



# Book of abstract

## 14<sup>th</sup> Conference of the European Foundation for Plant Pathology

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Editors: Anna Berlin and Georgios Tzelepis

**14th Conference of the European  
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# Future visions in plant pathology

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**In an ever-so fast changing world, it is more important than ever to uphold international dialogue and exchange of scientific ideas.**

International collaboration is needed to address the major challenges posed by climate change, globalization and non-sustainable use of natural resources. The theme of the 14th EFPP conference in Sweden, 'Plant Pathology in 2050: Visions for the Future', was chosen to address current and future challenges within plant pathology and plant protection. Importantly, scientific advances have resulted in many new opportunities in the form of new theory, methods and tools.

The 14<sup>th</sup> EFPP conference successfully captured the breadth of scientific progress in plant pathology during four intense days in Uppsala in June 2025. A total of 237 delegates registered to attend the conference, hailing from 23 different countries, and an additional 30 MSc students from the Swedish University of Agricultural Sciences were also given the opportunity to attend the sessions. The positive and enthusiastic support from the scientific community made the planning and execution of the conference very enjoyable. The scientific programme consisted of 14 invited speaker presentations, 48 oral presentations and 129 poster presentations organised into six plenary

sessions and eight parallel sessions. Two excursions were organised for one afternoon: one focusing on plant pathogens in agricultural crops and the other on forest health. The conference also included appreciated social activities including a welcoming reception at Gustavianum museum and a conference dinner at the Norrland nation in Uppsala. We would like to extend our sincere gratitude to the members of the scientific committee, to the sponsors, to the Swedish Society for Plant Pathology, and the local organizers. Special thanks should be given to the conference secretary, Dr. Anna Berlin and her assistant, Dr. Georgios Tzelepis. Finally, we would like to thank all participants that made the 14<sup>th</sup> EFPP meeting a success.

Finally, we would like to extend our wishes for a successful 15<sup>th</sup> EFPP meeting in Athens, Greece.

*Prof. Magnus Karlsson*

Chair of the scientific committee

*Prof. Emeritus Jonathan Yuen*

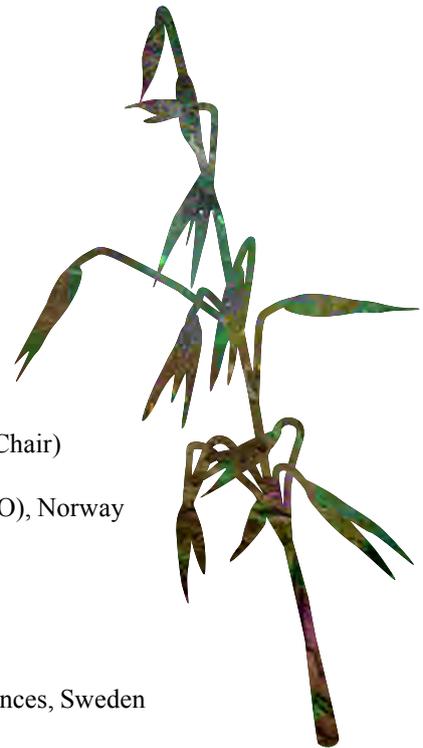
President, EFPP (2022-2025)



The 14<sup>th</sup> Conference of the European Foundation for Plant Pathology was arranged 2-5 June 2025 in Uppsala, Sweden.

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## Local organizers

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Anna Berlin, Meeting Secretary  
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## **Invited speakers**

## PS1.1 - Pathogens or biocontrol agents? The two faces of cyclic lipopeptide producing *Pseudomonas* strains

Monica Höfte<sup>1</sup>

Lu Zhou<sup>1</sup>, Bishnu Marahatta<sup>1</sup>, Jasmine De Rop<sup>1</sup>, Durga Prasad<sup>2</sup>, José Martins<sup>2</sup>

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Some *Pseudomonas* bacteria that belong to the *P. fluorescens* or *P. syringae* group produce cyclic lipopeptides (CLPs), bioactive compounds with biosurfactant activity which interact with other organisms and the plant. CLPs of the Mycin and Peptin family are always co-produced and show potent antifungal activity at low concentrations, but these compounds can also be involved in pathogenicity. *Pseudomonas* strains producing Mycin and Peptin-type CLPs cause plant diseases such as sheath brown rot on rice, midrib rot on lettuce, wheat leaf blight, stem necrosis on tomato, bacterial canker of trees, etc. The CLPs in these strains have a dual role, they cause necrosis on above-ground plant parts, but also protect the producer against competing microorganisms. Intriguingly, Mycin and Peptin producers have also been isolated from the rhizosphere or endosphere of plants where they behave as mutualists and protect the host against soilborne pathogens such as *Rhizoctonia* and *Pythium*. In many strains, CLP production is temperature dependent with high production at low temperatures. Regulation of CLP production is complicated and involves multiple LuxR type regulators that respond to environmental factors. Some strains are parasitic on above-ground plant parts, but beneficial in the rhizosphere where they show temperature-dependent biocontrol activity mediated by CLP production. Moreover, some *Rhizoctonia* strains that co-occur with CLP-producing *Pseudomonas* strains are able to inactivate the CLPs by enzymatic degradation. Our results show that CLP-producing *Pseudomonas* strains operate along a parasite-mutualist continuum that is affected by location in the host, environmental factors and competing microorganisms. The challenges to use these bacteria or their metabolites as bioprotectants will be discussed.

## PS1.2 - The global movement of fungal crop pathogens: perils, predictions and panacea?

Sarah Gurr<sup>1</sup>

<sup>1</sup> University of Exeter, UK and Utrecht University, The Netherlands

This presentation will consider the impact of climate change on the burden of crop disease. However, of the various microbial challenges to food security, the threat of fungal (and oomycete) infection of our calorie and commodity crops outstrips that posed by bacterial and viral diseases combined (Fisher *et al.*, 2012 *Nature*; Fones *et al.*, 2020 *Nature Food*; Case *et al.*, 2025 *Nature*).

We face a future blighted by known adversaries, by new variants of old foes and by new diseases. Modern agricultural intensification practices have heightened the challenge - the planting of vast swathes of genetically uniform crops, guarded by one or two inbred resistance genes, and use of single target site antifungals has hastened emergence of new virulent and fungicide-resistant strains (Fisher *et al.*, 2018 *Science*; Fisher *et al.*, 2022 *Nature Reviews Microbiology*). Climate change compounds the saga as we see altered disease demographics - pathogens are moving poleward in a warming world (Bebber *et al.*, 2013 *Nature Climate Change*; Chaloner *et al.*, 2021 *Nature Communications*; Chaloner *et al.*, 2022 *Nature Climate Change*).

