based and hyperspectral imaging are considered an innovation. Furthermore, the current limitations in curative treatments and regulatory restrictions on synthetic fungicides highlight the urgent need for integrated and innovative strategies. Thus, emerging perspectives on disease management, including the role of cultural practices, and biological control agents, are explored. Significant progress has been made in unraveling the complex etiology and diversity of fungal trunk pathogens in Mediterranean orchards, the road ahead

calls for stronger interdisciplinary collaboration. A synergistic approach that combines mycology, genomics, climatology, and sustainable agronomy is essential to design effective monitoring systems and deploy targeted interventions.

ORAL PRESENTATIONS

Hazelnut trunk diseases, ecology clarification and development of biological management strategies. V. PIATTINO¹, I. MARTINO¹, M. MASPERO², T. DE GREGORIO², D. SPADARO^{1,3}, V. GUARNACCIA^{1,3}. ¹*Department of Agricultural, Forest and Food Sciences (DISAFA), University of Turin, Largo Braccini 2, 10095 Grugliasco (TO), Italy;* ²*Hazelnut Company division, Ferrero Group, 16 Rue de Trèves – L – 2633 Senningerberg, Luxembourg;* ³*Interdepartmental Centre for Innovation in the Agro-Environmental Sector, AGROINNOVA, University of Turin, Largo Braccini 2, 10095 Grugliasco (TO), Italy.* E-MAIL: valeria.piattino@unito.it

Trunk diseases, a major threat for hazelnut, are characterized by internal necrotic lesions, cankers and production of reddish spore masses on the bark. Highly spreading in the main hazelnut growing areas, these diseases cause plant death and yield losses. Historically, the main trunk disease has been associated with *Cytospora* spp. such as *Cytospora corylicola*. However, a polyphasic approach conducted on fungal strains obtained from Italian and Spanish hazelnut plants, identified *Anthostoma decipiens* as the predominant causal agent, with reddish conidial masses serving as the main inoculum, potentially influenced by climate change. Due to the limited number of available active ingredients for hazelnut crop in Italy, the development of innovative management strategies is needed. Thus, effectiveness of commercial biological products was evaluated on detached twigs and potted plants. The twigs and plants were wounded with a scalpel, treated twice at 5-day intervals, inoculated with mycelial plugs of *A. decipiens*, and the twigs incubated in a moist chamber at 25°C with a 12 h photoperiod for 20 days, while the plants placed in a greenhouse for 4 months. Disease severity was assessed by measuring the length of the internal necrotic lesion. Data were analyzed using ANOVA followed by Tukey's post-hoc test (p -value > 0.05). The effectiveness of commercial biological products containing *Bacillus* spp., *Trichoderma* spp. and *Pythium oligandrum* was statistically comparable to that of chemical fungicides used as a positive control, revealing a reduction in lesion length between 30% and 55%.

This research was financially supported by the Hazelnut Company division of Ferrero.

Effect of wood age on the canker development in almond through multi-inoculations with *Botryosphaeria dothidea* and *Neofusicoccum parvum*. C. LUQUE-CRUZ¹, B.I. ANTÓN-DOMÍNGUEZ¹, L. ROMERO-CUADRADO², N. CAPOTE², C. AGUSTÍ-BRISACH¹.

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Botryosphaeria dothidea and *Neofusicoccum parvum* are the most frequent and aggressive fungal species associated with young almond decline syndrome in southern Spain. In this study, the effect of age of almond wood on canker development was evaluated by inoculating periodically these pathogens. For this purpose, green (1-year-old) or lignified (2-3 years-old) detached shoots and potted almond plants ('Avijor' grafted on GF-677) were inoculated with conidial suspensions using the following treatments: single inoculations of each pathogen, simultaneous co-inoculation, or sequential co-inoculations with a 4-day interval between pathogens. Canker developed in lignified wood were significantly smaller than those in green wood, regardless of the inoculation treatment. There were not significant differences in aggressiveness among treatments in lignified wood. In green wood, *B. dothidea* and *N. parvum* showed significantly higher aggressiveness in single inoculations, or when *N. parvum* was inoculated first, followed by *B. dothidea* 4 days later. Due to the higher aggressiveness observed in green tissues, the inoculum density of each fungus was quantified by multiplex qPCR. The highest relative inoculum density was found in plants inoculated only with *B. dothidea*, in plants co-inoculated simultaneously with both pathogens, or in those where *B. dothidea* was inoculated 4 days before *N. parvum*. In these treatments, the highest amount of DNA corresponded to *B. dothidea*. Despite the inoculum levels recorded, these two co-inoculation treatments resulted in the smallest lesions. These findings suggest that interactions during multi-infections alter the relative aggressiveness of these pathogens.

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Genetic structure of the *Cryphonectria parasitica* population in the Lake Garda area of San Zeno di Montagna (Northern Italy). D.H. TABET¹, A. ESPOSITO², A. BRIGHENTI³, S. BARIC^{1,4}. ¹Faculty of Agricultural, Environmental and Food Sciences, Free University of Bozen-Bolzano, Piazza Università 5, 39100 Bozen-Bolzano (BZ), Italy. ²Department of Biotechnology, University of Verona, Strada le Grazie 15, 37134 Verona (VR), Italy. ³Consorzio di Tutela del Marrone di San Zeno DOP, Via Ca' Montagna 11, 37010 San Zeno di Montagna (VR), Italy. ⁴Competence Centre for Plant Health, Free University of Bozen-Bolzano, Università 5, 39100, Bozen-Bolzano (BZ), Italy. E-MAIL: DaniaHanna.Tabet@unibz.it

The sweet chestnut (*Castanea sativa*) faces threats from multiple diseases, one of which is the chestnut blight caused by *Cryphonectria parasitica*. Disease attenuation in several European regions has been attributed to the presence of hypovirulent fungal strains infected by the mycovirus *Cryphonectria hypovirus* 1 (CHV-1). Hypovirulence is transmitted horizontally via hyphal anastomosis between strains of the same vegetative compatibility (vc) types, and vertically through asexual conidia. Given the importance of the effective hypovirus transmission, a comprehensive understanding of the population structure of *C. parasitica* is crucial for successful biological control of chestnut blight. This study investigated the genetic structure of *C. parasitica* isolated from 89 bark canker samples collected in San Zeno di Montagna (Province of Verona, northern Italy). DNA-based methods were employed to analyze the isolates for the variability at six vegetative compatibility (vic) loci, the mating type locus and eleven microsatellite markers, as well as the internal transcribed spacer (ITS) region. Additionally, RNA isolation, real-time RT-PCR and Sanger sequencing of the ORF-A region were conducted to investigate the presence and diversity of CHV-1. Among 91 *C. parasitica* isolates, 12 distinct vc types were identified. Both mating types were found with nearly equal ratios, and two different ITS haplotypes were revealed. The presence of CHV-1 was determined in 35.2% of isolates and sequence analysis pointed to a high degree of genetic variation. Consequently, the high genetic population variability of *C. parasitica* coupled with sexual

reproduction and the low frequency of CHV-1 may hinder the effectiveness of biological control strategies in this region.

This study was performed within the project “Esplorazione del microbioma del cancro corticale del castagno per migliorare il controllo biologico della malattia in Veneto” (Meta-Blight) funded by Fondazione Cariverona. We are grateful to the Consorzio di tutela del Marrone di San Zeno D.O.P. for the collaboration in this project.

Fungal canker pathogens and pruning wound management strategies in California apple orchards.

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Apple orchards in California—including San Joaquin, Santa Cruz, and Sonoma counties—have shown increasing incidences of branch dieback and wood canker disease, particularly in cultivars such as ‘Fuji’, ‘Gala’, and ‘Granny Smith’. A 2021–2023 survey across nine commercial orchards revealed *Diplodia* spp. (including *D. seriata*, *D. mutila*, and *D. bulgarica*) as the most prevalent pathogens, followed by *Eutypa lata*. Additional fungal taxa included *Diaporthe* spp. (*Di. australafricana*, *Di. eres*, *Di. chamaeropsis*, *Di. foeniculina*), *Cytospora parasitica*, *Kalmusia variispora*, and *Phaeocremonium* sp. All isolates were pathogenic on wounded apple wood in field trials, confirming their role in the canker complex. In 2024, dormant-season field trials were conducted in Sebastopol, CA to evaluate the efficacy of pruning wound protectants. Treatments included thiophanate-methyl, fluopyram + tebuconazole, *Trichoderma asperellum* + *Trichoderma gamsii*, *Aureobasidium pullulans*, and thyme oil. Branches were treated and then inoculated with *D. seriata*. Thiophanate-methyl showed complete control (0% mean percent infection, MPI), followed by fluopyram + tebuconazole (54.2% MPI). The biological products offered limited protection, with *Trichoderma*-based and *Aureobasidium*-based treatments yielding 75–91.7% MPI, and thyme oil showing the least efficacy (95.8% MPI). These results emphasize the dominance of *Diplodia* spp. in apple canker etiology in California and demonstrate the critical need for effective pruning wound protection. Synthetic fungicides remain the most reliable in protecting pruning wounds, though biologicals may play a role in integrated disease management.

Investigation of symptomatic and genetic diversity of *Xanthomonas arboricola* pv. *Juglandis* isolates in walnut orchards and biological control possibilities with bacterial antagonists. D. ERTIMURTAŞ¹, H. ÖZAKTAN². ¹Directorate of Plant Protection Research Institute Bornova, Department of Biological Control, 35040, Izmir, Türkiye. ²Ege University, Faculty of Agriculture, Department of Plant Protection, 35040, Izmir, Türkiye. E-MAIL: ertimurtasdamla@gmail.com

Walnut can be affected by walnut bacterial blight (WBB) and brown apical necrosis (BAN), which are mainly caused by *Xanthomonas arboricola* pv. *juglandis* (*Xaj*). Depending on the prevalence of these emerging diseases, an increase in yield losses is observed in the last quarter century. In this research, surveys were carried out in Western Anatolia during 2018–2019. Thirty-one bacterial strains were identified as *Xaj* by classical methods and 20 of them were confirmed to be pathogenic on immature walnut fruits. In addition, molecular identification by 16S rRNA showed a 99.83 to 100% similarity among the different *Xaj* isolates. Genetic variability among 31 *Xaj* isolates associated with WBB and BAN was best revealed by ERIC-PCR, where our set of bacteria was divided into 9 subgroups. It was found that *Xaj* strains obtained from same symptom, neighbouring regions and with similar pathogenicity were in the same group. The phylogenetic tree obtained by MLSA (Multi Locus Sequence Analysis) using 7 housekeeping genes highlighted differences due to changes in geographical variation and disease symptoms. While *Xaj* strains from WBB were categorised in 4 groups, 2 of 3 *Xaj* from BAN were in the same group. In biocontrol studies, 585 antagonist candidate bacterial isolates were obtained from healthy walnut trees. Out of three bacterial isolates showed biocontrol potential in *in vitro* and *in vivo* conditions. The most successful bacterial antagonists were identified as *Bacillus velezensis* and *Erwinia billingiae* as a result of sequence analysis.

POSTER PRESENTATIONS

Investigating the etiology, cultivar susceptibility, and fungicide efficacy against twig canker disease caused by *Diaporthe amygdali* in peach orchards of Greece. D. PAPADIMITRIOU¹, S. TESTEMPASIS^{1,2}, G.S. KARAOGLANIDIS¹. ¹Laboratory of Plant Pathology, Faculty of Agriculture, Forestry and Natural Environment, Aristotle University of Thessaloniki, 54124 Thessaloniki, Greece; ²Department of Agriculture, School of Agricultural

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Twig canker, caused by the fungus *Diaporthe amygdali*, is a significant disease affecting almond and peach orchards, especially in Mediterranean regions of Europe. The disease targets annually emerging shoots, including fruit-bearing ones, leading to substantial yield losses. Characteristic symptoms include shoot blight, rapid drying, and gummosis of infected twigs. Despite its importance, *D. amygdali* has been insufficiently studied in Greece, so the aim of this study was: (a) the molecular identification, at species level, of twig isolates mainly of peach trees affected by fungal cankers, b) the determination of the susceptibility of 23 peach cultivars at field level along with the investigation of the comparative susceptibility between clingstone and freestone cultivars and c) the investigation of the inhibition of the fungus in GF 667 rootstocks, using 20 different commercial fungicides and a potential biological control agent. Molecular analysis of 100 isolates, using nucleotide sequencing and phylogenetic analysis of the β -tubulin and calmodulin gene regions, identified *D. amygdali* as the predominant pathogen. The highest disease incidence and severity were observed in cultivars 'Papayanni', 'Fortuna', and 'VLG', while 'Early May', 'Cal-2000', and 'Ferlot' showed the lowest severity. Regarding fungicides, fluopyram, boscalid, and copper hydroxide showed the lowest efficacy, while *Aureobasidium pullulans* (biological control) and tebuconazole demonstrated high effectiveness. The fungicide dithianon was the most effective, causing almost no necrosis. This study provided valuable insights into the etiology of twig canker, cultivar susceptibility, and effective disease management strategies, highlighting key factors in controlling *D. amygdali* in peach orchards.

Characterization of new fungal species causing pomegranate decline in Italy. S. TUNDO^{1,2}, D. GERIN³, A. BOLZONELLO¹, R. CARACCILO¹, L. SELLA¹, F. FARETRA³, F. FAVARON¹, S. POLLASTRO³. ¹Dipartimento di Territorio e Sistemi Agro-Forestali (TESAF), Università degli Studi di Padova, Viale dell'Università 16, Legnaro, Italy; ²Dipartimento di Agronomia, Animali, Alimenti, Risorse naturali e Ambiente (DAFNAE), Università degli Studi di Padova, Viale dell'Università 16, Legnaro, Italy; ³Dipartimento di Scienze del Suolo della Pianta e degli Alimenti - Università degli Studi di Bari Aldo Moro, via Amendola 126/A Bari, Italy. E-MAIL: donato.gerin@uniba.it; angela.bolzonello@unipd.it

Pomegranate is an ancient fruit crop, with origins tracing back to at least 4,000 years ago in central Asia. The global expansion of pomegranate cultivation is due to its adaptability to adverse environmental conditions and the increasing demand for foods with improved organoleptic characteristics. With this study, we report the occurrence of pomegranate fungal pathogens isolated from canker/dieback of trees located in Basilicata, Apulia and Veneto regions (Italy). The isolates were identified based on BLASTn analysis of ITS1-5.8S-ITS2 rDNA internal transcribed spacer (ITS) region, translation elongation factor 1- α (*TEF1*), calmodulin (*cmdA*), β -tubulin (*TUB2*), and large subunit (*LSU*) of rDNA as *Diaporthe eres*, *D. foeniculina*, *Neopestalotiopsis rosae*, *Ophiostoma stenoceras* and *Xenoacremonium* sp. Pathogenicity of these fungi was demonstrated by artificial inoculations of 2-years-old pomegranate trees cv. Wonderful. *Coniella granati* and *Neofusicoccum parvum*, fungal species already described for their involvement on pomegranate decline, were also isolated. The colony growth of each isolate was assessed at different temperatures (from 5 to 35°C) on malt extract agar in dark, and for all species the optimum temperature was in the range 25–30°C. All isolates were grown in self- and cross-pairing with the other pathogens and the most important interactions were represented by the competition due to the different growth rates of the isolates. In some cases (e.g. *D. foeniculina*/*O. stenoceras*), a mutual intermingling between hyphae was observed and both pathogens were re-isolated from plugs collected in the contact zone. This work provides knowledge on new pomegranate pathogens, representing a threat for the cultivation of pomegranate.