This presentation will highlight some current notable and persistent fungal diseases. It will consider the evolutionary drivers which underpin emergence of new diseases and man-made “accelerators” of spread. I will set these points in the context of a series of different disease models, initially with statistical correlative models, and thence with more recent mechanistic models - parametrised by data collected from pathogen, host, climate and with a temporal axis. Such models have enabled us to look across biological scales, that is from the global level to crop to host-pathogens *per se*, in our development of more accurate predictive movement models. I shall conclude with some thoughts about disease control in the future (Steinberg *et al.*, 2021 *Nature Communications*; Cannon *et al.*, 2022 *Plos Pathogens*).

## **PS2.1 - From lesions to lessons: The past, present and future of filamentous plant pathogen genomics**

**Thorsten Langner**<sup>1</sup>

Cristina Barragan<sup>2,3</sup>, Sergio Latorre<sup>4</sup>, Lucía Gómez Gil<sup>1</sup>, Paul Grablowitz<sup>1</sup>, Joe Win<sup>2</sup>, Adeline Harant<sup>2</sup>, Yu Sugihara<sup>2</sup>, Hernan Burbano<sup>4</sup>, Sophien Kamoun<sup>2</sup>

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Over the past two decades genomics has emerged as a central technology to study filamentous plant pathogens. Since the first genome of the blast fungus, *Magnaporthe oryzae*, was published in 2005, genomics has not only advanced our understanding of pathogen population dynamics and improved disease diagnostics but has also accelerated molecular research and provided insights into gene regulatory mechanisms. With the increasing availability of high-quality assemblies, we have now entered the pangenomics era, enabling detailed insights into genome dynamics and structural organization. The genome of filamentous plant pathogens is often compartmentalized into relatively stable core and highly variable accessory regions. An extreme form of this compartmentalization are supernumerary chromosomes which are only present in some individuals of a species. These chromosomes can be horizontally transferred and facilitate structural rearrangements, thereby contributing to gene flow and extensive presence-absence and copy number variations in pathogen populations. Here, I will discuss recent advances in our understanding of plant pathogen genome dynamics and provide an outlook into possible future applications of plant pathogenomics.

## **PS2.2 - What genomes tell us about the emergence of crop pathogens**

**Daniel Croll**<sup>1</sup>

<sup>1</sup> Laboratory of Evolutionary Genetics, Institute of Biology, University of Neuchatel, Switzerland

Adaptation of plant pathogens proceeds at speeds that easily overwhelm the rate of resistant cultivar deployment and fungicide development. Low cultivar diversity and the application of single fungicides can exacerbate these dynamics. Understanding the molecular basis of pathogen adaptation is critical to define more sustainable containment strategies. Emerging traits such as virulence on resistant cultivars or fungicide resistance often appear in a geographically structured manner. Such geographic mosaics can be determined by regional variation in selection pressures or as a consequence of the pathogen's population history. I will introduce the use of large panels of pathogen genomes from the same species. Such genome panels can recapitulate historic colonization patterns and reveal potential constraints on pathogen genetic diversity. In combination with phenotyping assays, large genome panels enable the tracking of fungicide resistance mutations across continents and reveal how effector gene loci are undergoing adaptive sequence rearrangements. Recent research has pointed to selfish elements (i.e. transposable elements) as key factors in pathogen evolution, facilitating adaptation to biotic and abiotic factors in their environment. Selfish elements, by nature, can also impact the integrity of genomes and lead to deleterious dynamics for the pathogen. In conclusion, large-scale genomic investigations of individual pathogen species unravel essential mechanisms of pathogen adaptation.

### PS2.3 - Transposons drive environmental adaptation in a clonally evolving fungal pathogen

Cristina López Díaz<sup>1</sup>

Dilay Hazal Ayhan<sup>2</sup>, Ana Rodríguez López<sup>1</sup>, Lucía Gómez Gil<sup>1</sup>, Li-Jun Ma<sup>2</sup>, **Antonio Di Pietro**<sup>1</sup>

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Genomes of clonally reproducing fungal pathogens are often compartmentalized into conserved core and lineage-specific accessory regions (ARs) enriched in transposable elements (TEs). ARs and TEs are thought to promote pathogen adaptation, but direct experimental evidence is sparse. Using an evolve and re-sequence approach, we found that serial passaging of the cross-kingdom fungal pathogen *Fusarium oxysporum* through tomato plants or axenic media rapidly increased fitness under the selection condition. TE insertions were the predominant type of mutations in the evolved lines, with a single non-autonomous hAT-type TE accounting for 63% of total events. TEs inserted preferentially at sites of histone H3 lysine 27 trimethylation, a hallmark of ARs. Recurrent evolutionary trajectories during plate adaptation led to increased proliferation concomitant with reduced virulence. Unexpectedly, adaptive mutations in accessory genes strongly impacted core functions such as growth, development, quorum sensing and virulence. Thus, TEs and ARs function as adaptive drivers in this important fungal pathogen.

## PS3.1 - A route to leaf galls

**Armin Djamei**<sup>1</sup>

<sup>1</sup> University of Bonn

Smut fungi, a diverse group within the basidiomycetes, are biotrophic specialists, meaning they thrive by infecting living plants—particularly grasses. Among their hosts are some of the world's most important crops, including sorghum, millet, barley, and maize. A standout example is the maize smut fungus *Ustilago maydis*, which has become a model organism for studying smut fungi. This pathogen is notorious for inducing large galls on the maize plant's above-ground tissues, a clear sign that it has reprogrammed the plant's metabolism and growth processes. These galls are acting as an induced organ and sink-tissue of the plant where the spore-formation of the fungus occurs inside. The plant-sided signaling and developmental pathways underlying gall formation are not well understood.

The key to the remarkable manipulation of the host plant lies in small, secreted fungal molecules known as effectors. Over the past decade, our research has systematically screened hundreds of these effector proteins to uncover their specific roles within the plant. This effort has greatly expanded our understanding of how *U. maydis* manipulates its host at the molecular level.

In this presentation, I will discuss the latest insights into the fungal "effectome"—the complete set of effectors produced by the fungus—and the biotrophic interaction between *U. maydis* and maize. A special focus will be on a fascinating group of effectors that promote cell division and pluripotency in plant cells by hijacking conserved plant signaling pathways. Our findings reveal surprising insights how components of an unexpected plant developmental pathway are reused to serve a microbial pathogen.