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Fungal pathogen surveillance in almond orchards: A comprehensive approach through the ATENA project. M.I. DRAIS, C. MACOR, D.M. PINTO, F. PAPA, L. ROSSINI, M. CONTARINI, A. MAZZAGLIA. Department of Agriculture and Forest Sciences, University of Tuscia, Viterbo, Italy. E-MAIL: drais@unitus.it

The (ATENA-Agricoltura di precisione e Intelligenza Artificiale per l'innovazione sostenibile dell'arboricoltura) project supports the sustainable intensification of almond production by integrating advanced technologies and data-driven approaches into the design and management of intensive and super-intensive orchard systems. Central to this initiative is the Decision Support Design Platform (DSDP), a low-code, user-oriented framework that provides agronomists and growers with tools to optimize productivity, resource efficiency, and disease control. A key module within the DSDP is dedicated to phytopathological surveillance, focusing on the detection and identification of fungal pathogens affecting almond orchards. The monitoring system integrates weekly field inspections, symptomatic tissue sampling, and spore trapping for early detection of airborne propagules. Laboratory diagnostics follow a multi-step protocol, including morphological identification based on mycelial and conidial features, molecular analysis using conserved markers (e.g., ITS, β -tubulin), and germination trials to assess spore viability and pathogenic potential. Initial results confirmed the presence of *Monilinia laxa* and *Diaporthe* spp. as major pathogens in the surveyed orchards. The integration of this surveillance data into the DSDP enhances early warning capabilities, supports site-specific phytosanitary measures, and enables adaptive management by aligning control strategies with real-time and historical field conditions. This comprehensive, integrated approach strengthens disease prevention and contributes to the ATENA project's broader goal of promoting resilient, efficient, and environmentally sustainable almond cultivation systems.

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Unveiling *Diaporthe* spp. associated with almond canker in central Italy. M.I. DRAIS, I. GIUBILEI, C. MACOR, A. MAZZAGLIA. *Department of Agriculture and Forest Sciences, University of Tuscia, Viterbo, Italy.* E-MAIL: drais@unitus.it

The incidence of almond-associated fungal diseases, particularly twig cankers and shoot blight caused by *Diaporthe* spp. is increasing and poses a significant threat to crop productivity, especially in areas with high humidity and mild temperatures. Among these, constriction canker caused by *Diaporthe amygdali* is considered one of the most severe diseases affecting *Prunus dulcis* (almond) across Europe, particularly in Mediterranean

countries. Field surveys conducted in Lazio Region (Italy) revealed widespread symptoms of twig canker and shoot blight in almond orchards managed under intensive cultivation systems. The highest disease incidence was recorded in orchards planted with the 'Tuono' cultivar. A total of 25 *Diaporthe*-like isolates were obtained through isolation in the laboratory. Morphological characterization and molecular identification based on sequencing and phylogenetic analyses of the ITS, *tef1- α* , and *tub2* gene regions were used to identify the isolates. The effect of various temperatures (5, 10, 15, 20, 25, 30, and 35°C) on mycelial growth was assessed on PDA medium, while lesion development was evaluated on inoculated detached almond shoots. Pathogenicity tests on detached almond shoots are ongoing to assess differences in aggressiveness among isolates. This study provides an updated assessment of *Diaporthe*-induced cankers in Central Italy, offering integrated morphological and molecular insights into the identity and behavior of these pathogens.

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CONCURRENT SESSION G2 – Innovation in alternatives to synthetic fungicides

SESSION KEYNOTES

Use of basic substances as alternative to synthetic pesticides for the management of plant diseases. G. ROMANAZZI. *Department of Agricultural, Food and Environmental Sciences, Marche Polytechnic University, Via Brecce Bianche, 60131, Ancona, Italy.* E-MAIL: g.romanazzi@univpm.it

Plant pathogens can induce up to 40% of loss and waste of the potential production of fresh fruit and vegetables. Nowadays the application of synthetic fungicides is the main strategy to prevent and manage plant diseases on fresh produces. At the same time, the consumers request products free from fungicide residues, and they are concerned about their use in the field to protect plant health, often forgetting the drugs they use for human or pet health. Retailers use (and at times induce) this fear, to promote more expensive products with zero pesticide residues or with concentrations way lower than allowed maximum residue levels (e.g. even 33% of MRL), and a cap number of active ingredient (in some cases even 3 to 5). Registration of a synthetic pesticide nowadays has

a cost of around 300 million Euro, and it can take up to 13 years. The unfavourable environment toward the synthetic pesticides leads to reduction of the research to discover new active ingredients, while a high number of compounds is disappearing from the market for banning or for expiring or registration (that for a standard pesticide lasts 10 years, while for a low risk fungicide last 15 years). While in 2014 they were registered in EU 30 new compounds and more than half were synthetic products, since 2019 no new synthetic products were registered, and overall yearly registrations were much less than 10. The risk is then to have reduced solutions for plant protection, together with the introduction of new pathogens from other areas (so called aliens) and with the increase of virulence of previously mild pathogens, that were contained by the application of wide spectrum compounds, and/or are taking advantage by the evolution of climate conditions. Therefore, the adoption of alternatives to synthetic fungicides, like biocontrol agents and natural compounds, is mandatory. Among those, the basic substances represent compounds that are already used as a food or feed and can have a secondary activity in plant protection. Then, it is not needed to test their side effects on human and environment, so registration cheaper and can be obtained in a shorter time (around 1-2 years and 50000 Euro, including registration for organic agriculture), and last forever. There are currently 28 basic substances registered for use toward plant diseases, including chitosan hydrochloride, chitosan, vinegar, lecithin, *Equisetum arvense* extract, *Urtica* extracts, *Salix* spp. cortex, magnesium hydroxide, cow milk, whey (liquid or powder), talc and sodium bicarbonate. Some of these basic substances were used in open field and were able to contribute to plant disease management, with example of partial or even total replacement of synthetic fungicides. So, new biosolutions with a reduced impact can, at least partially, replace the synthetic fungicides not anymore on the market, although they require a deeper knowledge by technicians supporting growers to optimise the application and get a protection level similar to what achieved with the synthetic fungicides, sometimes with a higher number of applications.

ORAL PRESENTATIONS

Phytotoxins: a virulence factor or an opportunity? A. EVIDENTE. *Institute of Biomolecular Chemistry, National Research Council (CNR) Viale Campi Flegrei 34, 80078 Naples Italy.* E-MAIL: evidente@unina.it

Nature is an almost inexhaustible source of a myriad of compounds with different chemical structure and biological activities. Fungal phytotoxins, that play an important role in many plant diseases induced by fungal pathogens, belong to different classes of natural compounds. They frequently have original chemical structures and diversified biological activities. Thus, could be a fundamental tool to protect crops from pathogens, avoiding the use of chemical pesticides. Phytotoxins could be used: (a) to develop rapid and specific diagnostic methods in order to detect diseases and be the basis for developing simple kits to be used directly in the field by farmers (b) to help in selecting plants naturally resistant from those susceptible to a pathogen by knowing the genes responsible of innate resistance; furthermore they could help in different research fields: (c) formulating new biopesticides (fungicides, bactericides, but also herbicides and insecticides, etc.) (d) developing new drugs with important applications in medicine due to their activity and specificity frequently associated with a new mechanism of action (anticancer, antiviral, antimalarial, etc.). Significant examples of this kind of bioactive metabolites are ophiobolin A and sphaeropsidin A, two fungal terpenes active as wilt inducing phytotoxins now also developing as promising anticancer metabolites.

TRV-assisted RNA silencing of *VdSNF1* and *VdNLP1* potentiates the development of innovative management strategies against *Verticillium dahliae* in herbaceous crops. C. TSOUKAS, P. SARIDIS, A. VENIERAKI, E.J. PAPLOMATAS. *Laboratory of Plant Pathology, Department of Crop Science, Agricultural University of Athens, Iera Odos 75, 11855, Athens, Greece.* E-MAIL: epaplom@aua.gr

Verticillium dahliae, a devastating vascular pathogen, affects a wide range of economically important crops. Limited effective control methods are currently available. The advancement of RNA interference (RNAi) comprises promising strategy to manage this pathogen. The aim of this study was to utilize a TRV-based host-induced gene silencing (HIGS) approach to suppress pathogen's critical genes. For this, genes involved in several functions such as regulation of expression of cell wall-degrading enzymes (*SNF1*), and induction of necrosis and ethylene production (*NLP* genes) were selected. Initially, specific gene segments were chosen to avoid off-target effects, ensuring a pathogen-specific outcome. Each segment was cloned into the TRV2 vector and *Agrobacterium tumefaciens* AGL-1 cells were subsequently transformed with the resulting constructs. Afterwards, cotton and tomato

plants were agro-infiltrated and artificially inoculated with *V. dahliae*. The effect of TRV-assisted HIGS was evaluated against a highly defoliating (D) and a non-defoliating (ND) isolate in cotton plants, while it was also used against Race 1 (R1) and Race 2 (R2) strains in tomato. Silencing of the *VdSNF1* gene reduced the severity in the ND strain by approximately 50%, while plants expressing the partial *VdNLP1* gene demonstrated 51% less disease upon inoculation with the D strain. Similarly, at 20 dpi of the ongoing experiment, silencing of the *VdSNF1* gene resulted in approximately 40% and 56% less disease against the R1 and the R2 isolates, respectively. These findings suggest that RNA silencing is a promising approach that could lead to the development of innovative control strategies against vascular wilt pathogens.

This project is carried out within the framework of the National Recovery and Resilience Plan Greece 2.0, funded by the European Union – NextGenerationEU (Implementation body: HFRI).

Exploring plant-based protein hydrolysates as resistance inducers in wheat. A. MOSTACCI¹, C. SCAVARDA², G.L. SALA³, L. DE FERRA⁴, O. INCERTI¹, N. DE DIEGO⁵, L. SPÍCHAL⁵, A. IPPOLITO¹, S.M. SANZANI¹. ¹Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, via Amendola 165/A, 70126 Bari, Italy. ²A. Costantino & C. SpA, Via Francesco Romana, 11, 10083 Favria (TO), Italy. ³Biofer S.p.A., Via Gabrio Serbelloni 7, 20122 Milan, Italy. ⁴Sts Pharma Srl, Via Gabrio Serbelloni 7, 20122 Milan, Italy; ⁵Czech Advanced Technologies and Research Institute, Šlechtitelů 27, 77900 Olomouc, Czech Republic. E-MAIL: a.mostacci@phd.uniba.it; simonamarianna.sanzani@uniba.it

By 2030, as part of the Green Deal Farm to Fork Strategy, the European Commission intends to reach the goal of a 50% reduction in the overall chemical pesticide use. An alternative approach to meet the objective is to empower plant natural defense responses using biostimulants, which can positively act on plant biological processes including the defense against stresses. In the present study, a formulation (AgricostanD) based on a protein hydrolysate (PH) derived from soybean flour, was evaluated *in vivo* as a resistance inducer against *Fusarium culmorum*, a causal agent of fusarium head blight on durum wheat in southern Italy (cv. Svevo). Seeds, showing infection symptoms, were subjected to an additional artificial inoculation with *F. culmorum*, thus undergoing a double biotic stress. In this condition, the seeds priming with AgriD led to notable effects on

germination rate, growth speed, and the plant's tolerance to both biotic stresses up to the tillering stage. An induction of plant defence might be hypothesized, as AgriD at the label dosage did not show an antifungal direct effect *in vitro* against plant pathogens. To improve PH formulation, several enzymatic hydrolyses were conducted using proteases of various origins on soybean flour and pea protein isolate. The aim is to generate original formulations, which will be evaluated through *in vivo* and *in vitro* assays to explore their potential involvement in plant responses to biotic and abiotic stresses.

Asia Mostacci was supported by a PhD fellowship funded within D.M.117/2023, PNRR, Missione 4, componente 2 "Dalla Ricerca all'Impresa" - investimento 3.3 – Cofunded by A. Costantino & C. S.p.a - Applicazioni di biostimolanti di origine vegetale per migliorare la risposta agli stress delle piante e/o loro prodotti, CUP H91I23000050007.

Overview of plant defense inducers' mode of action against the biotrophic pathogen *Podosphaera xanthii*. T. MARGARITOPOULOU¹, K. KOTSARIDIS², M. SAMIOTAKI³, D. TSIRIVA⁴, E. MARKELLOU¹. ¹Laboratory of Mycology, Scientific Directorate of Phytopathology, Benaki Phytopathological Institute, Stefanou Delta 8, 14561, Kifissia, Greece. ²Laboratory of Virology, Scientific Directorate of Phytopathology, Benaki Phytopathological Institute, Stefanou Delta 8, 14561, Kifissia, Greece. ³Protein Chemistry Facility, Biomedical Sciences Research Center "Alexander Fleming", Vari, 166 72, Greece. ⁴Research and Development Department, Phytorgan SA, Nea Kifissia, Greece. E-MAIL: th.margaritopoulou@bpi.gr

The high adaptability of the biotrophic fungus *Podosphaera xanthii* to diverse environments, along with the increasing negative effects of excessive fungicide use, necessitates the exploration of alternative control methods. Plant Defense Inducers (PDIs) activate plant defense mechanisms similarly to pathogens and are considered eco-friendly and sustainable for disease management. However, their mode of action is still not fully understood. Using transcriptomic, proteomic, and metabolomic approaches alongside physiological studies and targeted analyses, we demonstrated that two PDIs—the extract from the knotweed plant *Reynoutria sachalinensis* and a formulation of chitosan nanoparticles loaded with salicylic acid—effectively induce plant defences against *P. xanthii*. Our results revealed a direct correlation between the production and signalling of major cell membrane phospholipids, such as phosphatidylserine, phosphatidylethanolamine, and phosphatidic acid, which

are involved in the initial stages of signal perception. This correlates with the reprogramming of the protein quality control mechanism in the endoplasmic reticulum during the later stages of signalling, which also accompanies induced defense. Moreover, the application of PDIs enhanced the expression of plant defense-related proteins, including glutathione transferases, kinases, and chitinases. Protein-protein interaction enrichment indicated the formation of an interactive network that promotes defense. Simultaneously, we found that the pathogen secretes proteins related to ubiquitination during host invasion to manipulate the plant's protein translation and quality control machinery. Notably, the application of the PDIs significantly downregulated the expression of various secreted proteins from the pathogen, resulting in reduced powdery mildew severity.

This research was financially supported by the project "Innovations in Plant Protection for sustainable and environmentally friendly pest control, InnoPP - TAEDR-0535675 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", by the project "Nanoshield - T2EDK-02113", and Benaki Phytopathological Institute.

Biocontrol-mediated activation of defense mechanisms in grapevine against *Fusarium equiseti* infection. M.L. TELLO-MARISCAL¹, J.M. ALONSO DE ROBADOR², B. PINTOS², A. GOMEZ-GARAY². ¹National Institute for Agricultural and Food Research and Technology (INIA). National Research Council (CSIC), Ctra. N-VI. 28040 Madrid, Spain. ²Research Group FiVe-A, Department of Genetics, Physiology and Microbiology, Faculty of Biology, Complutense University of Madrid, Ciudad Universitaria, 28040 Madrid, Spain. E-MAIL: marisa.tello@inia.csic.es

Grapevine trunk diseases (GTDs) are among the most devastating threats to viticulture worldwide, causing significant economic losses and compromising vineyard longevity. In this study, we isolated and characterized a *Fusarium equiseti* strain from infected grapevine (*Vitis vinifera* L.) tissues, confirming its pathogenicity through Koch's postulates and assessing virulence via enzymatic and physiological assays. Given the limitations of chemical phytosanitary treatments, we evaluated the potential of *Meyerozyma guilliermondii* CECT13190, a patented yeast strain (ES-2792777 B2), as a biocontrol agent (BCA). The BCA demonstrated multiple plant growth-promoting traits, including indole-3-acetic acid (IAA) production, phosphate solubilization, siderophore synthesis, and atmospheric nitrogen fixation. Dual-culture

and resazurin assays showed strong antagonism against *F. equiseti*, supported by in planta experiments that revealed both preventive and curative effects on infected grapevine plants. Proteomic analyses revealed that *M. guilliermondii* modulates host plant defense by enhancing expression of key pathogenesis-related (PR) proteins, including β -1,3- glucanase, thaumatin, and chitinase, and activating systemic acquired resistance (SAR). Gene expression studies confirmed transcriptional regulation of these defense components under both prophylactic and therapeutic treatments. Our findings highlight the complex tripartite interaction between host plant, pathogen, and biocontrol agent, showcasing the ability of *M. guilliermondii* to induce molecular, enzymatic, and metabolic defense responses in grapevine. These results provide a strong foundation for sustainable GTD management strategies and contribute to the growing body of evidence supporting biocontrol as a viable alternative to synthetic fungicides in viticulture.

This research was supported by the Ministry of Science, Innovation and Universities, Spain.

POSTER PRESENTATIONS

Interactions between ontogenetic and acquired resistance shape grapevine metabolic responses to *Plasmopara viticola*. L. NASSI^{1,2}, G. FEDELE^{1,2}, I. RAGNO-LI³, L. ZHANG³, L. LUCINI³, V. ROSSI^{1,2}, T. CAFFI^{1,2}. ¹Department of Sustainable Crop Production, Università Cattolica dal Sacro Cuore, Piacenza, Italy. ²Research Center on Plant Health Modelling (PHEM), Department of Sustainable Crop Production, Università Cattolica dal Sacro Cuore, Piacenza, Italy. ³Department for Sustainable Food Process, Università Cattolica del Sacro Cuore, Piacenza, Italy. E-MAIL: luca.nassi@unicatt.it

Plant Resistance Inducers (PRIs) are promising alternatives to conventional plant protection products for controlling the grapevine pathogen *Plasmopara viticola*. This study aimed to investigate the metabolic effects caused by PRI-based treatments on grapevine leaves and to evaluate how these effects combine with the morpho-metabolic changes given by ontogenetic resistance. For this purpose, the effects of two well-known PRI-based plant protection products, Laminarin (LAM) and Potassium Phosphonate (PHO), were evaluated and compared with an untreated control (TNT). Ten leaves from the treated vine shoot, ranging from apical to basal, were sampled 48 h after PRIs treatment (Priming assessment) and 48 and 96 h post artificial inoculation of *P. viticola* (Defence response assessment). The effects on their

metabolism were assessed through an untargeted metabolomics approach using LC-MS. The annotated metabolites through plant-based specific databases and the list of compounds were subjected to unsupervised (PCA) and supervised (AMOPLS-DA and OPLS-DA) statistical analysis that allowed to highlight the main discrimination factors that influenced leaf metabolism. After treatments and 48 h after pathogen inoculation, the metabolic profiles were strongly affected by the leaf age and LAM showed a different modulation of metabolite profile compared to the other treatments. LAM and PHO shared a similar metabolite profile only after 96 h post inoculation. Overall, LAM and PHO treatments activated significantly different metabolic profiles respect to the timing of priming and pathogen inoculation. Moreover, the results of this study confirmed a significant influence of leaf age on leaf metabolism response.

This study was supported by the PhD in Agro-Food System (Agrisystem) of the Università Cattolica del Sacro Cuore (Italy).

***Salvia* spp. extracts contain bioactive terpenoids to control *Plasmopara viticola* infections on grapevine.**

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Downy mildew, caused by *Plasmopara viticola*, is one of the major threats to viticulture, especially in areas with warm and humid climate conditions. Grapevine downy mildew is controlled by frequent application of copper and synthetic fungicides, with possible negative impacts on human health and the environment. Alternative products, such as extracts of medicinal plants, are highly desired to develop sustainable plant protection strategies. Alcoholic extracts of *Salvia officinalis* previously showed inhibitory activity against phytopathogens. This study aims to characterize the activity of sage extracts against grapevine downy mildew and to identify bioactive metabolites. Alcoholic extracts of sage shoots, leaves, and flowers showed disease reduction in grapevine leaf disk assays comparable to copper, while a stem extract was only partially active against *P. viticola*. Fractions of shoot extracts were obtained using preparative liquid

chromatography and subjected to inhibitory activity tests on leaf disks. Active fractions were analyzed with untargeted metabolomics using liquid chromatography-high resolution mass spectrometry (LC-HRMS), and 25 putative bioactive compounds were annotated as terpenoids, such as eight abietane diterpenoids, three icetexane diterpenoids, three pentacyclic triterpenoids, two secoabietane diterpenoids, and nine other terpenoids. Five annotated compounds were identified by LC-HRMS analysis using reference standards, and their inhibitory activity was confirmed on leaf disks. The identification of novel bioactive compounds from *Salvia* spp. and the characterization of their mode of action will pave the way for the development of new sustainable alternatives for grapevine protection.

Inactive yeasts as preventive treatments against *Botrytis cinerea* on grapevine.

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The demand for alternatives to synthetic fungicides has been increasing due to the rise of pathogens resistant to active principles, consumers and producers' consciousness, and EU directives on sustainable farming. *Botrytis cinerea* is a worldwide distributed pathogen of grapevines causing losses of grape production. In the last years, yeasts and their derivatives have been studied as promising alternatives to fungicides for the management of plants' pathogens. In this study, a screening of various inactive yeast suspensions from the food industry was performed. The objective of this research was to check several yeast derivatives efficacy in plant stimulation and defence activation toward *B. cinerea*. Grapevine plants, cultivar Merlot grown in pots in a greenhouse, were treated twice using inactive yeast-based suspensions. Stomatal conductance was monitored 48 hours later to assess the plant metabolism. Inoculation of *B. cinerea* was performed on detached leaves at 72 hours after the second application and extension of the lesions was observed three days later. Results suggest that inactive yeast *Saccharomyces cerevisiae* grown on exhausted medium of *Salvia officinalis* efficiently reduce *B. cinerea* lesions on leaves in comparison to the untreated control. An increase in plants photosynthetic activity has

also been observed after the application of the exhausted medium of *S. officinalis*. Those extracts could offer interesting alternatives to synthetic fungicides. Further in vivo trials need to be conducted to investigate the efficiency of these compounds in protecting Grapevine production.

Testing the efficacy of cyanobacterial silver nanoparticles against fungal plant pathogens. C. AGLIETTI, M. CIANI, M. GIAMBERINI, A. ADESSI, L. GHELARDINI. *Department of Agricultural, Food, Environmental and Forest Sciences and Technologies (DAGRI), University of Florence, Piazzale delle Cascine 28, 50144 Florence, Italy.* E-MAIL: chiara.aglietti@unifi.it

Plant diseases caused by fungi and bacteria can be managed and controlled using a variety of techniques that have been implemented during time. However, traditional management often relies on the use of synthetic chemical pesticides, despite the growing need for more sustainable and eco-friendly solutions. Among these, silver nanoparticles (AgNPs) application has proven effective in inhibiting the growth of fungal and bacterial pathogens, but there is still a gap in identifying sustainable synthesis technologies and user-friendly application methods in plant health defense. This study explores the green synthesis of AgNPs using cyanobacteria, promising biofactories for the eco-friendly synthesis of metal nanoparticles (MNPs). The antifungal efficacy of AgNPs was assessed *in vitro* against *Botryosphaeriaceae* sp., *Gnomoniopsis castaneae* and *Fusarium* sp. by testing AgNPs obtained by cell-free culture medium, extracellular polysaccharides (EPS) extracted from cell-free culture medium, and cellular extract. The antifungal activity of the different cyanobacteria-derived AgNPs on the different fungal species was assessed through *in-vitro* growth tests. To verify the persistence of the antifungal effect, at the end of the treatment each fungal colony was reinoculated onto PDA petri dishes without AgNPs. The mycelial growth of two of the three fungal species tested was clearly inhibited by high concentration of AgNPs (100ppm), although the effect was not found to be persistent on reinoculated PDA dishes. Cellular extract was the most effective synthesis method, inhibiting fungal radial growth of c.a 60% (1.5 cm against 4 of control). These results underline the potential of cyanobacteria-derived AgNPs as a sustainable antifungal method for plant disease control.

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biannual projects for Temporary Researchers, edition 2024- 2025 (D.R. n. 419 02/05/2023)".

Copper nanoparticles against *Colletotrichum gloeosporioides*: effectiveness and insights on the mode of action. A. MALANDRAKIS^{1,2}, N. KAVROULAKIS², M. MATHIOUDAKIS², M. KISANDRAKI², V. KATZOURAKIS³, C. CHRYSIKOPOULOS³. ¹*School of Chemical and Environmental Engineering, Technical University of Crete, 73100 Chania, Greece.* ²*Institute for Olive Tree, Subtropical Plants and Viticulture, Hellenic Agricultural Organization "Dimitra", Agrokipio-Souda, 73164 Chania, Greece.* ³*Department of Civil Infrastructure and Environmental Engineering, Khalifa University of Science and Technology, Abu Dhabi 127788, UAE.* E-MAIL: amalandrakis@tuc.gr

Anthracnose, caused by the fungal pathogen *Colletotrichum gloeosporioides*, is among the most prevalent and severe postharvest diseases affecting various fruits, including olive, citrus, and avocado. In an era where chemical control is being challenged by fungicide resistance development and strict EU legislation, causing the withdrawal of a large number of established active ingredients used for disease management, metallic nanoparticles (MNPs) represent promising, eco-friendly fungicide-alternatives for controlling pathogens. In this study, the effectiveness and potential fungitoxicity mechanisms of copper nanoparticles (Cu-NPs) was tested against *C. gloeosporioides* strains isolated from orange and avocado fruit both *in vitro* and *in vivo*. Cu-NPs outperformed their ionic counterpart Cu(OH)₂ (which was used as a reference fungicide) in reducing mycelial growth and disease symptoms. Pearson correlation analysis revealed a positive cross resistance between Cu-NPs and the demethylase inhibitor (DMI) fungicide difenoconazole. A significant antagonism between Cu-NPs and detoxifying inhibitor agents DEM, PBO as well as DMI fungicides difenoconazole, prochloraz, and prothioconazole was observed as indicated by the reduced effectiveness of applied combinations against the pathogen. Furthermore, an induced C-14 alpha-demethylase overexpression was found, at least partly, to be responsible for the observed superior fungitoxic action of Cu-NPs against *C. gloeosporioides*. Overall, inhibition of ergosterol biosynthesis is proposed as a candidate fungitoxicity mechanism for the mode of action of Cu-NPs against the pathogen.

This research was financially supported by the European Union-Next Generation EU, Greece 2.0 National Recovery and Resilience plan (project code: TAEDR-0535675).

Green synthesized silver nanoparticles against *Pseudomonas savastanoi* pv. *savastanoi*: effectiveness and mode of action. N. KAVROULAKIS¹, A. MALANDRAKIS^{1,2}, M. KISSANDRAKI¹, V. KATZOURAKIS³, C. CHRYSIKOPOULOS³. ¹*Institute of Olive Tree, Subtropical Plants and Viticulture, ELGO-DIMITRA, Agrokipio-Souda, 73164 Chania, Greece.* ²*School of Environmental Engineering, Technical University of Crete, 73100 Chania, Greece.* ³*Department of Civil Infrastructure and Environmental Engineering, Khalifa University of Science and Technology, Abu Dhabi 127788, UAE.* E-MAIL: kavroulakis@elgo.gr

Knot disease is an important olive tree bacterial infection caused by *Pseudomonas savastanoi* pv. *savastanoi*, leading to significant economic losses in olive production worldwide. Control measures against the pathogen include cultural methods, selection of resistant cultivars, use of biocontrol agents and the application of copper-based bactericides which are the only available chemical control option. Although copper is a multisite inhibitor, various cases of *P. savastanoi* pv. *savastanoi* resistant to this metal have been reported leading to control failures. This lack of available alternatives in combination with the loss of copper effectiveness make the need for alternative control measures an urgent matter. In this study, the effectiveness and potential antimicrobial mechanisms of green synthesised silver nanoparticles (GAg-NPs) were tested against two *P. savastanoi* strains isolated from olive orchards both *in vitro* and *in vivo*. GAg-NPs exhibited superior bactericidal action compared with chemically synthesized nano silver (Ag-NPs) and a reference Cu(OH)₂-containing bactericide in reducing bacterial growth and disease symptoms. *In vitro* experiments using the strong chelating agent EDTA in combination with NPs indicated that silver ion release is not the main mechanism responsible for the antibacterial action of GAg-NPs against the pathogen. Overall, GAg-NPs have demonstrated a great potential to be used as copper alternatives against Olive knot disease and a promising tool against bactericide resistance.

This research was financially supported by the European Union-Next Generation EU, Greece 2.0 National Recovery and Resilience plan (project code: TAEDR-0535675).

Environmentally-friendly methods and tools for disease control in table grapes. E. CHALBAOUI, P. ALBANESE, F. SANTORO, S. GUALANO, F. DIGIARO, F. VALENTINI. *Centre International de Hautes Etudes Agronomiques Méditerranéennes Bari (CIHEAM Bari), Via Ceglie, 9 - 70010 Valenzano (BA), Italy.* E-MAIL: chalbaouieya@gmail.com

Powdery mildew, downy mildew, gray mould, and thrips represent major challenges in table grape cultivation, particularly in the Apulia region of southern Italy, where they can severely reduce both yield and fruit quality. Traditionally, these threats have been managed through chemical treatments. However, increasingly stringent EU regulations on pesticide use—driven by environmental concerns and consumer awareness of food residues—are prompting a shift toward more sustainable viticultural practices. This study investigates the efficacy of hydrogen peroxide, a GRAS (Generally Recognized As Safe) product, applied using a Micro 28 precision spraying system, as an alternative to conventional chemical-based Integrated Pest Management (IPM). The trial was conducted in a commercial vineyard in Acquaviva delle Fonti (Bari) from February to September. Disease and pest presence, weather conditions, and grape ripeness were monitored using a decision support system, visual inspections, and laboratory analyses. The hydrogen peroxide treatment, when applied with the Micro 28 system, showed comparable disease control to traditional IPM while significantly reducing treatment time and costs. Its effectiveness is attributed to the precision and timeliness of applications, which are critical for optimal performance of GRAS products. Importantly, this method also contributes to a lower environmental impact by reducing machinery use, thereby minimizing carbon emissions and soil compaction. These findings highlight the potential of combining GRAS products with innovative application technologies for sustainable disease management in viticulture. Further research is necessary to validate the results across different environments and growing seasons.