## PS3.2 - Leveraging heterogeneous surveillance data for modeling and inferring disease spread

**Samuel Soubeyrand**<sup>1</sup>

<sup>1</sup> INRAE, BioSP, 84914 Avignon, France

Surveillance data about diseases are primarily collected to inform the sanitary status of territories, draw alerts, and initiate control actions accordingly. They can also be used to model and infer the spatiotemporal dynamics of the responsible organisms. Numerous approaches have been pursued toward this goal, drawing on various types of surveillance data (field observations, remote sensing...). Throughout case studies, I will present how some of these approaches have been utilized to produce knowledge about the large-scale dynamics of plant pathogens (e.g., *Xylella fastidiosa*, yellowing viruses...). I will also highlight several promising research directions in this field.

Ref.: Soubeyrand S. et al. (2024). Building integrated plant health surveillance: a proactive research agenda for anticipating and mitigating disease and pest emergence. *CABI Agriculture and Bioscience* 5:72. <https://doi.org/10.1186/s43170-024-00273-8>

## **PS4.1 - Tree resistance breeding for forest health in a changing world**

**Malin Elfstrand<sup>1</sup>**

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Trees and forest ecosystems have a role in the global carbon cycle accounting for three quarters of terrestrial gross primary production. Despite their importance, forest ecosystems and many tree species are under threat and the growth is stagnating in many forest ecosystems. In northern temperate forests, the main threats to tree species are invasive and emerging pests and diseases. The numbers of recognized invasive and emerging pests, diseases are growing rapidly, and they often interact with other abiotic and biotic factors creating novel disease and outbreak dynamics. This creates unprecedented challenges for forest health and sustainability.

Assisted migration and tree breeding are strategies that can improve the planting stocks' tolerance to climate change, pests and diseases. The identification and use of tree genotypes with lower disease susceptibility has an important role in management practices irrespective of if the objective is increased productivity or the restoration of threatened tree species into natural populations. However, tree resistance breeding is presented with a number of challenges associated with trees relatively long generation times and long lived nature, and with a lack of understanding of their basic resistance biology and genetics. Key opportunities and challenges in forest tree resistance breeding and deployment will be discussed.

## **PS4.2 - Tomato brown rugose fruit virus in Canada: Progress in research and future prospects for disease control**

**Aiming Wang<sup>1</sup>**

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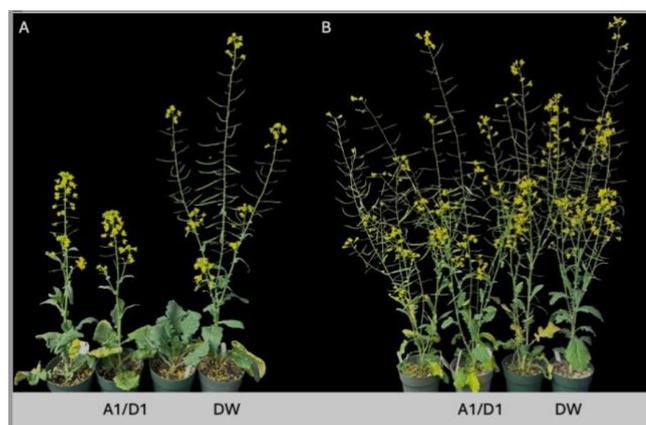
Tomato brown rugose fruit virus (ToBRFV) is an emerging and rapidly spreading plant RNA virus that currently threatens tomato and pepper production worldwide. ToBRFV belongs to the genus *Tobamovirus* in the family *Virgaviridae* and was first discovered in the Middle East in 2015. Since then, the virus has spread to at least 40 countries across four continents including Asia, Europe, North America, and Africa. ToBRFV are highly contagious with tomato as the primary host. The virus is mainly transmitted via contaminated seeds and mechanical contact. In tomato, its infection usually causes 30% to 70% yield losses. ToBRFV can break down genetic resistance to tobamoviruses conferred by all known *R* genes including the durable resistance gene *Tm-2<sup>2</sup>*. In 2019, ToBRFV was first found in tomato plants from greenhouses in Canada. The industry quickly responded with implementation of enhanced biosecurity, disinfection and hygiene measures and application of tolerant cultivars. In collaboration with provincial governments and grower associations, researchers from Agriculture and Agri-Food Canada (AAFC), a Canadian federal government department, launched a ToBRFV research program to better understand ToBRFV and its interactions with tomato, generate novel freedom-to-operate genetic resistance, develop attenuated ToBRFV strains for cross-protection against ToBRFV, and establish best management practices (BMPs). Here, I would like to share research progress in these research areas and discuss future prospects for the control of ToBRFV in Canada.

### PS4.3 - Success story in Canadian blackleg disease management in canola: Does *Verticillium longisporum*, threaten this success

Dilantha Fernando<sup>1</sup>

<sup>1</sup> Department of Plant Science, St. Paul's College, University of Manitoba, Winnipeg, Manitoba, Canada R3T 2N2

Blackleg in canola was the most devastating disease in canola in Canada for nearly 4 decades. Successful national and international collaborations, sound funding, and highly qualified personnel introduced several strategies that led to the success of mitigating the disease. Naming R-genes, introducing labeling cultivars with R-genes, introducing a R-gene rotation in fields, using KASP markers to identify the pathogen avirulence (*Avr*) and virulence (*avr*) alleles, identifying *AvrLm3* and *AvrLm9* masked by the presence of *AvrLm4-7* (epistatic effect), and sequencing 250 global isolates of the blackleg pathogen made the success. Multiple genes significantly reduce disease when Rlm4 protects Rlm3 breakdown. However, introducing new R-gene cultivars and a new pathogen (*Verticillium longisporum*) causing Verticillium stripe disease is now threatening this success. *V. longisporum* is spreading and establishing rapidly in the prairie landscape. We observed an interaction and presence of both (*Leptosphaeria maculans*, *L. biglobosa*, and *Verticillium longisporum*) in the same plant. The threat is real and rapid. The evolution of avirulence to virulence with the introduction of new R-genes (analysis revealed numerous single-nucleotide polymorphisms (SNPs) and structural mutations), the identification of novel R-genes (in elite genotypes - qualitative), minor genes (quantitative), and the identification of S-genes in a TILLING (Targeting Induced Local Lesions in Genomes) population with mutations (achieved through functional disruption of susceptible (S) genes) from a canola cultivar, and its mutant lines showing single resistance to a pathogen or dual resistance to both pathogens, and developing LAMP, and qPCR probes to detect both pathogens simultaneously is helping us look at the future of the canola crop in Canada with promise. These latest findings, mainly through genetics, and how we arrived at the initial success in mitigating the blackleg disease through understanding the host-pathogen interaction, spatial scale studies in the field, and working with growers for this success, will be presented.