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Application of plant-formulations to improve growth parameters and resistance to stresses of Mediterranean crops. A. BEROUAKEN¹, C. PITUELLO², A. SCENNA¹, O. INCERTI¹, A. IPPOLITO¹, S. M. SANZANI¹. ¹*Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, via Amendola 165/A, 70126 Bari, Italy.* ²*SICIT Group S.p.A., Via Arzignano 80, 36072 Chiampo (VI), Italy.* E-MAIL: simonamarianna.sanzani@uniba.it

Moringa oleifera Lam. (Moringaceae), consumed in several tropical and subtropical countries, is a highly valued multi-purpose herbal plant with numerous medici-

nal properties and high nutritional value, being rich in polyphenols, bioactive peptides, carotenoids, and glucosinolates. Besides, a great interest is emerging towards the exploitation of Moringa as an added-value food ingredient. In a recent study the preharvest application of two lab-made aqueous extracts from *Moringa oleifera* leaves (MLEs) improved yield, quality, and storability of lettuce grown in a hydroponic system, as compared to an untreated control and other biostimulant formulations on the market. In the present investigation a Moringa-based formulation has been tested to improve the growth parameters of two relevant crops as wheat and tomato. Preliminary assays on seeds were conducted confirming a significant increase in germination rate and root and shoots length. As such the plant should withstand better stressing events. Further *in planta* assays are in progress.

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Antimicrobial activity, and defense gene activation by natural extracts: toward sustainable plant health management. C. BILEN¹, L. VACCARO¹, S. LAERA¹, P.R. ROTONDO¹, S. POLLASTRO¹, M. MARASHI¹, T. MASCI¹, R. SPANÒ², D. EL CHAMI³, F. FARETRA¹, R.M. DE MICCOLIS ANGELINI¹. ¹Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, Bari, Italy. ²National Research Council -Institute for Sustainable Plant Protection, Via Amendola, 122/D Bari, 70126, BA, Italy. ³TIMAC AGRO Italia S.p.A., 26010 Ripalta Arpina, CR, Italy. E-MAIL: ritamilvia.demiccolisangelini@uniba.it

The growing demand for sustainable agricultural practices has accelerated research on eco- friendly plant health management strategies, focusing on natural substances like plant extracts with bioactive compounds for antimicrobial activity or plant defense-inducing properties. This study evaluates natural extracts as sustainable alternatives for managing plant diseases. A variety of substances, including essential oils, extracts from ornamental and edible plants, by-products from beer and coffee processing, *Aloe vera* extracts, and selected commercial formulations, were assessed for their antimicrobial efficacy through *in vitro* and *in vivo* assays. Several extracts demonstrated notable bioactivity: *Agapanthus africanus* showed significant antifungal effects against

Botrytis cinerea, while essential oils from thyme, tea tree, and lavender were effective against both *B. cinerea* and *Pseudomonas syringae* pv. *tomato*. Greenhouse trials on tomato plants revealed the protective effects of *Aloe vera* polysaccharides and ornamental plant extracts, particularly *Eremophila nivea* and *Limoniastrum monopetalum*, against PVY^{c-to}, a recombinant strain of *Potato virus Y* necrogenic on tomato. Additionally, microencapsulated thyme essential oil and alginate- based formulations showed enhanced *in vivo* performance, particularly against the bacterial and viral pathogens. Gene expression analyses at 12- and 72-hours post-treatment indicated strong upregulation of the defense-related gene *PR1*, with extracts from coffee residues, *A. vera* gel, and the synthetic resistance inducer BION[®]. The latter used as reference, elicited the most pronounced response, while *PR4* exhibited a more moderate induction. These findings highlight the promising role of natural plant-derived and agro-industrial substances in providing both direct antimicrobial action and stimulation of plant immune responses, contributing to sustainable health management strategies.

This research was financially supported by Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) - MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 - D.D. 1032 17/06/2022, CN00000022), and INVESTIMENTO 3.3 D.M. 352/2022 and TIMAC AGRO Italia S.p.A. and by Innovazione di prodotto e di processo nel florovivaismo biologico per il verde pubblico, healing e kitchen garden BIOGARDEN (MASAF Progetti di Ricerca in agricoltura Biologica Avviso pubblico per la concessione di contributi per la ricerca in agricoltura biologica n. 9220340 del 8 ottobre 2020 - CUP H93C24000920001).

Preliminary assessment of plant powders and aqueous extracts from different botanical species to inhibit fungal growth under *in vitro* conditions. M.G. MOREA, G. RICCIARDI, A. LIBUTTI, A. CARLUCI. Department of Agricultural Sciences, Food, Natural Resources and Engineering, University of Foggia, Via Napoli 25, 71122, Foggia, FG, Italy. E-MAIL: maria.morea@unifg.it

Many botanic plants are reported to be rich in antioxidant and polyphenolic compounds, resulting in effectiveness to prevent microbial contaminations (bacteria and fungi). The natural compounds might be useful in biocontrol against fungal pathogens instead of synthetic fungicides. This preliminary study was carried out to assess the ability of five botanical species to control 15

fungal soilborne pathogens by the use of their dehydrated tissues and aqueous extracts. Vegetative tissue portions of *Raphanus raphanistrum*, *Brassica nigra*, *Sinapis alba*, *Matricaria chamomilla* and *Solanum lycopersicum* were collected and left to dry at room temperature for one week and then at 30°C until constant weight, before to be finely grounded and used at 2% as plant powders (PPs) by inclusion in PDA medium. The same PPs were used to obtain aqueous extracts (AEs) by infusion in distillate water (ratio w:v; 1:10) at room temperature for 48 h. The aqueous fractions were obtained by two centrifugations at 20.000 rpm and then added to PDA medium at four different concentrations (25, 50, 75 and 100%). For both kinds of experiment (inclusion PPs and AEs), three PDA Petri dishes were inoculated with the fungal species, and every 48 h the diameter growths were measured until 21 days. The data collected were used to calculate the inhibition activity played from each PPs and AEs. The dataset was subjected to statistical analyses (multifactorial and one-way Anova, and principal components). This study highlighted that AEs were more effective to inhibit fungal growth.

This research was financially supported by Fondazione Puglia (Ricerca Scientifica e Tecnologica) – Italy through the grant “Estrazione di Biomolecole da residui vegetali di origine agro-industriale e loro impiego per la Sostenibilità delle colture Agrarie – BioSostAgr (2023-2025).

Embryo rescue of local *Vitis vinifera* varieties and F1 progenies for rethink the powdery mildew protection strategies. F. SPATARO, G. BOTTALICO, A. CAMPANALE, A. AGNUSDEI, F. DALENA, E. CHIAROMONTE, F. FARETRA, S. POLLASTRO. *Department of Soil, Plant and Food Sciences, University of Bari, Via G. Amendola, 165A/ 70126 Bari, Italy.* E-MAIL: giovanna.bottalico@uniba.it

This study explored the efficacy of *in vitro* embryo rescue as a valuable tool for the early and efficient production of F1 progeny of *Vitis vinifera* and subsequent assessment of resistance/susceptibility to powdery mildew. *In vitro* culture of grape seeds (pips) was performed starting from immature and mature grape bunches of six varieties including wine and table grapes with seeded and seedless berries, selected for different levels of tolerance/resistance to powdery mildew and their hybrids. Following surface sterilization, the seeds/embryos were cultured on medium supplemented with GA3, IAA, and activated charcoal. An equivalent number of seeds was also directly sown in soil. *In vitro* and *in vivo* germination were monitored, as well as the growth of seedlings

transferred to a greenhouse, where resistance/susceptibility to powdery mildew was evaluated through natural infection. A higher germination percentage was proved for *in vitro* tests compared to direct sowing in soil, as well as significantly earlier and more uniform germination time, particularly when embryos were excised from immature seeds. For evaluations on the F1 progeny, the most promising results in terms of powdery mildew resistance were obtained for an unidentified variety. This study demonstrates that embryo rescue is an effective method to accelerate seed germination and for the early assessment of resistance/susceptibility to powdery mildew in F1 progeny, offering a significant advantage for grapevine breeding programs.

This research was financially supported by Agritech National Research Center and received funding from the European Union Next-Generation EU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) –MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

BIOBIVE PROJECT: eco-innovative delivery systems for resilient horticulture and sustainable control of soil-borne pathogens. M. CARLUCCI, F. CONVERTINO, A. HACHEM, C. CARBOTTI, A. PACIFICO, G. VOX, F. NIGRO, E. SCHETTINI. *Department of Soil, Plant and Food Sciences (DiSSPA), Via Giovanni Amendola 165/A, University of Bari- Aldo Moro, Bari, 70126, Italy.* E-MAIL: franco.nigro@uniba.it

The BioBIVE project introduces an innovative approach aimed at developing bio-based and bioactive delivery platforms for the management of plant diseases in a variety of horticultural crops. The global agricultural sector faces significant challenges from soil-borne diseases, which result in considerable economic losses. The increasing global relevance of these diseases is evidenced by their high frequency in specific regions, exacerbated by intensive cropping systems and climatic conditions that favor pathogen proliferation. Soil-borne fungal pathogens such as *Phytophthora nicotianae*, *Fusarium oxysporum* f. sp. *lycopersici*, *Pseudopyrenochaeta lycopersici*, and *Verticillium dahliae* primarily infect roots, causing root rot, wilting, and chlorosis. These pathogens can persist in soil and plant debris for extended periods, rendering their control particularly challenging. A thorough understanding of their biology, ecology, and epidemiology is essential for developing sustainable disease management strategies that extend beyond conventional

chemical control. Effective management requires an integrated approach, incorporating advanced diagnostic tools, resistant plant varieties, and innovative agricultural practices to reduce pathogen spread and minimize crop losses. BioBIVE Project aims to incorporate natural active compounds - such as plant-derived basic substances, phlorotannins (polyphenols from marine algae), and beneficial microorganisms - into eco-friendly, biodegradable delivery systems. These systems will include mulch films, biochar, and sprayable biodegradable coatings, designed to enhance crop productivity and resilience while reducing the environmental impact of agricultural practices.

This study has received funding from the BioBIVE Project (no. 101130442), funded by the EU through the Horizon Europe Framework Programme (HORIZON-CL4-2023-RESILIENCE-01-34).

Future innovation for pesticide use reduction in agriculture – FORTUNA. D. TSITSIGIANNIS. *Department of Crop Science, Iera Odos 75, Agricultural University of Athens, Athens, 11855, Greece.* E-MAIL: dimtsi@aua.gr

FORTUNA is a collaborative research project supported by Horizon Europe. Its primary objective is to identify the forthcoming research requirements essential for realizing the EU policy objectives outlined in the Farm to Fork and Biodiversity strategies. These strategies are geared towards reducing the use and risk of chemical pesticides, as well as the use of more hazardous pesticides, by 50% by 2030. Advanced Integrated Pest Management (IPM) strategies are characterised by implementing a high degree of preventive measures to avoid establishment of pests and reduce significantly the use of chemical pesticides. However, the implementation of IPM at a multidisciplinary level is still lacking due to economic reasons and a low acceptance by farmers. A pivotal aspect of FORTUNA involves the identification and analysis of available methods, tools, and mechanisms from biological/ecological, technological, and socio-economic standpoints. These endeavors aim to facilitate a significant reduction in the usage and associated risks of chemical pesticides, ultimately fostering the adoption of innovative pesticide-free farming practices. The outcomes and strategies derived from these efforts are discussed with stakeholders across the food and supply chain. Such stakeholder engagement aims to foster a transition away from heavy reliance on chemical pesticides while concurrently supporting robust yields and ensuring food security. A critical aspect of this process involves identifying strategies that entail lower risks of

productivity and profitability losses compared to other approaches, with a focus on long-term sustainability and viability.

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Ozonized olive oil as an innovative natural fungicide: a promising alternative to synthetic agents. M.E. MERZOUKI¹, D. BOUZID², S. MERZOUKI³, M.M. ZERROUG². ¹*Department of Science and Technology, Faculty of Science and Technology, Optics and Precision Mechanics, Ferhat Abbas University Setif 1, 19000 Sétif, Algeria.* ²*Laboratory of Applied Microbiology, Faculty of Natural and Life Sciences, Ferhat Abbas University Setif 1, 19000 Sétif, Algeria.* ³*Private Medical Clinic, Sétif, Algeria.* E-MAIL: bouzid.djihane@yahoo.fr

The increasing resistance of pathogenic fungi to conventional antifungal agents necessitates the development of innovative, natural alternatives. This study investigates the antifungal potential of ozonized olive oil, obtained by bubbling a medical-grade oxygen-ozone gas mixture through cold-pressed *Olea europaea* L. oil. The antifungal activity was evaluated against five filamentous fungi (*Aspergillus niger*, *Penicillium digitatum*, *P. expansum*, *Aspergillus itaconicus*, *Fusarium solani*) and two yeasts (*Candida albicans*, *Aureobasidium pullulans*) using agar diffusion and broth dilution methods. Results demonstrated a strong inhibition of fungal growth, with *Fusarium solani* being the most sensitive (inhibition zone of 36.66 ± 4.71 mm). Minimum fungistatic and fungicidal concentrations (MFCs/MFCc) ranged from 0.38 to 18.3 mg mL⁻¹. Notably, the antifungal effect on *C. albicans* was significantly stronger than in previous reports, with an MFC of 0.38 mg mL⁻¹. The ozonation process leads to the generation of active compounds including ozonides and peroxides, which are believed to disrupt fungal membranes and cellular metabolism via oxidative stress mechanisms. These findings highlight the potential of ozonized olive oil as a safe, biodegradable, and effective natural fungicide. Its use could reduce dependency on synthetic fungicides and contribute to sustainable agriculture and infection control. Further investigation for olive oil-based formulations and their field application is needed.

In vitro evaluation of natural compounds against *Verticillium* wilt of olive. M. MOUMNI¹, S. M. MAKAU^{1,2},

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Verticillium wilt, caused by the soilborne fungus *Verticillium dahliae*, poses a significant threat to olive trees (*Olea europaea*) globally, leading to substantial economic losses. Its persistence in soil makes management challenging. In this study, two *V. dahliae* strains (AN1, AN2) were isolated from diseased olive trees in Cerignola, Italy. An *in vitro* study was conducted to evaluate antimicrobial activities in contact phase of COS (chito-oligosaccharides)-OGA (oligo-galacturonides), chitosan hydrochloride, *Urtica* spp., and *Equisetum arvense*, all at 0.2% concentration. Five essential oils (EOs) – *Rosmarinus officinalis*, *Rosmarinus officinalis* var. *verbenone*, *Lavandula hybrida*, *Origanum majorana*, and *Thymus vulgaris* – were evaluated at 0.1% concentration in the contact phase and at two distinct concentrations/EO in the vapor phase. *Urtica* spp. showed the highest mycelial growth inhibition (92.9% for AN1, 82.6% for AN2), followed by COS-OGA (74.8% and 76.0%) and chitosan hydrochloride (65.8% and 63.4%). *E. arvense* was the less effective. In contact assays, *T. vulgaris* completely inhibited mycelia growth of both isolates, and *L. hybrida* completely inhibited AN1. *O. majorana* showed 89.4% (AN1) and 63.3% (AN2) inhibition. In vapor phase, *T. vulgaris* was the most effective, achieving 99.0% and 82.5% inhibition at 90.9 µL/L and 45.4 µL L⁻¹, respectively against AN1. *In vivo* investigations are required to validate the possible application of these treatments for *Verticillium* wilt disease management.