**Figure 1:** The phenotypic changes of wild type (A) and 124-5 mutagenized line (B) after *Verticillium longisporum* A1/D1 inoculation and distilled water (DW) treatment. First three plants of each line were infected with *V. longisporum* A1/D1 and fourth plant was treated with distilled water. The images were taken after 40 days of post-inoculation.

## **PS5.1 - AI tools in plant pathology: Risks and opportunities**

**Eduardo Sáenz-de-Cabezón<sup>1</sup>**

<sup>1</sup> Universidad de La Rioja, Spain

Artificial intelligence (AI) is rapidly emerging as a set of transformative tools across various scientific disciplines, and plant pathology stands to gain significantly from its integration. This keynote address will explore AI tools in this field, highlighting the opportunities and the potential risks that accompany their adoption.

AI offers unprecedented opportunities to revolutionize plant disease management. Machine learning algorithms can analyze vast datasets from diverse sources, including images, spectral data, and environmental parameters, enabling early and accurate disease detection, often before symptoms are visible to the naked eye. AI-powered diagnostic tools can aid in rapid pathogen identification, facilitating timely intervention and reducing crop losses. Predictive models can forecast disease outbreaks, allowing for proactive application of control measures and minimizing environmental impact. Furthermore, AI can optimize breeding programs by accelerating the screening of disease-resistant varieties.

However, the integration of AI in plant pathology also presents potential risks. The reliance on large datasets raises concerns about data quality, accessibility, and standardization. Algorithmic bias can lead to inaccurate predictions or unfair outcomes, particularly for underrepresented crops or regions. The "black box" nature of some AI models can hinder interpretability and trust, making it challenging to understand the biological mechanisms underlying disease development. Ethical considerations surrounding data privacy, ownership, and the potential displacement of human expertise must be carefully addressed.

## PS5.2 - Single-cell and spatial dissection of plant-pathogen interactions

Tatsuya Nobori<sup>1</sup>

<sup>1</sup> The Sainsbury Laboratory

Plants lack specialised and mobile immune cells, requiring any cell encountering pathogens to mount cell-autonomous immune responses and communicate with surrounding cells to deploy non-cell-autonomous responses. Despite its conceptual importance, the cellular basis of the plant immune response remains largely unknown. To address this, we conducted a time-course experiment integrating single-nucleus multiomics (snRNA-seq and snATAC-seq) with spatial transcriptomics in *Arabidopsis thaliana* leaves infected by bacterial pathogens (*Pseudomonas syringae*), capturing the spatiotemporal dynamics of immune responses. This approach revealed previously uncharacterized immune cell states and their potential gene regulatory networks. Importantly, we discovered rare Primary IMMunE Responder (PRIMER) cells that initially activate immunity upon pathogen invasion, along with surrounding bystander cells in the mesophyll. Our molecularly defined single-cell spatiotemporal atlas provides functional and regulatory insights into immune cell states in plants. Additionally, I will discuss our efforts to develop innovative methodologies for high-resolution analysis of plant-microbe interactions, including PHYTOmap, a novel method for multiplexed single-cell 3D spatial gene expression analysis in whole-mount plant tissues. Lastly, I will outline our roadmap toward a comprehensive understanding of the diversity, functionality, regulation, and communication of plant immune cell states, which serve as critical building blocks of plant-pathogen interactions.

## **PS6.1 - From prevention to control: The BeXyl project's response to the *Xylella fastidiosa* threat in Europe**

**BLANCA B. LANDA**<sup>1</sup>

The BeXyl Consortium<sup>1</sup>

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The BeXyl project is a Horizon Europe-funded, multi-actor initiative that addresses the serious threat posed by *Xylella fastidiosa*, a quarantine plant pathogen and priority pest in the European Union. By fostering interdisciplinary research and innovation, BeXyl aims to deliver effective strategies for managing *X. fastidiosa*-related diseases, particularly in key tree crops such as olive. The project focuses on delivering impact across three main pillars: strengthening capacities for prevention, surveillance, and rapid response; (ii) advancing biologically based control strategies; and (iii) providing science-based support for plant health policies across EU and associated countries.

Aligned with the EU Plant Health Law and the Green Deal goals for sustainable agriculture and forestry, BeXyl reinforces the foundational pillars of plant protection: prevention and control. This talk will highlight efforts to enhance early detection and border inspection through: (i) development and validation of novel molecular diagnostic tools and evaluation of EPPO-recommended methods; (ii) application of remote sensing technologies for large-scale field surveillance; (iii) development of on-site phenotyping approaches to assess plant infection status. Additionally, we will present how the exploration and application of synthetic microbial communities could offer an innovative biological strategy for the sustainable management of *X. fastidiosa*.

BeXyl will support research, innovation, and knowledge transfer to strengthen crisis preparedness, to prevent new *X. fastidiosa* outbreaks and mitigate further spread of currently infested areas, ultimately contributing to the resilience and productivity of European agriculture and forestry.

**Funding:** This research was funded by BeXyl Project (Beyond *Xylella*, Integrated Management Strategies for Mitigating *Xylella fastidiosa* impact in Europe (Grant ID 101060593)

## **PS6.2 - Context, climate, and control of plant colonization by beneficial bacteria**

**Derek Lundberg<sup>1</sup>**

<sup>1</sup> Swedish University of Agricultural Sciences, Uppsala

Climate change exposes crops to extreme weather and changes patterns of plant disease. Meanwhile, global food demand continues to increase. The impact of these immense problems can be reduced by using beneficial microbes to improve plant growth, but how and why some beneficial microbes thrive in complex agricultural field settings, while others do not, remains difficult to explain. Therefore, turning lab findings into reliable agricultural products often fails.

# **Molecular plant-microbe interactions**

## **O2.1 - XopG2 effector of *Xanthomonas campestris* pv. *campestris* is a major Race 5 determinant**

Molecular plant-microbe interactions

**Najeeb Ullah**<sup>1</sup>

Ana Dominguez Ferreras<sup>1</sup>, Shannon Greer<sup>1</sup>, David J. Studholme<sup>2</sup>, Murray Grant<sup>1</sup>, Vardis Ntoukakis<sup>1</sup>

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Black rot caused by *Xanthomonas campestris* pv. *campestris* (*Xcc*) poses a significant risk to brassicas, such as cabbage, cauliflower, and Brussels sprouts. *Xcc* has 11 known races defined by distinct sets of effector proteins. Yet, our understanding of the role of race-specific effectors in *Xcc* pathogenicity is still limited. Through comparative genomics, we identified the XopG2 effector (a predicted metalloprotease) as the determinant of *Xcc* Race 5. Deletion of *XopG2* in *Xcc* R5 (*Xcc* R5  $\Delta$ XopG2) and mutations in the metalloprotease active site of XopG2 avoid recognition by the resistant *Brassica oleracea* line SxD1 while complementation of *Xcc* R5  $\Delta$ XopG2 with XopG2 restores the wild type R5 phenotype. Furthermore, expression of *XopG2* in the highly virulent *Xcc* Race 6 compromised its pathogenicity. Consistent with these observations, we identified naturally occurring allelic variants of *XopG2* with a truncated metalloprotease active site that evolved to evade recognition. Using proteomics and split luciferase assays, we identified EDR2 (enhanced disease resistance 2) as the interacting partner of XopG2. Currently, we are working to identify resistance proteins guarding EDR2.