This work was supported by the Croatian Science Foundation under the project number UIP-2020-02-7413.

Recent findings on *Sclerotinia* spp. control with a formulation based on eugenol, geraniol and thymol, on different leafy vegetables and fennel. M. PAGNANI¹, F. GUASTAMACCHIA¹, A. GUARNONE², D. BITONTE², S. CIANNONE². ¹Sipcam Oxon, ²Sipcam Italia, R&D Department, Via Sempione 195, 20016 Pero, MI, Italy. E-MAIL: mpagnani@sipcam.com

Leafy vegetables are highly specialized crops that require careful agronomic and phytosanitary management. Among the most damaging pathogens is crown rot, caused by *Sclerotinia sclerotiorum* and *S. minor*. Sipcam studied the application of a commercial fungicide based on Eugenol, Geraniol, and Thymol (3LOGY®), to control *Sclerotinia* in several open-field and greenhouse-grown horticultural crops such as lettuce, baby leaf, herbs, edible flowers, cabbage, and fennel. Field and greenhouse trials were conducted in 2021 and 2022 in collaboration with research centers. Trials followed EPPO guidelines, using randomized block designs with four replicates and natural infection conditions. Treatments were applied with a backpack sprayer before symptom onset. Disease incidence and severity were assessed and data were statistically analyzed using ANOVA and the Student-Newman-Keuls test ($P < 0.05$). Four trials were conducted on fennel, baby leaf lettuce, rocket, and open-field lettuce. 3LOGY® demonstrated high efficacy in all trials. In fennel, it achieved up to 91.1% efficacy on disease severity. In baby leaf lettuce and rocket, combinations with biological and chemical fungicides achieved 100% control.

The inhibitory and fungicidal effect of Saharan plants essential oils on *Fusarium* head blight species. H. HECHACHNA^{1,2}, L. BENFEKIH¹, N. GOURINE², M. YOUSFI². ^{1,2}Laboratory for Research on Medicinal and Aromatic Plants, Department of Biotechnology and Agroecology, Faculty of Nature and life sciences, Saad Dahlab University Blida 1, Route de Soumaa, 09000 Blida, Algeria. ²Laboratory for Research on fundamental Sciences, Po Box 35G, Ammar Telidji University of Laghouat, Laghouat 03000, Algeria. E-MAIL: leila.benfekih@univ-blida.dz

Fusarium head blight is a serious fungal disease of wheat caused by species belonging to the *Fusarium* genus worldwide. *F. graminearum* and *F. culmorum* have been reported as the most common causal agents for *Fusarium* head blight as well as major producers of mycotoxins leading to significant yield losses in Algerian cereal crops. Natural compounds such as essential oils (EOs) have been recognized for their efficient antifungal activity, proposed for using in agricultural fields. In this regard, the antifungal effect of three tested essential oils extracted from aerial plant parts of *Artemisia herba alba*, *Artemisia campestris* and *Teucrium polium* harvested in Laghouat region (North Central Sahara, Algeria) was determined on the isolates *F. graminearum* (F5883) and *F. culmorum*. The EOs displayed effective antifungal action, which was most likely due to the presence of bioactive components when comparing plant harvest sea-

son. Our findings showed that *F. culmorum* isolate was more sensitive than *F. graminearum* strain to *A. campestris* essential oil from the plant harvested in autumn season than for the one collected in winter, with minimal inhibitory concentration and minimal fungicidal concentration values of 1.25 $\mu\text{L mL}^{-1}$ (v/v). The *A. herba alba* EO exhibited a lower (5 $\mu\text{L mL}^{-1}$ v/v) fungicidal activity against both *Fusarium* species whereas *T. polium* essential oil from the plant collected in autumn and winter did not affect these fungi (MIC and MFC >20 $\mu\text{L mL}^{-1}$).

Antifungal activity of methanolic and aqueous extracts of *Pistacia atlantica* from central Algeria: an in vitro study.

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Medicinal plants are a rich source of bioactive compounds that may serve as new antifungal agents. The Atlas pistachio (*Pistacia atlantica*) is widely used in traditional medicine for its antimicrobial properties. This study aimed to evaluate in vitro the antifungal activity of aqueous and methanolic extracts from the leaves and unripe fruits of *P. atlantica* against five phytopathogenic fungi: *Fusarium* sp., *F. solani*, *F. oxysporum*, *Aspergillus fumigatus*, and *A. flavus*. Aqueous and methanolic extracts were prepared by maceration of the plant material. Total polyphenol and flavonoid contents were quantified spectrophotometrically using standard methods. Major phenolic constituents were identified by LC-MS/MS. Antifungal activity was assessed on PDA agar by measuring inhibition zone diameters and calculating percentage inhibition of mycelial growth (PI %) at various extract concentrations. The methanolic leaf extracts stood out for their high phenolic content (352.10 \pm 0.70 mg GAE g⁻¹ extract and 163.60 \pm 5.67 mg RE g⁻¹ extract) and contained twenty-one phenolic compounds identified by LC-MS/MS, including rutin, gallic acid, coumaric acid (the principal phenolic), maleic acid, naringenin, and hydroxycoumaric acid. High-doses methanolic leaf extracts significantly inhibited in vitro the growth of *Fusarium*, with inhibition rates ranging from 44.44% to 75.16%, whereas no notable antifungal effect

was observed against *A. fumigatus* or *A. flavus*. Methanolic leaf extracts of *P. atlantica*, exhibit particularly strong antifungal activity against *Fusarium* species and represent a promising natural source of antifungal compounds. These preliminary findings pave the way for further studies on biofungicide formulation and the assessment of their efficacy and phytotoxicity under crop production conditions.

**CONCURRENT SESSION H1 –
Smart innovations in plant
pathology: the digital Era**

SESSION KEYNOTES

Decision Support Systems for tactical management of plant diseases. V. ROSSI. Research centre on Plant Health Modelling (PHEM), Università Cattolica del Sacro Cuore, Via Emilia Parmense 84, 20100 Piacenza, Italy. E-MAIL: vittorio.rossi@unicatt.it

The increasing demand for sustainable crop protection requires tools that combine precision, reliability, and real-time support. Decision support systems (DSSs) are informatic tools that provide farmers (or agronomists) with the information needed for addressing key questions in crop protection, e.g., is there a disease risk? If so, are plants susceptible to infection? Or, are they protected from a previous fungicide application? Then, which plant protection product (PPPs) should I use and at what dosage? And finally, are environmental conditions suitable for the PPPs? Modern DSSs are based on the following main components: i) an integrated system for monitoring the crop environment (plants, pathogens, and the physical environment) and may include multiple sensors (proximal and remote, including satellites), weather stations or algorithms estimating weather data for specific geographic coordinates, Apps for pathogen detection and disease assessments, etc.; ii) structured databases in which monitoring data are stored and checked for consistency/quality; iii) databases for plant protection products that can be used in the specific agricultural context (e.g., in a country, in conventional, IPM or organic farming, under production specifications, etc.) and provide all the key information on the product, including dosages and the risk that target pathogens develop resistance; iv) mathematical models that use monitoring data and database information as input to predict plant growth and development, the plant canopy volume, infection periods for multiple pathogens, disease onset and development, physical mode of action of fun-

gicides, etc.); v) decision algorithms that transform model output into agronomic information (e.g., the plant is highly susceptible, the infection risk is high, the residual activity of a previous fungicide application does not provide sufficient crop protection, etc.); vi) a user interface that allows the user to navigate within the DSS, interact with the DSS, and receive up-to-date information and easy-to-use decision supports; vii) a crop diary in which the users insert information on agronomic interventions, which may also be used as model inputs. Mathematical models and decision algorithms are the core of a DSS and determine its reliability; different kinds of modeling approaches exist (e.g., data-based vs process-based models) that provide different performances in terms of accuracy, robustness, versatility and scalability. Demonstration of the DSS reliability under varying agricultural environments and situations, and benefits originated by the DSS use is then a key step for DSS success. In conclusion, DSSs have the potential to become a cornerstone of precision plant protection, enabling a shift from calendar-based to data-driven, adaptive crop protection that enhance efficacy and (economic, environmental and social) sustainability in modern agriculture.

ORAL PRESENTATIONS

Early detection of *Botrytis cinerea* in horticultural crops using deep learning segmentation, transformer-based classification, and molecular quantification. E. KALOGEROPOULOU¹, P. CHRISTAKAKIS², D. KAPETAS², C. KLARIDOPOULOS³, E.M. PECHLIVANI². ¹Laboratory of Mycology, Scientific Directorate of Phytopathology, Benaki Phytopathological Institute, 14561 Athens, Greece. ²Information Technologies Institute, Centre for Research and Technology Hellas, 57001 Thessaloniki, Greece. ³KnowHow S.A., 15451 Athens, Greece. E-MAIL: e.kalogeropoulou@bpi.gr

Early and accurate detection of *Botrytis cinerea*, the causative agent of grey mould disease, is critical for mitigating yield losses in high-value horticultural crops such as cucumber, tomato and pepper. This study integrates advanced artificial intelligence (AI) approaches and molecular techniques to detect *B. cinerea* during early and even latent infection stages. Multi-spectral image datasets were acquired at various wavelengths ranging from 450 to 900nm, including RGB and generated vegetation indices, from both artificially and naturally infected plants under controlled conditions. Deep learning models, including U-Net++ and YOLO, were used for high-accuracy leaf segmentation. Fea-

ture descriptors were extracted using advanced Vision Transformer architectures and subsequent classification was performed using ensemble methods involving K-Nearest Neighbors (KNN), Long Short-Term Memory (LSTM) and ResNet models. The optimal configurations achieved a high overall accuracy of 92% for cucumber, with segmentation mean average precision (mAP50) scores exceeding 81% for tomato and 86% for pepper. Classification F1-scores reached 71,12% and 81,13% for tomato and pepper, respectively. Complementary RT-qPCR analyses quantified fungal biomass in plants, confirming AI-based predictions even at asymptomatic stages. This integrated approach demonstrates a robust and scalable framework for early disease detection, supporting the development of *in situ* diagnostic systems in precision agriculture.

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Machine learning-based prediction of olive tree health against Vercillium wilt using rhizosphere microbiome profiles. L.F. ARIAS-GIRALDO¹, L. ANTONIELLI², S. COMPANT², J.A. NAVAS-CORTÉS¹, B.B. LANDA¹. ¹Institute for Sustainable Agriculture (IAS-CSIC), Córdoba, Spain. ²Center for Health & Bioresources, Bioresources Unit, AIT Austrian Institute of Technology GmbH, Austria. E-MAIL: lfarias@ias.csic.es

Verticillium wilt, caused by *Verticillium dahliae*, poses a major threat to Mediterranean olive production due to its severe impact and the lack of effective post-infection treatments. Modulating the soil microbiome has emerged as a promising strategy to manage this disease, given the rhizosphere's role in naturally suppressing soil-borne pathogens. In this study, we analysed the rhizosphere microbiome of 720 olive trees (359 healthy and 361 diseased) from 180 commercial orchards in Andalusia, Spain. Bacterial and fungal communities were characterized through amplicon sequencing of the 16S rRNA (V1-V2) and ITS1 regions. Environmental, climatic, soil, and agronomic data were also collected for each orchard. Using classical ecological approaches and supervised machine learning, we explored microbial patterns associated with environmental variables that predict tree health. Random Forest and Lasso regression models were tested on four datasets: bacterial ASVs (N=1251), fungal ASVs (N=360), merged ASVs (N=1611), and a set

of differentially abundant ASVs between healthy and diseased trees (N=55). Random Forest slightly outperformed Lasso, achieving the best results with the merged dataset (ROC-AUC ~ 0.85), especially in accurately identifying diseased trees. Four ASVs, *Peribacillus* spp., *Fusarium* spp., *Aspergillus* spp., and *Pseudomonas* spp., emerged as key predictors of tree disease status across both models, highlighting their potential as microbial biomarkers for olive tree health. Although the DA subset yielded competitive results, the inclusion of additional ASVs in the merged dataset suggests complex microbial interactions that enhance prediction accuracy.

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Stem colonisation and proximal sensing-based detection of *Xylella fastidiosa* infection in olive trees. M. ROMÁN-ÉCIJA, C. OLIVARES-GARCÍA, B.B. LANDA, J.A. NAVAS-CÓRTEZ. *Instituto de Agricultura Sostenible (IAS), Consejo Superior de Investigaciones Científicas (CSIC), 14004 Córdoba, Spain.* E-MAIL: mromanecija@ias.csic.es

The emergence of *Xylella fastidiosa* (Xf) poses a serious threat to agricultural and natural ecosystems. Xf ability to adapt to varying environments raises concerns about potential host shifts and increased virulence in the Mediterranean region. Moreover, accurate and prompt detection is crucial for effective disease control. Understanding the interactions of European Xf strains and local olive cultivars responses is key for designing targeted management strategies. The pathogenicity and extent of stem colonization of five Xf strains of different subspecies were investigated and the sequence types (ST) inoculated on three olive cultivars widely grown in Spain. All tested strains were able to infect olive plants, although detection rates declined over time. Strains belonging to subspecies *pauca* (XYL1961/18 of ST80 and De Donno of ST53) exhibited higher colonization levels and bacterial loads compared to those of subspecies *multiplex*. Additionally, we assessed the physiological impact of Xf infection on the inoculated olive plants analyzing leaf spectral signatures using non-invasive handheld proximal sensors, allowing the estimation of 74 vegetation indices. Among these, 17 to 31 indices, mainly associated to carotenoids, flavonoids, xanthophylls and blue band related indices, proved to be reliable indicators of infection status, achieving over 90% classification accuracy across olive cultivars. Our findings emphasize the importance of understanding host-pathogen interactions

between local cultivars and Xf strains adapted to Mediterranean conditions. Proximal sensing technologies proved to be useful tools for early detection of Xf-infected plants, even before symptoms appearance. Furthermore, these approaches could be integrated into breeding programs to support the development of Xf-resistant olive cultivars.

This research was financially supported by Projects BeXyl (Grant ID 101060593, EU-Horizon Europe), XF-ACTORS (Grant ID 727987, EU-Horizon 2020), E-RTA2017-00004-C06-02 (AEI-INIA Spain) and the Spanish Olive Oil Interprofessional, and the Projects PIE202240E067, PIE202340E021 and the PTI-SolXyl of CSIC.

Modelling multitrophic interactions between grapevine, microorganisms causing sour rot of bunches, and the *Drosophila* spp. Vectors. G. FEDELE^{1,2}, C. BRISCHETTO^{1,2}, L. CHIAPPAROLI^{1,2}, V. ROSSI^{1,2}, T. CAFFI^{1,2}. ¹Department of Sustainable Crop Production (DI.PRO.VE.S.), Università Cattolica del Sacro Cuore, 29100 Piacenza, Italy; ²Research Center on Plant Health Modelling (PHeM), Università Cattolica del Sacro Cuore, 29100 Piacenza, Italy. E-MAIL: giorgia.fedele@unicatt.it

Sour rot (SR) is one of the late-season, non-Botrytis, rots affecting grapevines, which can cause severe yield losses and relevant decay for wine making. A model framework was designed for describing the complex relationships between the following: i) development of grapevine bunches and changes in SR susceptibility; ii) occurrence, growth and pathogenicity of causal microorganisms (specifically, yeasts and acetic acid bacteria, AAB); iii) the vectoring activity of the *Drosophila* flies; and iv) the environmental conditions. The ultimate aim was to predict SR epidemics so that to inform disease management actions. Since several aspects of SR etiology, epidemiology and ecology are still not completely understood, a series of research activities were carried out for model parametrization. First, a characterization of the microbiota in both healthy and affected bunches was conducted across different bioclimates, to identify microorganisms associated with SR-infected bunches, their changes in response to bioclimatic conditions, and their relationship with the *Drosophila* flies. Then, nine yeasts and two AAB species were selected and characterized for their temperature requirements for growth on a liquid substrate that mimicked the chemical composition of ripened grape berries, in the range of 5 to 40°C in. Finally, the pathogenicity of the selected species was tested on grape berries through single and sequential inoculations. Further research is planned to study the seasonal micro-

biome dynamics of healthy and SR-affected berries, and of *Drosophila* spp. flies, to better parameterize the role of flies on disease development.

POSTER PRESENTATIONS

Importance of pathosystems' knowledge and process-based modelling approach in developing decision support systems for optimal crop management. S.E. LEGLER, E. GONZALEZ-DOMINGUEZ. *Horta srl, Piacenza, IT 29122, Italy.* E-MAIL: s.legler@horta-srl.com

Decision Support Systems (DSS) in agriculture are digital tools that can integrate different technologies to assist farmers, agricultural professionals, and decision-makers in taking informed decisions. These systems use data, models, and algorithms to provide insights, predictions, and recommendations that can optimize agricultural practices. The goal is to improve productivity, resource management, sustainability, and profitability in farming operations. Although several alternatives are available for agricultural stakeholders, adoption is still low, with the main obstacle being convincing farmers that a DSS can address their specific needs, even if it has been developed in a different country/context. Horta's DSSs are based on key aspects that enable them to address the challenges of demonstrating DSS robustness to farmers, to enter new contexts (countries/environments/crops) or to respond to climate change. These key aspects are: i) comprehensive agronomic and pathological knowledge, supported by continuous yearly field and controlled-environmental trials vs. machine learning data analysis, ii) mechanistic process-based vs. empirical data-based modelling, and iii) flexibility/customization vs. standard black-box solutions. Thanks to these features, Horta's DSS are now used by thousands of farmers and advisors, as well as agri-food industries on 16 crops in 12 countries. Two case studies are of particular interest to demonstrate how Horta's deep understanding of pathosystems knowledge and process-based models can help a DSS in territorial wide-spreading and in facing new cultivation contexts: the grape DSS used from Prosecco, to Champagne, to Napa Valley region, and the tomato DSS used by Italian processed tomato industry to Spanish greenhouse cherry tomato growers.

Postgraduate in agricultural sciences: challenges and opportunities in Egypt. A. HUSSEIN¹, Y. AHMED².

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Although Egypt has experienced a significant increase in tertiary education enrolment, the number of postgraduate students in agricultural sciences has seen considerable decline. This study aimed to discuss the challenges facing students in pursuing postgraduate studies. Data were collected from 206 students/graduates from 21 universities/institutes through questionnaire and in-person interviews. Less than half of the surveyed students and graduates have decided to pursue postgraduate studies. Although 86% of the participants were from agriculture and biological sciences backgrounds, medical applications emerged as the top choice for postgraduate research (65%), followed by agricultural applications (49%). The main motivation was the influence of the curricula taught during the undergraduate studies, then assumptions about higher income, better media coverage, publication in higher-impact journals, and access to grants. The students/graduates identified the main challenges they face include scientific thinking, foreign languages, writing resumes and cover letters, academic writing, and finding information online. A large portion of the sample indicated that undergraduate education has not efficiently prepared them to address those challenges. Many students/graduates sought advice from university professors, however, 54% evaluated it as partially useful, and 9% found it was not useful. Undergraduate agricultural education in Egypt needs to undergo reforms to attract higher-caliber students to pursue postgraduate studies and careers in agricultural research. Research and development (R&D) should be integrated into the undergraduate syllabus, and universities should establish specialized support units beyond the academic education. Furthermore, online platforms are essential in the era of digital information to enhance access to information and learning opportunities.

Machine Learning-Based Analysis of Reflectance Spectra in Eggplants under Biotic and Abiotic Stresses. C. TSELIOS^{1,2}, E. MANIATI², T. MPATZAKA³, I. RAPTIS³, A. KALOGERAS¹, D. ALEXANDROPOULOS^{1,2} and D. KIZIS⁴. ¹Industrial Systems Institute, Athena Research Center, Patras Science Park Building Platani, 26504 Patras, Greece. ²Department of Materials Science, University of Patras, 26504 Rion, Greece. ³ThetaMetris S.A., 12132 Athens, Greece. ⁴Laboratory of Mycology,

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This study investigates the use of reflectance spectroscopy combined with machine learning to classify eggplant spectral profiles in response to different stress treatments. Four plant-treatment groups were generated: control (C), water-stressed (S), *Verticillium dahliae* infected (V), and combined stress (VS). Two experiments were conducted: the first involved groups C, S, and V, while the second included all four groups. Reflectance spectra were collected over multiple measurement days, with repeated sampling across leaf area to monitor stress progression. A machine learning pipeline was developed using Savitzky-Golay filtering for noise reduction and principal component analysis (PCA) to extract dominant spectral features. The extracted features were used to train a Random Forest classifier to distinguish plant groups. Model performance was evaluated with 80–20 train-test splits and assessed using accuracy and confusion matrix metrics. In the first experiment, high classification accuracy was observed during early-to-middle measurement days, supported by confusion matrices showing clear class separation. In the later days, classification accuracy decreased to 70–80%, likely due to overlapping spectral signatures at advanced stages; although tissue damage was visible, spectral differences between stress conditions became less distinguishable. Earlier measurements captured physiological changes prior to visible symptoms, enabling more reliable classification. In the second experiment, including the VS group, accuracy remained consistently high (81–88%), demonstrating robust spectral differentiation. These findings support the sensitivity of reflectance spectroscopy for detection of early plant responses to stress and emphasize the value of temporal monitoring for non-invasive plant diagnostics, particularly during the onset of stress when intervention remains effective.

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Integrating digital tools for plant health: use case pilots of STELLA pest surveillance system in Greece. S. DIMITROPOULOU, D. TSITSIGIANNIS. *Department of Crop Science, Laboratory of Plant Pathology Agricultural University of Athens, Greece.* E-MAIL: dimtsi@aua.gr

The EU-funded STELLA project is developing an integrated Pest Surveillance System (STELLA PSS) aimed at enabling early warning, detection, and response to quarantine and regulated non-quarantine plant pathogens (RNQPs). By combining advanced sensing technologies, Internet of Things (IoT) devices, and artificial intelligence (AI)-driven analytics, STELLA addresses key limitations of conventional pest monitoring methods, such as delayed detection, restricted accessibility in remote areas, and high resource demands. In Greece, STELLA PSS is being implemented across two Use Case Pilots targeting *Ceratocystis platani* - a quarantine fungal pathogen responsible for canker stain in *Platanus orientalis* - as well as two RNQPs: *Verticillium dahliae* and *Pseudomonas savastanoi* pv. *savastanoi*, causal agents of Verticillium wilt and Olive knot disease, respectively. Pilot deployments are situated in the Kireas riparian forest on Evia Island and selected olive groves in the Attiki and Attiki regions, providing real-world assessment under both natural ecosystems and commercial cultivation conditions. The system leverages proximal (EDEN Viewer) and remote sensing (UAVs and satellite imagery), *in situ* IoT sensors, and spatiotemporal modelling to monitor plant host health, environmental parameters, and pest risk dynamics in real-time. This digital surveillance framework improves spatial resolution, expedites diagnosis, and lowers the cost of monitoring efforts. Furthermore, stakeholder engagement is fostered through mobile applications and citizen science initiatives, improving data collection and raising public awareness. STELLA PSS presents a scalable, science-based solution for proactive pest management, fully aligned with EU plant health policies.

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Remote sensing technology for grape health status monitoring under nursery conditions. F. DALENA, D. CORNACCHIA, R. CORONELLI, G. INCAMPO, F. MANNERUCCI, A. FARELLA, G. POPEO, S. PASCUZZI, S. POLLASTRO, F. FARETRA, D. GERIN. *Department of Soil, Plant and Food Sciences, University of Bari, Via G. Amendola, 165A/ 70126 Bari, Italy.* E-MAIL: stefania.pollastro@uniba.it

Remote sensing applications are becoming a valuable tool for crop production, including the viticulture sector. Different studies report the use of unmanned aerial vehicles (UAVs) for providing grapevine vigour indexes

related to the canopy shape and showing relationships with plant physiology and productivity. With this study, remote sensing UAV-based technology was applied under grape nursery condition on a mother plant field and on a rootstock production field, to evaluate possible relationships with grapevine trunk diseases. In the mother plant field, the 1103 Paulsen and Kober 5BB varieties were grown, while rootstock field was planted with Kober 5BB, 140 Ruggieri and 1103 Paulsen varieties. The NDVI data collected by DJI Mavic 3M, (DJI Sciences and Technologies Ltd, Shenzhen e Pechino, Cina) and analysed by using JMicrovision software (jmicrovision.github.io), allowed to follow the canopy development, along June-August months. In the mother plant field, the presence of weeds caused an overestimation of the NDVI values for grapevines and further AI-based analysis are currently being developed. In the rootstock production field, a lower NDVI index were observed for Kober 5BB compared to 140 Ruggieri and 1103 Paulsen. NDVI data were positively correlated $r \geq 0.7$ with the percentage of un-sprouted grapevine cuttings. These latter show internal wood browning symptoms whose fungal species of the Botryosphaeriaceae family, and belonging to the genera *Diaporthe*, *Pestalotia*, *Fusarium*, and *Dematophora* were isolated. In conclusion, the use of remote sensing represents a promising methodology for monitoring young vine health under nursery condition.

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New frontiers in plant pathology: innovative and cutting-edge tools to detect and manage plant pathogens. F. SAID¹, H. ALI², M.H. KHAN³. ¹Department of Entomology, Abdul Wali Khan University, Mardan, Pakistan. ²Entomology Section, Agricultural Research Institute Peshawar, Pakistan. ³Plant Protection Division, Nuclear Institute for Food & Agriculture, Peshawar. E-MAIL: dr.fazal@awkum.edu.pk

Plant pathogens have long been associated with different plant diseases across the globe and pose threats to agricultural commodities, enhancing food insecurity, crop yield both in terms of quality as well as quantity. For the sake of food security and improved crop yield, it is essential to address such issues and for that timely detec-

tion of plant pathogen and management is indispensable enabling us ensure sustainable food production. The current case study focuses on cutting-edge innovations and new frontiers in the field of plant pathology and integrated disease management. We explore the development and application of molecular diagnostics, remote sensing technologies, and machine learning algorithms for early and accurate disease detection. Additionally, we discuss advances in disease management, including bio-control agents, resistance breeding, and precision agriculture practices. Case studies demonstrating the successful implementation of these technologies and strategies are presented, showcasing their impact on reducing disease incidence and improving crop health. Finally, we address the challenges and future directions in plant pathology research, emphasizing the need for multidisciplinary approaches and enhanced collaboration between researchers, farmers, and policymakers.

CONCURRENT SESSION H2 – Mycotoxins and climate change: management strategies to mitigate their impact on food safety

SESSION KEYNOTES

Epidemiology and Biodiversity of mycotoxigenic fungi: a constantly evolving world. A. MORETTI. *Research National Council of Italy, Institute of Sciences of Food Production, CNR-ISPA, Bari, Italy.* E-MAIL: antonio.moretti@cnr.it

Mycotoxins, secondary metabolites produced by toxigenic fungi, are natural contaminant of food and feed and represent a serious threat worldwide, because of their impact on animal, human and plant health. According to a recent report, around 25% of food is contaminated by at least one mycotoxin, with important implications for the health of humans and animals and the global trade and economy. Hot and dry weather conditions, prolonged periods of rainfall deficits, and tighter supplies and market uncertainty have contributed in the last years to reduce agrifood production and increase the risk of mycotoxin contamination worldwide. Therefore, mycotoxin management requests advanced strategies along the whole supply chain, from pre- to post-harvest stages. Among the toxigenic fungi that produce mycotoxins *in planta*, the species belonging to *Alternaria*, *Aspergillus*, *Fusarium* and *Penicillium* genera are the most common, showing a great variability of their mycotoxin profiles, even in species phylogenetically closely related. In addition, such variability can be detected also at an

intraspecific level. The results of ISPA team studies, carried out in cooperation with other research institutions, for several species belonging to the above-mentioned genera, and conducted through metabolomics analyses and Whole Genome Sequencing approach, are here reported. In particular, we will report the variability of Aflatoxins, Ochratoxin A and Fumonisin B2 (FB2) production related to the occurrence of gene clusters (*afla*, *ota* and *fum*), in *Aspergillus* spp, where both the intact and deleted gene clusters co-exist, variability of FBs and Beauvericin in *Fusarium* where toxigenic potential can be related to Single Nucleotide Polymorphism occurrence, variability of trichothecenes production in the *Fusarium* where a different distribution among the TRI genes cluster has been detected. Taken together, these data show that mycotoxin gene clusters can dramatically differ within a single species or among very closely related species and the lack of a given mycotoxin production, at least *in vitro* conditions, is frequently, but not always, related to the absence of related gene clusters. Due to the examples of genetic variants in several toxigenic fungi with respect to mycotoxin gene clusters, we need continuous epidemic studies from different crops and geographic areas by using a consistent and statistically significant approach to generate data that better would reflect the current dynamics of species profile and their genetic variability, also in the perspective of the current climate change scenario. Several results of our epidemiological studies for all the four *Alternaria*, *Aspergillus*, *Fusarium* and *Penicillium* genera will be provided. In addition, a further current challenge should be to use genomic approach to better understand the relationships between genotyping and phenotyping for pathogenicity, mycotoxin profile and stress related genes, in order also to better understand how the genetic plasticity of the toxigenic fungi can be a precious tool for their adaptation to the current climate changes.