## O2.2 - The role of ethylene in the *Verticillium dahliae*-pepper interaction: defense mechanism or disease driver?

Molecular plant-microbe interactions

**Roos Bex**<sup>1,2</sup>

Shirley Marcou<sup>1</sup>, Yukiyo Sato<sup>3</sup>, Florà Magnus<sup>1</sup>, Lien Tyvaert<sup>1</sup>, Thomas Depaepe<sup>4</sup>, Siel Goethals<sup>1</sup>, Dominique Van Der Straeten<sup>4</sup>, Bart Thomma<sup>3</sup>, Kathy Steppe<sup>2</sup>, Monica Höfte<sup>1</sup>

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Ethylene is a key plant hormone involved in various physiological processes, including defense responses during pathogen infections. Some pathogens, like *Verticillium* species, manipulate ethylene biosynthesis to enhance disease severity. The most notorious species is *V. dahliae*, a vascular pathogen that causes Verticillium wilt in more than 200 host species, leading to extensive crop yield losses. During infection, ethylene is produced by both the plant and the pathogen. Microorganisms can use several pathways for ethylene biosynthesis, including the 1-aminocyclopropane-1-carboxylic acid (ACC) pathway, which is also found in plants. Another pathway converts 2-oxoglutarate and L-arginine into ethylene via the ethylene-forming enzyme (EFE). To validate that *V. dahliae* uses both ethylene biosynthesis pathways, *in vitro* experiments with nutrient-limiting conditions, simulating the xylem, were conducted to assess ethylene production and gene expression. Subsequently, plant infection experiments were set up using the *V. dahliae* VD1-pepper (*Capsicum annuum* L.) pathosystem. Gene expression of several key enzymes in ethylene biosynthesis pathways of both the plant and pathogen was analyzed at several time points during infection. Additionally, an *EFE* deletion mutant was generated to investigate the role of this pathway in disease progression. Changes in symptom severity, pathogen colonization, ethylene biosynthesis gene expression and total ethylene production were analyzed. Our findings reveal that plant ethylene biosynthesis is significantly upregulated during infection and that ethylene production by the EFE pathway of the pathogen also contributes critically to disease development.

## O2.3 - Extracellular: Exploring plant-pathogen interactions in the apoplast

Molecular plant-microbe interactions

**Brian Mooney**<sup>1</sup>

Jie Huang<sup>1</sup>, Felix Homma<sup>1</sup>, Farnusch Kaschani<sup>2</sup>, Joy Lyu<sup>1</sup>, Markus Kaiser<sup>2</sup>, Renier van der Hoorn<sup>1</sup>

<sup>1</sup> University of Oxford

<sup>2</sup> Universität Duisburg-Essen

Plant diseases cause massive crop losses annually, threatening global food security and devastating local economies. The colonisation of plant tissues by many pathogens begins in the apoplast (extracellular space). Upon pathogen detection, host plants secrete an array of defence-related enzymes to resist infection. In response, adapted pathogens release ‘effector’ proteins that may neutralise these defences, *e.g.* by forming inhibitory protein complexes with apoplastic hydrolase enzymes. Such interactions are crucial in determining the progression of plant disease, but only a handful have been described in detail. We aim to characterise apoplastic plant-pathogen interactions in the *N. benthamiana* – *P. syringae* pathosystem using two complementary approaches: 1) AI-guided prediction of protein-protein interactions and 2) chemical proteomics.

We screened > 10,000 potential protein-protein interactions using AlphaFold multimer to identify candidate pathogen-derived suppressors of diverse host apoplastic hydrolases including the cysteine protease ‘Cathepsin-B’ and the glycosidase ‘Beta-galactosidase 2’ (BGAL2). Putative interactions were later validated by activity-based protein profiling (ABPP) and co-immunoprecipitation assays (Co-IP). We also deploy reactivity-based protein profiling (RBPP) to investigate plant-pathogen interactions for the first time, revealing novel host targets of pathogen manipulation. These findings shed new light on the mechanisms used by plant pathogens to overcome apoplastic defences and highlight targets for protein engineering to avoid pathogen-mediated suppression and bolster plant resilience.

## O2.4 - Early gene expression and effectors of *Plasmodiophora brassicae* in broccolini, broccoli, and gai lan

Molecular plant-microbe interactions

**Claudia Vacca**<sup>1</sup>

Donovin Coles<sup>1</sup>, Jeff Powell<sup>1</sup>, Jonathan Plett<sup>1</sup>

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*Plasmodiophora brassicae*, the causal agent of clubroot, is a threat to all brassica crops, including the hybrid broccolini and its parental lines, broccoli and gai lan. In this study we investigated early-stage host-pathogen interactions using transcriptome assemblies for *P. brassicae* during infection in all three hosts. Using differential expression analysis we identified key effector genes, which suggested potential mechanisms of host immune modulation.

To define infection timepoints, we performed comparative microscopy on infected root tissues from each host, which allowed visualization of pathogen development. This analysis enabled us to identify three key stages for transcriptome profiling: root hair/cortex aggregation of spores, the beginning of secondary plasmodium formation, and mature secondary plasmodium development. We observed these stages happening at different times post-inoculation in the three cultivars. We then conducted RNA sequencing and differential gene expression analysis to characterize *P. brassicae* effector dynamics across these infection stages.

Following comparative transcriptomics analysis, we identified the differences in host responses, with broccolini exhibiting a distinct expression profile from its parental cultivars. Several defense-related genes were differentially expressed, indicating variations in resistance strategies among broccolini, broccoli, and gai lan. Additionally, stage-specific expression patterns of *P. brassicae* effectors highlighted potential roles in pathogen establishment and proliferation.

These findings provide new insights into the molecular mechanisms governing early *P. brassicae* infection and highlight how broccolini's response is shaped by its parental species, offering a basis for targeted breeding strategies. They also pose a foundation for functional validation of candidate effectors to improve disease resistance in brassica crops.