Biological detoxification of mycotoxins. CIMBALO, P. VILA-DONAT, A. MORENO, C. LUZ, S. RAFAI, L. ANTELO, G. MECA. *Biotech Agrifood, Faculty of Pharmacy and Food Sciences, Universitat de València, Avda. Vicent Andrés 6 Estellés s/n, 46100, Burjassot, Spain.* E-MAIL: alessandra.cimbalo@uv.es

Nowadays, biologically based detoxification strategies are attracting great interest in replacing conventional methods for counteracting highly toxic compounds in contaminated food products. Among these compounds, Aflatoxin B1 (AFB1) and Ochratoxin A (OTA) are two major mycotoxins known for their harmful effects on

human and animal health, as well as for their persistence in the body once ingested. Following this line, the aim of this study was to evaluate the detoxification potential of bacterial strains belonging to the genera *Lactobacillus* (n=10) and *Bacillus* (n=10) on these two mycotoxins, using naturally contaminated corn (AFB1) and barley (OTA) flour as the substrate. For this purpose, the selected strains were grown in their culture medium by inoculating 1% of bacterial suspension and then incubated for 12, 24 and 48h. At each time point, culture was withdrawn, diluted, and filtered prior for chromatographic analysis. All samples were processed and analyzed in duplicate to evaluate AFB1 and OTA detoxification efficiency. Following strain selection, an *in vitro* simulated human digestion was performed to assess detoxification potential under real gastrointestinal conditions, including oral, gastric, duodenal, and colonic phases. Mycotoxin quantification and metabolite detection was performed using a Liquid Chromatography system coupled with Quadrupole Time-of-Flight Mass Spectrometry (LC-QTOF-MS) operating in targeted MS/MS mode. Based on the results, the best AFB1 detoxification rates were observed in *Lactobacillus curvatus* 14 ($41.1 \pm 19.3\%$) and *Pediococcus pentosaceus* 4 ($25.4 \pm 11.3\%$) after 48 h, and *Bacillus firmus* 6 ($25.1 \pm 12.9\%$) after 24 h. Furthermore, metabolite analysis revealed that both lactic acid bacteria and *Bacillus* strains were capable of transforming AFB1 into various degradation products, with aflatoxicol being the most consistently detected metabolite across all strains and time points, at concentrations ranging from 31.00 to 55.40 $\mu\text{g L}^{-1}$. *Lactobacillus curvatus* 14 and *Bacillus siamensis* 4 exhibited the highest aflatoxicol levels, peaking at 47.45 $\mu\text{g L}^{-1}$ and 55.40 $\mu\text{g L}^{-1}$, respectively, at 48 h and 24 h. In addition, *L. curvatus* 14 and *Pediococcus pentosaceus* 4 produced aflatoxin D1, while *P. pentosaceus* 29 was the only strain to generate aflatoxin Q1. Among the *Bacillus* strains, *B. siamensis* 4, *B. firmus* 6, and *B. velezensis* 9 showed a broader metabolic profile, including aflatoxin D1, P2, B2a, and Q1, suggesting multiple biotransformation pathways. In contrast, some strains such as *B. licheniformis* 10 and *B. amyloliquefaciens* 20 primarily produced aflatoxicol, with no other detectable metabolites, possibly indicating more limited degradation mechanisms. On the other hand, the simulated digestion model showed strain-dependent AFB1 detoxification. *L. curvatus* 14 and *P. pentosaceus* 4 significantly reduced AFB1 in gastric and duodenal phases, while *firmus* 6 was less effective. In the colonic phase, AFB1 levels decreased over time even in the control, suggesting non-microbial degradation may also occur. For OTA, the most effective strains at 48 h were *P. pentosaceus* 4 ($59.22 \pm 6.78\%$), *Lactobacillus plan-*

tarum 5 ($70.00 \pm 7.00\%$), and *L. fermentum* 8 ($67.02 \pm 9.23\%$). Significant reductions were observed for after the colonic phase, reaching $72.26 \pm 7.54\%$ for *P. pentosaceus* 4 and $69.67 \pm 9.70\%$ for *L. curvatus* 14. These findings highlight the potential application of selected strains in biological detoxification strategies for reducing dietary AFB1 and OTA exposure.

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ORAL PRESENTATIONS

Dynamics of *Aspergillus* spp. section *Flavi* in Spanish dried figs: field assessment. Z. JANFI¹, M.T. GARCIA-LOPEZ², F.A. GUERRERO-PAEZ¹, M.A. ROMERO¹, C.M. DIEZ¹, J. MORAL¹. ¹UCOLIVO. Department of Agronomy. María de Maeztu Unit of Excellence, University of Cordoba, Cordoba, Spain. ²AgroPhenoLab, Institute for Sustainable Agriculture-Spanish National Research Council (IAS-CSIC), Cordoba, Spain. E-MAIL: tgarcialopez@ias.csic.es

Fig cultivation (*Ficus carica*) represents a significant agricultural sector in Spain, yielding approximately 59,900 tons annually. A persistent challenge facing dried fig producers is contamination with aflatoxins—potent mycotoxins synthesized by members of *Aspergillus* section *Flavi*. Despite their economic importance, critical knowledge gaps remain regarding contamination pathways and risk factors in fig production systems. This research investigated *Aspergillus* section *Flavi* population dynamics in Spanish fig orchards during the 2022-2023 growing seasons. We systematically monitored fungal presence in orchard soils, tree-hanging fruits, and ground-fallen figs, while quantifying airborne spore concentrations using volumetric air samplers deployed in both field and processing environments. Our findings revealed significantly higher *Flavi* colonization in ground-fallen figs compared to tree-harvested fruits (10% vs. 4.2% in 2022; 35.7% vs. 14.2% in 2023). Soil inoculum densities averaged 25.7 CFU/g in 2022 (range: 5.0-140.1) and 27.9 CFU/g in 2023 (range: 5.2-395.5). Airborne *Aspergillus* spores were detected throughout the summer months (mean: 0.04 CFU/m³), with concentration peaks in late August (maximum: 2.43 CFU/m³). Notably, *Aspergillus* spore density in cold storage facilities remained minimal (0.001 CFU/m³), substantially lower than other fungal genera including *Penicillium* (0.02 CFU/m³) and *Alternaria* (0.01 CFU/m³). These

results demonstrate that soil contact represents the primary contamination pathway for aflatoxigenic fungi in fig production. Implementation of agronomic practices that minimize fruit-soil contact, such as improved harvesting techniques and orchard floor management, could significantly reduce aflatoxin risk in Spanish dried figs.

This research was financially supported by the “HEPATOCARCINOGENIC FUNGI IN DRIED FIGS” CDTI Spanish project and the “Agrupación de Cooperativas del Valle del Jerte” company. M. Teresa García López’s contract is part of grant JDC2022-048362-I, from MICIU/AEI/10.13039/501100011033 and the European Union NextGenerationEU/PRTR.

Effect of ozone as an alternative treatment of *Aspergillus* contamination in pistachios. M. ABUKHMAISH¹, W. MELLIKECHE¹, A. RICELLI², M. GALLO¹, A.M. D’ONGHIA¹, G. COLELLI³. ¹International Centre for Advanced Mediterranean Agronomic Studies, Via Ceglie, 9-70010, Valenzano (BA), Italy. ²National Research Council - Institute of Molecular Biology and Pathology, P.le A. Moro, 5-00185, Rome, Italy. ³Department of Agricultural Sciences, Food, Natural Resources and Engineering, University of Foggia, Via Napoli, 25-71122, Foggia, Italy. E-MAIL: marahabukhmaish@gmail.com

Toxigenic *Aspergillus* molds pose a major challenge to the commercialization and export of pistachio nuts. Therefore, it is essential to develop effective postharvest strategies to control these contaminations, particularly with increasing restrictions on chemical pesticides. Ozone has emerged as a promising alternative treatment due to its powerful oxidizing properties and broad-spectrum antimicrobial activity. It has been previously tested on different food commodities and proven effective in reducing fungal contamination and mycotoxin content. This study explores the efficacy of ozone in reducing *Aspergillus* contamination by applying two doses (20 ppm and 13 ppm) for six hours on in-shell and shelled pistachios. The antimicrobial effect was evaluated by monitoring *Aspergillus* spp. occurrence at fixed intervals over six months of storage. Additionally, the impact of ozone treatment on the color and peroxide content of pistachios was assessed. Results showed that ozone treatments significantly reduced fungal contamination, with the higher dose of 20 ppm resulting in up to a 90% reduction in in-shell pistachios and 91.5% reduction in shelled pistachios. This reduction remained significant throughout long storage, with consistent results obtained 3 and 6 months after the treatment. Importantly, ozone had no significant effect on the color and peroxide levels of treated pistachios. These findings show that ozone

is a highly effective and safe postharvest treatment that reduces fungal contamination without compromising the quality of pistachios, offering a valuable solution for enhancing food safety during storage.

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First report of the relationship between *Drosophila suzukii* and mycotoxigenic fungi on blueberries. N. BASER¹, A. BEN HASSINE¹, S. KHEMIES¹, W. MEL-LIKECHE¹, A. RICELLI², A.M. D'ONGHIA¹. ¹*International Centre for Advanced Mediterranean Agronomic Studies (CIHEAM IAMB), Bari, Italy.* ²*CNR-Istituto di Biologia e Patologia Molecolari, Rome, Italy.* E-MAIL: baser@iamb.it

Drosophila suzukii is a ubiquitous pest insect which attacks a wide range of soft and stone fruits including blueberries (*Vaccinium corymbosum* L.). Adult insects can lay eggs deep inside healthy fruits inducing their deterioration. In addition to its direct damage, *D. suzukii* can also cause secondary infections by post-harvest fungi. This study was carried out on blueberries produced in Italy to determine their mycobiota. It revealed a high presence of diverse genera of fungi already reported in blueberry, including *Alternaria*, *Botrytis*, *Colletotrichum*, *Aspergillus*, and *Penicillium*. Based on these results, the vectoring relationship of *D. suzukii* with toxigenic fungal contaminants was studied on blueberries. It was revealed that indeed the insect is capable of spreading fungal spores among blueberries. Sterilized fruits infested with the insect showed a high occurrence of fungi compared to non-infested sterilized fruit. The fungal species transmitted by *D. suzukii* were mainly *Alternaria alternata* and *Aspergillus niger*, both known for their production of mycotoxins. HPLC analysis, aiming to detect any mycotoxin production of isolated fungi, revealed that several *A. alternata* isolates, transmitted by *D. suzukii*, were able to produce alternariol and tenuazonic acid mycotoxins.

Impact of agroforestry systems on fungal populations in wheat crops. E. FORGUES, M. FOULONGNE-ORIOLO, L. CARLES. *UR1264 Mycology and Food Safety (MycSA), INRAE, 71 Avenue Edouard Bourlaux, 33883 Villenave d'Ornon.* E-MAIL: elise.forgues@inrae.fr

As new agroecological practices like temperate agroforestry emerge, the issue of biological regulation, particu-

larly regarding plant pathogens within these systems is poorly documented. It is essential to gain knowledge of the benefits and potential risks associated with the deployment of such a practice applied to field crops. The aim of this study is to investigate the impact of the agroforestry system associated with wheat on fungal populations and in particular on the pathogens responsible for Fusarium Head Blight (FHB). FHB is one of the major diseases affecting cereals including wheat. A cohort of several *Fusarium* spp. are associated with FHB symptoms. While *F. graminearum* is often prevalent, the spectrum of other *Fusarium* species is strongly influenced by the environment. In this context, the presence of hedges or trees in intra-parcel wheat fields may modify fungal populations, by acting as pathogens reservoirs, or by modifying the microclimatic conditions favorable to their development. Using metabarcoding approaches targeting either ITS or EF1alpha gene regions, we will investigate the representativeness of fungal genera and the occurrence of *Fusarium* species in samples collected in the wild compartment (leaves, wild grasses) and in wheat fields (heads) during the growing season. Preliminary results from the first year of sampling on the spatio-temporal dynamics of fungal populations will be presented. Our study will contribute to fuel scientific knowledge on FHB epidemiology in agroforestry landscapes and will also provide broader insights into potential non-intended effects relevant to disease management strategies in diverse cropping environments.

POSTER PRESENTATIONS

Temporal dynamics of *Fusarium* spp. on wheats and possible relationship with their ecological requirements. M-A. GARCIA^{1,3}, R. MAHMOUD², M-O. BANCAL³, P. BANCAL³, A. ROUCOU⁴, R. VALADE⁴, S. BERNILLON¹, F. RICHARD-FORGET¹, M. FOULONGNE-ORIOLO¹. ¹*INRAE, Mycology and Food Safety (MycSA), 33140 Villenave d'Ornon, France.* ²*Institut Agro, Univ Rennes, CNRS, IRMAR-UMR 6625, 35042 Rennes, France.* ³*Université Paris-Saclay, INRAE, AgroParisTech, EcoSys, 91120 Palaiseau, France.* ⁴*ARVALIS Institut du Végétal, Boigneville, France.* E-MAIL: marie-anne.garcia@inrae.fr

Fusarium Head Blight (FHB) is one of the most devastating fungal diseases affecting wheat worldwide. This disease is caused by a complex of different fungal species, belonging to the *Fusarium* genus, that coexist within the same host plant. Beyond the significant impact on yield due to infection, FHB also alters grain qual-

ity, leading to economic losses, due to contamination by harmful mycotoxin produced by *Fusarium* species. Global changes, including agronomic and climatic factors, are known to affect the distribution of *Fusarium* species infecting wheat and the associated mycotoxins. Studying the evolution of *Fusarium* species in a changing environment is essential for predicting and controlling FHB, and therefore, ensuring the safety of cereals products. The purpose of this study was to understand *Fusarium* species dynamics and their adaptive potential in the context of climate change. The diversity of *Fusarium* species in French wheat fields has been studied over the last 15 years using a metabarcoding approach targeting the *EF1a* gene, together with mycotoxins profiling. A large database gathering numerous metavariables was available, enabling the study of the impact of agronomic and climatic factors on *Fusarium* diversity on wheat fields. Differences in *Fusarium* predominance were observed, suggesting distinct ecophysiological behaviour between species in response to environmental factors. Therefore, growth and mycotoxin production were *in vitro* analysed in response to combined temperature and water activity variations. Our findings will contribute to improving mycotoxin risk prediction models, thereby supporting the development of effective strategies for managing future mycotoxin threats in wheat production.

Preventing food-borne threats at the source: a one health approach to crop and animal biosecurity. F. ALI¹, M. SADIQ², I. AHMAD². ¹*Pet Street Veterinary Clinic Abu Dhabi UAE.* ²*University of Agriculture Peshawar Pakistan.* E-MAIL: drfarzand_2011@yahoo.com

Ensuring food safety begins long before animal production starts in the field, with healthy plants. As a veterinarian, the link between plant health and animal health is clear: contaminated crops can lead to unsafe feed, compromise animal health, and ultimately impact human food safety. Recent advances in plant pathology offer new tools for detecting and controlling plant diseases that threaten the safety and quality of crops used for animal feed and the broader food chain. By integrating veterinary and plant sciences, we can develop more effective strategies for preventing foodborne illness, reducing exposure to toxins (such as mycotoxins), and enhancing biosecurity. Strengthening education and collaboration across disciplines is essential to training professionals capable of addressing these challenges comprehensively. This approach supports a safer, more sustainable food system, one where plants, animals, and human health are protected together.