## O2.5 - Vertical virus transmission in RNA interference deficient plants

Molecular plant-microbe interactions

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RNA interference (RNAi) is a well-established antiviral defense mechanism in plants, insects, and other organisms, targeting viral genomes, either RNA or DNA, to impede their replication. However, its efficacy can be compromised by viral suppressors of RNAi (VSRs), which many plant viruses encode to counteract this defense. Recent studies on plant viruses such as TuMV, TyMV, TCV, and CMV have shown that their vertical transmission is completely suppressed under dysfunctional RNAi conditions, leading to sterile plants. Notably, this sterility was only reversed in CMV strains lacking VSRs, suggesting that some viruses mitigate their VSR virulence to persist in the host germ line.

To investigate whether vertical transmission in RNAi-deficient lines is a mechanism utilized by persistent viruses, we examined flower development and seed production in two additional viral species using *Arabidopsis thaliana* mutants with varying RNAi deficiencies. We assessed both plant and viral fitness across the RNAi mutant panel by quantifying seed production weight, germination rates, and viral incidence in progeny using ELISA, qRT-PCR, and protein gel analysis. Additionally, infections in RNAi suppressor overexpression lines (P1, P6/TAV, and 2b) were tested to evaluate the relationship between suppressor strength and vertical transmission efficiency.

We found that both unaltered viruses from two previously untested families supported flower development in RNAi-deficient lines, but only one successfully achieved seed production and vertical transmission. Moreover, viral inheritance was not compromised in any of the VSR-overexpressing lines. These findings suggest that persistent viruses bypass vertical transmission restrictions through mechanisms beyond weakening their VSRs. Our results provide new insights into the role of RNAi in shaping viral transmission strategies and establish a novel experimental pathosystem for studying the evolutionary continuum of parasitism, commensalism, and mutualism.

## **O2.6 - Development of a virus-based host induced gene silencing approach to control *Verticillium dahliae* in herbaceous plants**

Molecular plant-microbe interactions

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*Verticillium dahliae* is a devastating vascular pathogen affecting many economically important plants, with no effective control methods currently available. The advancement of biotechnological approaches like RNA interference (RNAi) is a highly promising strategy to manage phytopathogens. The recent discovery of important genes involved in pathogenicity of *V. dahliae*, can be utilized for its efficient control. RNAi technology is an environmentally friendly alternative to synthetic pesticides, based on the modification of gene expression in plants by targeting pathogenicity genes. The aim of this study was to utilize a TRV-based host-induced gene silencing (HIGS) method, to suppress critical genes of the pathogen. For this purpose, highly conserved and essential genes involved in vital functions such as signal transduction (G protein beta subunit), regulation of the expression of cell wall-degrading enzymes (sucrose non-fermenting-*SNFI*), and induction of necrosis and ethylene production (NLP-like genes) were selected. Based on bioinformatic analysis, specific gene segments were chosen to avoid off-target effects, ensuring specificity to the pathogen without affecting the expression of homologous genes in the host plant or beneficial microbiome. Each gene segment was cloned into the TRV2 vector and transformed into *Agrobacterium tumefaciens*. Cotton and tomato plants were agroinfiltrated with cells harboring the recombinant TRV vector, to ensure dsRNA processing by the host and deliverance of the siRNA molecules into the fungal cells upon artificial inoculation with a spore suspension of *V. dahliae*. The effectiveness of this method, assessed by quantification of the pathogen's biomass, fusion protein visualization with the *gfp* biomarker, reduction of disease index and silencing of targeted genes, are underway and will be discussed.

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## O6.1 - Differential gene-expression profiling of white lupin in response to infection by *Colletotrichum lupini*

Molecular plant-microbe interactions

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White lupin (*Lupinus albus* L.) is a grain legume with high agronomic potential both used as green manure as well as for its highly nutritious seeds. Albeit, it remains a niche crop in Europe, which is mostly due to difficulties in production. One of the main obstacles is its susceptibility to the hemibiotrophic fungus, *Colletotrichum lupini* causing anthracnose disease. After a brief biotrophic phase (up to 4 days post-inoculation) the fungus enters into necrotrophy, followed by reproduction with asexual conidia forming reportedly even 6 days post-inoculation, ready for re-infection and further spreading, which leads to total yield loss in unfavourable years with susceptible varieties.

Studies have shown that anthracnose resistance in white lupin is polygenically controlled and increased quantitative resistance is scarce within the genepool. We hypothesize that in these few resistant accessions, distinct transcriptional changes occur at crucial timepoints throughout the swift infection process. A better understanding of the molecular machinery behind the transcriptomic response would contribute considerably to the further improvement of the crop.

Through differential gene-expression profiling by total RNA sequencing of white lupin in response to inoculation with *Colletotrichum lupini*, we address both genotype- and time-dependent transcriptomic changes. We are comparing four different white lupin accessions representing different sources of resistance in contrast to two susceptible cultivars and non-inoculated controls at 11 timepoints spanning the entire fungal life-cycle (from 1 hour to 9 days post-inoculation) under controlled conditions.

We will present the results of a combined analytical approach. Using pairwise comparisons we determined differentially expressed genes at each timepoint for each white lupin accession. Simultaneously, we applied linear mixed models for the identification of key moments of white lupin defence against *Colletotrichum lupini*.

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## O6.2 - Host-dependent fitness of *Scaphoideus titanus*: Understanding cultivar susceptibility to flavescence dorée

Molecular plant-microbe interactions

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Identifying resistance traits is essential in combating incurable plant diseases like grapevine Flavescence dorée (FD). This quarantine disease, associated with a phytoplasma and mainly transmitted by the leafhopper *Scaphoideus titanus*, has no effective cure and leads to severe yield losses and economic setbacks. In Switzerland, the cultivar Chasselas is scarcely affected by FD, despite being the most cultivated variety in FD-affected regions. However, susceptibility trials revealed no significant difference in phytoplasma tolerance, suggesting that the reduced incidence of FD in Chasselas may be linked to its interaction with the disease vector *S. titanus* rather than direct pathogen tolerance. Here, we investigate the mechanisms of Chasselas that prevent phytoplasma transmission by comparing it with Pinot Noir, a variety severely affected by the disease in Switzerland. Under natural vineyard conditions, *S. titanus* populations were consistently larger in Pinot Noir than in adjacent Chasselas vineyards, and a higher number of egg eclosions was observed in vegetal material collected from Pinot Noir vineyards compared to Chasselas. However, a 9-hour choice test revealed no clear preference of *S. titanus* for either variety. In addition, insect fitness and reproductive performance assessed under controlled conditions showed that *S. titanus* exhibited a significantly longer lifespan and higher reproductive output on Pinot Noir compared to Chasselas. Upcoming experiments will assess whether female egg load varies based on host plant exposure, whether plant-derived volatile organic compounds (VOCs) influence insect attraction, and whether the cultivar affects the dietary behavior of the insect. The findings of this study highlight a significant effect of the cultivar Chasselas on *S. titanus* fitness and fertility, which will be further investigated to identify key molecular mechanisms responsible for its lower susceptibility to the insect vector. Uncovering these traits has important implications for sustainable vineyard management and will aid in the development of resistant grape varieties.

### **O6.3 - Can the growth penalty caused by resistance inducers be exploited for crop protection? Insights from cerato-platanin**

Molecular plant-microbe interactions

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The practical use of resistance inducers to protect crops from diseases is hampered by the “growth-defense trade-off”. The plant induced to defend itself sustains metabolic expenditure and antagonistic hormonal cross-talks that may negatively affect its growth and yield. With this work, we aimed to exploit this phenomenon to fight weeds. The sensitivity of arable weeds to defense elicitors is largely unknown, since reports existing on the topic mostly focus on *Arabidopsis* and crops. The work was carried out with a fungal elicitor named Cerato-platanin (CP), extensively studied by our research group and known to induce PAMP-triggered immunity (PTI) in *Arabidopsis* and *Platanus acerifolia*. CP is a protein (120 a.a.) belonging to the “Cerato-platanin family” that is produced by the plant pathogenic fungus *Ceratocystis platani*. The ability of CP to be perceived by arable weeds was assessed by seedling growth inhibition assays performed on Poaceae and Amaranthaceae known for the resistance to herbicides. CP-treated seeds from *Lolium multiflorum*, *Digitaria sanguinalis* and *Amaranthus hybridus* showed reduced shoot and/or root length during germination, revealing sensitivity to the protein. In contrast, *A. palmeri*, *A. tuberculatus* and *Avena* sp. seedlings were not affected by the presence of CP, suggesting insensitivity. Interestingly, CP did not affect growth of *Triticum aestivum* seedlings, highlighting the possibility of selectively targeting certain weed species while safeguarding the crop. A dose-response assay performed in *L. multiflorum* with CP and BSA as protein control showed specific and concentration-dependent activity by CP. Western-blot analysis showed absorption of CP by *L. multiflorum* seedlings during germination, while RT-qPCR analyses confirmed the activation of defenses. Preliminary results obtained with CP-derived peptides suggest that *L. multiflorum* perceives the N-terminal region of CP. This study highlights the possibility of using the knowledge acquired on plant immunity to provide novel sustainable solutions for crop protection.

## O6.4 - Distinct roles of two *Rcr1*-interacting proteins in *Rcr1*-mediated clubroot resistance in brassica crops

Molecular plant-microbe interactions

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Clubroot, caused by *Plasmodiophora brassicae*, is a major disease affecting *Brassica* crops and poses a significant threat to canola (*Brassica napus*) production in Western Canada. Host resistance is the most effective and sustainable management strategy. While numerous clubroot resistance (CR) genes have been identified through genetic mapping, the molecular mechanisms underlying CR gene-dependent resistance remain unclear, hindering the development of durable-resistant cultivars. Building on our previous mapping-by-sequencing work, we conducted map-based cloning in resistant Chinese cabbage (*B. rapa*) lines and identified several Toll-Interleukin-1 receptor/nucleotide-binding site/leucine-rich-repeat (TIR-NBS-LRR, TNL) resistance genes. Pathogenicity assays revealed that *Rcr1*, one of the cloned TNL genes, is widely present in resistant *B. rapa* lines and the first generation of Canadian clubroot-resistant cultivars. *Rcr1* expression in the susceptible canola line DH12075 via *Agrobacterium*-mediated transformation conferred strong resistance. However, resistance of DH12075<sup>*Rcr1*</sup> was lost when either the N- or C-terminal domain of the *Rcr1* protein was replaced with GFP using CRISPR/Cas9. To identify additional components involved in *Rcr1*-mediated resistance, yeast-two-hybrid screening coupled with next-generation sequencing identified 75 and 43 proteins interacting with the *Rcr1* TIR and LRR domains, respectively. Among 6 selected *Rcr1*-interacting proteins, CRISPR/Cas9 knockout of a cysteine protease or a thaumatin-like protein significantly impaired resistance in DH12075<sup>*Rcr1*</sup>. Notably, cysteine protease disruption in DH12075 alone reduced disease severity, whereas thaumatin-like protein knockout had no significant impact. These findings reveal distinct yet crucial roles of the two *Rcr1*-interacting proteins in *Rcr1*-mediated immunity. This study provides the first functional confirmation and characterization of a single dominant CR gene across multiple *Brassica* crops, offering valuable insights to improve breeding for durable clubroot resistance.

## O6.5 - Uncovering yellow rust resistance in European spring wheat

Molecular plant-microbe interactions

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Spring wheat (*Triticum aestivum* L.) is a vital crop in northern Europe, especially in areas where winter wheat cultivation is challenging. Climate change is exacerbating pest and disease pressures, including the emergence of new races of yellow rust (*Puccinia striiformis* f.sp. *tritici*), which significantly reduce yields. The CResWheat project, a collaborative pre-breeding initiative, aims to enhance the climate resilience of Nordic spring wheat by improving yield stability and disease resistance.

In 2022 and 2023, yellow rust resistance was assessed in multi-environment trials at four sites in Sweden, Denmark, and Norway. Most of the material used was of European origin, with a few cultivars sourced from USDA and CIMMYT. Genome-wide association studies (GWAS) identified quantitative trait loci (QTL) for resistance on chromosomes 2D, 3A, 4A, 5A, 5B, 6A and 7A, with the 6A QTL previously reported in Nordic germplasm.

Resistant genotypes have been selected to develop segregating populations, which will be evaluated in multi-environment trials across the Nordic region and Estonia. The project's key outcomes include DNA markers for breeding, new populations with stacked beneficial traits, and enhanced collaboration through knowledge and material exchange—fostering the development of climate-resilient spring wheat in northern Europe.

## **P1 - Switching on resistance to *Ralstonia solanacearum*: exploring gene expression, metabolite production and soil microbes**

Molecular plant-microbe interactions

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*Ralstonia solanacearum* is a soil borne bacterium with a devastatingly large host range, encompassing over 200 plant species, including key crops such as tomatoes and potatoes. *R. solanacearum*, with its already far-reaching distribution, is continuing to spread via new strains, such as II-B1, also known as race 3 biovar 2, which have adapted to cooler climates. This pathogen has been reported to cause over \$850 million in yield annually and with this increasing pressure from new strains, work to identify methods of resistance is vital to reduce pressure in countries with existing outbreaks and protect those that *R. solanacearum* threatens to invade.