Evaluation of biocontrol agents for mitigation of aflatoxin contamination in pistachios. V. BARTZIS¹, M. VARVERI¹, S. DIMITROPOULOU¹, C. KAFTANI², M.D. KAMINIARIS¹, M. GEORGIADOU², C. PAPPAS², D.I. TSITSIGIANNIS¹. ¹*Department of Crop Science, Laboratory of Plant Pathology Agricultural University of Athens, Greece,* ²*Department of Food Science and Human Nutrition, Laboratory of Chemistry Agricultural University of Athens, Greece.* E-MAIL: dimtsi@aua.gr

Pistachios, one of the most economically significant nuts globally, are particularly susceptible to fungal infections that produce aflatoxins. Aflatoxins are a major group of mycotoxins known for their teratogenic and carcinogenic effects on humans and animals. Among various evaluated mitigation strategies, biological control methods have demonstrated the highest efficacy in reducing aflatoxin contamination. The aim of this study was to evaluate the ability of two yeast strains of *Aureobasidium pullulans* and *Meyerozyma caribbica*, one bacterial strain of *Pantoea agglomerans* and one non-toxigenic strain of *Aspergillus flavus* to reduce aflatoxin contamination levels in pistachio orchards in Greece. These microbial strains were originally isolated epiphytically from pistachios. Previous studies have demonstrated that these isolates can effectively suppress aflatoxin biosynthesis in pistachio fields on the island of Aegina. For the purpose of this study, the strains were applied to two distinct pistachio orchards: a commercial orchard located on Aegina Island and an experimental orchard at the Agricultural University of Athens. Analysis of harvested pistachio kernels demonstrated that aflatoxin formation was significantly inhibited by all four strains, with reductions ranging from 86% to 99%. These results align with earlier studies and further support the potential of these strains in developing biocontrol products for the effective management of aflatoxins in pistachio production.

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Study of *Drosophila suzukii*'s role in the transmission of postharvest fungal contaminants on blueberries and the hypothesis of their control by ozone. S. KHEMIES¹, W. MELLIKECHE¹, A. RICELLI², N. BASER¹. ¹*International Centre for Advanced Mediterranean Agronomic Studies (CIHEAM IAMB), Bari, Italy.* ²*CNR-Istituto di*

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Drosophila suzukii (Matsumura; *Diptera: Drosophilidae*), commonly known as the Spotted Wing *Drosophila* (SWD), is an invasive pest native to temperate Asia that has recently spread to North America and Europe. Unlike typical vinegar flies, *D. suzukii* females possess a characteristic serrated ovipositor, allowing them to lay eggs in ripening fruits, including blueberries (*Vaccinium corymbosum* L.), which leads to significant economic losses and reduced fruit quality. Beyond direct damage, *D. suzukii* also facilitates the transmission of postharvest fungal pathogens, increasing the risk of mycotoxin contamination and posing serious food safety concerns. In this study, we aimed to investigate the fungal postharvest pathogens associated with *D. suzukii*. Fungal isolations were performed from both wild-caught and laboratory-reared insects to explore the possibility that spore transmission occurs through physical contact between the insect and the fruit. The isolated fungi were identified using molecular tools, including PCR and LAMP. Given the predominant presence of *Aspergilli* spp., their possible ability to produce ochratoxin A (OTA) and aflatoxin B₁ was investigated using High-Performance Thin-Layer Chromatography (HPTLC). We also conducted fungal isolations during different periods of the year to monitor potential variations in fungal associations. Furthermore, we evaluated the potential of ozone treatment as a postharvest strategy to minimize fungal contamination on blueberry. Our results showed that some of the fungal isolates are able to produce OTA, and ozone treatment at 5 ppm for 48 hours significantly reduced fungal growth and sporulation, as well as the egg hatchability of *D. suzukii* on blueberries without causing damage to the texture of the fruit.

***Aspergillus flavus*-based product for biological control of aflatoxins in corn.** V. BOSCO, F. BOVE, J. MATHIEU, O. GURKAN, S. LAMPERTI. *Corteva Agriscience Italia – Via dei Comizi Agrari 10 26100 Cremona (CR).* E-MAIL: federica.bove@corteva.com

Aflatoxins are highly toxic secondary metabolites mainly produced by *Aspergillus flavus*, contaminating diverse crops including corn, peanuts, cottonseed, pistachios and other tree nuts, as well as various cereals and legumes worldwide and posing serious health risks to humans and animals. Climate change is increasing the prevalence of aflatoxin contamination in regions like Central and Southern Europe beyond the tradition-

ally subtropical regions. Effective control strategies are urgently needed, especially for economically important crops like corn. AF-X1 is an innovative biocontrol product developed by Università Cattolica del Sacro Cuore and Corteva Agriscience, specifically for corn protection. It utilizes an atoxigenic *A. flavus* strain (MUCL54911) isolated from Italian soils, which is genetically characterized by a large deletion in chromosome 3, resulting in the complete absence of the aflatoxin gene cluster and inability to produce cyclopiazonic acid, providing dual mycotoxin control. AF-X1 is formulated as sterilized sorghum seeds coated with MUCL54911 spores (1.0×10^5 CFU g⁻¹), applied at 25 kg ha⁻¹ during corn stem elongation (BBCH 30-39). Field trials across Italy (2013–2019) demonstrated that AF-X1 reduced aflatoxin B₁ by 86.4% and total aflatoxins by 87.3% compared to untreated controls, consistently keeping contamination below regulatory limits without affecting yield or quality. AF-X1 represents a sustainable and effective solution for managing aflatoxins in corn.

A patented bioactive formulation supporting SDG-Based food safety: hydroxytyrosol acetate as a natural antifungal agent. D. BOUZID¹, S. MERZOUKI², M.E. MERZOUKI³, M.M. ZERROUG². ¹Laboratory of Applied Microbiology, Faculty of Natural and Life Sciences, Ferhat Abbas University Setif 1, 19000 Sétif, Algeria. ²Private Medical Clinic, Sétif, Algeria. ³Department of Science and Technology, Faculty of Science and Technology, Optics and Precision Mechanics, Ferhat Abbas University Setif 1, 19000 Sétif, Algeria. E-MAIL: bouزيد.djihane@yahoo.fr

Mycotoxins and fungal spoilage remain major threats to food safety worldwide, especially in fruit and cereal supply chains. In response, we present a patented method to produce a natural antifungal formulation enriched in hydroxytyrosol acetate, derived from extra virgin olive oil via controlled oxidative transformation. The ozonation process leads to the emergence of bioactive phenolic esters, including hydroxytyrosol acetate, which are absent in untreated oil. This compound showed strong *in vitro* inhibition of *Fusarium solani*, *Penicillium expansum*, and *Aspergillus niger*, as well as preventive effects on spoilage development in fresh produce under experimental conditions. Unlike synthetic fungicides, this formulation is food-grade, biodegradable, and solvent-free, making it suitable for post-harvest applications. The ability to combine natural preservation, regulatory compliance, and sustainability aligns with current food safety regulations. This innovation supports the Sustainable Development Goals (SDGs), particularly SDG 2 (Zero

Hunger) and SDG 12 (Responsible Consumption and Production), offering a scalable opportunity to reinforce food safety in storage and distribution.

CONCURRENT SESSION H3 – Certification programs counteract the introduction and spread of dangerous, harmful organisms. QVI - The Italian Experience

SESSION KEYNOTES

Certification of nursery plants to preserve the fruit tree industry and genetic resources in NENA region.

T. YASEEN. *Regional Plant Protection Officer, Regional Office of the Near East and North Africa, Food and Agriculture Organization of the United Nations (FAORNE)*. E-MAIL: Thaer.Yaseen@fao.org

The Near East and North Africa (NENA) region is home to a rich diversity of fruit tree species that are vital to food security, rural livelihoods, and the preservation of agricultural heritage. However, the sustainability of the fruit tree industry is increasingly threatened by the spread of transboundary plant pests and diseases, the loss of genetic diversity, and the widespread use of uncertified, low-quality nursery material. Climate change, expanded trade, and limited phytosanitary infrastructure further exacerbate these challenges. Therefore, the implementation of nursery plant certification schemes offers a sustainable solution to protect the fruit tree sector and safeguard the region's unique and vital genetic resources. Despite its importance, countries in the NENA region face significant constraints in establishing and managing effective certification systems. These include weak regulatory and institutional frameworks, limited diagnostic and inspection capacities, weak coordination among public and private sector stakeholders, and a shortage of trained personnel. Additionally, many farmers still rely on traditional propagation methods due to limited access to tested, certified materials. The situation is even more difficult in organic production systems, where certified organic propagative material may be unavailable. Furthermore, the lack of awareness and incentives for nurseries to participate in certification schemes remains a significant barrier. Nevertheless, some countries have made notable progress in developing and implementing effective models. Countries such as Morocco, Tunisia and Lebanon have demonstrated the potential of well-structured certification systems to enhance the quality and traceability of nursery plants, promoting the conservation of locally

fruit tree genotypes. In this context, the Food and Agriculture Organization of the United Nations (FAO) has played a pivotal role in supporting member countries across the NENA region by advocating for the development of nursery plant certification schemes, fostering regional collaboration, and formulating technical guidelines and protocols tailored to key crops such as date palm and olive. Building on these efforts, FAO recently launched the NENA Plant Health Strategy (2025–2035), which aims to strengthen regional plant health systems with a particular focus on plant certification. This initiative promotes the adoption of harmonized standards and certification protocols, enhances regulatory and diagnostic capacity, and strengthens institutional coordination. These actions are critical for enabling sustainable fruit production, improving market access, and preserving the region's agrobiodiversity. Moving forward, coordinated regional action and targeted investment in institutional and technical infrastructure are crucial to scaling up successful models and ensuring the production and dissemination of healthy, true-to-type nursery plants.

The Italian experience in the development and adoption of the voluntary certification protocol for the vines and fruit plant propagating material. B.C. FARAGLIA¹, O. CORBO². ¹Masaf – Central Phytosanitary Service - National voluntary certification service, Rome. ²CREA DC - Masaf – Central Phytosanitary Service - National voluntary certification service, Rome. E-MAIL: b.faraglia@masaf.gov.it

In Italy the qualification of fruit plant propagation materials through the implementation of a certification protocol was started at the regional level in 1980. In consideration of the interest shown by nurseries and fruit growers, voluntary certification of fruit species was envisaged at ministerial level in 1987. In 1991, the National Voluntary Certification Service was established and by 1993, the technical protocols for citrus, stone fruit, pome fruit, strawberry, olive and walnut were defined. With the introduction of EU mandatory rules as the CAC (Conformitas Agraria Communitatis) quality standards starting in 1992, and those relating to quarantine and phytosanitary measures (the Plant Passport in 1993 and Regulation EU 2016/2031 on protective measures against plant pests), the national rules on voluntary certification have undergone numerous modifications. The implementing measures to this rule define the level for the qualification of the plant propagating material: CAC and Certified EU. They provide that individual EU member states may adopt additional measures

at the Certified EU level in order to better qualify and guarantee the health of the fruit plant propagation material. In 2021, Italy implemented and definitively adopted the national voluntary certification protocol called QVI (Qualità Vivaistica Italia), constituting a precise trade mark for this quality level. It covers all the species ruled, as well as kiwi and artichoke which are agamically propagated. The evolution of a 45-year legislative technical process for the highest qualification of plant propagation material produced by Italian nurseries is illustrated and discussed in this report.

The role of the Italian nursery association CIVI-Italia within the National Voluntary Certification Scheme. L. CATALANO, B. NOVELLI, G. CONSALVO. *CIVI-Italia, Corso Vittorio Emanuele II, 101, 00186 Rome, Italy.* E-MAIL: l.catalano@agrimeca.eu

CIVI (Interprofessional Centre for Nursery Activities) - Italia is the national consortium of nursery associations and producers' unions. Its main task is to qualify nursery production in support of the fruit, citrus, olive and vine production chains. It is the exclusive national association, officially representing the nursery sector, in promoting and implementing plant propagation certification programs in Italy. Since the introduction of voluntary certification schemes for fruit plant propagation material in Italy, a strong partnership has been established between public institutions and professional nursery associations. CIVI-Italia was officially recognized by the Italian Ministry of Agriculture through a Ministerial Decree in 1993, at the same time with the launch of the national voluntary certification system. With the implementation of the new national voluntary protocol, aligned with the reformed EU regulations and enacted by Ministerial Decree in June 2020, CIVI-Italia was appointed as the managing body of QVI – QUALITÀ VIVAISTICA ITALIA. This quality mark is used to identify and guarantee products that have been first certified as virus-free. The Italian experience demonstrates that certification of fruit plant propagating material has been carried out under the coordination and supervision of the public authorities - Ministry and Regional Plant Health Services, in close cooperation with the nursery associations. The key aspects of this collaboration between public authorities and the nursery association, including the responsibilities assigned, the roles undertaken, and the organization of activities, are presented and discussed with the overarching goal of qualifying Italian nursery production.

PLENARY CLOSING

KEYNOTE LECTURE

Plant Health – Food Safety – One Health. G. STANCANELLI. *Environment, Plants and Ecotoxicology (PLANTS) Unit, European Food Safety Authority (EFSA), Via Carlo Magno 1A, Parma (Italy).* E-MAIL: giuseppe.stancanelli@efsa.europa.eu

The European Food Safety Authority (EFSA) serves as the central scientific body providing risk assessments and evidence-based scientific advice on food and feed safety, animal health and plant health within the European Union (EU). EFSA's scientific outputs inform EU policy and regulatory frameworks, support national risk management authorities, and contribute to a harmonized, science-driven European food safety system. In the area of food safety, EFSA undertakes risk assessments on chemical and biological hazards across the food chain, including risk assessment for: pesticides; food additives; food flavourings; food enzymes; feed additives; food contact materials; contaminants in the food chain; genetically modified organisms; food claims; novel foods; microbiological risks in the food chain. In plant health, EFSA provides scientific advice on the risks posed by quarantine, new or emerging plant pests threatening the EU agriculture, forestry, landscape or biodiversity. EFSA plant health covers a broad range of activities: horizon scanning for new or emerging plant health threats; pest risk assessment; climate suitability assessment including climate change scenarios; modelling the introduction and spread of plant pests and the efficacy of phytosanitary measures; commodity risk assessment for High Risk Plants and derogation requests to provision of the EU Plant Health Law; support to early detection and surveillance of plant pests; databases on plant pests and plant commodities (e.g.: *Xylella* host plants database, Scolytinae database, apple pests database); research to reduce key knowledge gaps on new and emerging plant pests. EFSA communicates on the risks to food chain, animal and plant health by publishing open access its scientific outputs and by proactive risk communication strategies aiming at informing stakeholders and the general public. Various communication campaigns on food safety, plant health and animal health contribute to raising awareness among the EU citizens. EFSA, in cooperation with other EU agencies and international partners, aims to develop and apply a One Health risk assessment approach integrating human health, animal health, plant health and environmental health. This is evident in antimicrobial resistance

(AMR), where data collection on antimicrobials use and resistance need to cover human health, animal health and plant health. Other key issues addressed in a One Health risk assessment approach include zoonotic diseases and climate changes. Plant health is a major component of food security and environmental health, but it is also closely connected to food safety (in particular regarding pesticides and mycotoxins). Multidisciplinary risk assessment need to take into account these interdependencies, facilitating the integration of activities. For example, an integrated surveillance systems in border control points allow early detection of biological hazards that may affect animal health, plant health and human health. The One Health framework enables to align risk assessment with broader sustainability goals, while ensuring resilience in the face of climate change, biodiversity loss, and global trade changes.