This project aims to improve our understanding of the partial resistance exhibited by *Solanum dulcamara*, otherwise known as bittersweet nightshade, and test if this can be transferred to the closely related and susceptible crop, *Solanum lycopersicum*. We will focus on identifying differences in their response to infection of *R. solanacearum* at three levels: gene expression; metabolite production; and influence on the surrounding soil microbiome. Currently, to understand their differences at gene level, a gene co-expression network has been created to highlight differences in expression over time post inoculation. Additionally, the presence of microbes in the soil have been assessed in different conditions, including inoculation with *R. solanacearum*, beneficial metabolites or both. These two experiments are beginning to shed light on two out of three of our aims, improving our understanding of how resistance that already exists can be harnessed to protect key crops.

## **P2 - A novel effector from *Magnaporthe oryzae* strain targets the host nucleus and modulates plant defense responses**

Molecular plant-microbe interactions

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Rice blast, a disease caused by the fungal pathogen *Magnaporthe oryzae*, is a significant threat to rice production, often resulting in substantial yield losses. In East Africa, this fungus affects various cereal crops, including domestic sorghums and millets. Like other fungal pathogens, *M. oryzae* secretes effector proteins to manipulate host immune responses and cellular processes, enabling successful colonization and infection. In this study, we investigated a novel effector (named Mo2829Fm) from a *M. oryzae* high virulent strain isolated from diseased finger millet in Uganda. Gene expression analysis showed a significant induction of *Mo2829Fm* in the early stages of infection, followed by a decrease in the later phase. Gene deletion in *M. oryzae* and transient expression in *Nicotiana benthamiana* plants demonstrated its involvement in virulence, possibly through the suppression of genes involved in plant immunity, including Pathogen-Related proteins (PR), WRKY transcription factors, and mitogen-activated protein kinases (MAPKs). In silico analysis of the Mo2829Fm predicted it to be a cytoplasmic effector unique to the *M. oryzae* species. Confocal microscopy revealed a clear nuclear host localization of this effector. The structure of this effector contains two peptides linked by a serine protein linker. Our results showed that one peptide displayed a non-specific subcellular localization, while the latter is localized only in the nuclei. However, both peptides showed to be involved in virulence. Collectively, these findings underscore the distinctive nature of Mo2829Fm as a unique effector in *M. oryzae*, capable of subverting basal immunity by targeting the host nucleus.

### **P3 - Combining resistance to *Berkeleyomyces* spp. and TMV in tobacco hybrids and characteristics of the breeding lines**

Molecular plant-microbe interactions

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Two diseases that cause significant economic losses in Poland are tobacco mosaic, caused by the tobacco mosaic virus (TMV), and black root rot attributed to *Berkeleyomyces* spp. (former *Thielaviopsis basicola*). The best way to prevent losses caused by disease is to introduce genetic resistance into cultivars. However, the incorporation of pathogen resistance genes into the genome is often associated with a reduction in plant morphological traits and yield. The aim of the study was to obtain tobacco breeding lines combining resistance to TMV and black root rot to determine how resistance factors affect the tobacco agronomic performance.

The initial plant material included: 1) the American Burley-type cultivar TN90, which is resistant to TMV and *Berkeleyomyces* spp. and 2) two Polish Virginia tobacco cultivars: Wiślica which is susceptible to *Berkeleyomyces* spp. and Wigola, which is resistant to *Berkeleyomyces* spp. Intervarietal crosses were made, TN90 × Wiślica and TN90 × Wigola, followed by multiple backcrosses to Wiślica or Wigola, respectively. Selection of resistant plants in successive breeding generations (F<sub>1</sub>, BC<sub>1</sub>, BC<sub>2</sub>, BC<sub>3</sub>, BC<sub>4</sub>, BC<sub>5</sub>) was conducted using molecular markers linked to the N gene determining TMV resistance and *Berkeleyomyces* resistance bioassays. Resistant individuals from the BC<sub>5</sub> generation were crossed with the cytoplasmic male-sterile (CMS) form of paternal cultivar. The biometric measurements of the obtained breeding lines (WP7, WP9, WP10) showed that the combining resistance to TMV and *Berkeleyomyces* spp. did not negatively impact the morphological traits of tobacco. The plant height, number of leaves and leaf area were found to be similar to those of the parental cultivar of Virginia type. However, the percentage of cured leaves classified into the best grades was reduced in the yield. It indicated a decline in the quality of the raw tobacco, as well as a decrease in its nicotine content.

#### **P4 - Effects of $\alpha$ -solanine and $\alpha$ -chaconine on bacterial efflux pump activity and biofilm formation in pectinolytic bacteria**

Molecular plant-microbe interactions

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Pectinolytic bacteria, pathogenic to potato (*Solanum tuberosum* L.), cause significant economic losses in potato production worldwide. These bacteria employ multiple virulence mechanisms, including plant cell wall-degrading enzymes (PCWDEs), quorum sensing (QS), efflux pumps, and biofilm formation, all of which facilitate colonization and persistence in host tissues. As part of its defense strategy, potato synthesizes glycoalkaloids (GAs), steroidal secondary metabolites with antimicrobial properties. The predominant GAs in potato tubers,  $\alpha$ -solanine and  $\alpha$ -chaconine, have been studied for their inhibitory effects on bacterial growth (Sołtys-Kalina *et al.*, 2023). However, their influence on bacterial stress adaptation mechanisms, such as efflux pump activity and biofilm formation, remains poorly understood. Our recent evidence suggests that the ratio of  $\alpha$ -solanine to  $\alpha$ -chaconine in wild *Solanum* species and diploid hybrids may modulate bacterial virulence, highlighting the need for further investigation of their synergistic effects. This study explores the effects of synthetic  $\alpha$ -solanine and  $\alpha$ -chaconine, both individually and in a ratio reflecting their natural proportion in wild *Solanum* species, on the persistence and virulence traits of *Dickeya solani* and *Pectobacterium brasiliense*. We investigate whether these compounds influence bacterial efflux pump activity and biofilm formation, potentially interfering with QS-mediated regulation of virulence. Our findings will provide novel insights into the antimicrobial and regulatory effects of GAs, contributing to a better understanding of plant-derived defense mechanisms and their potential application in sustainable disease management strategies in potato cultivation.

Sołtys-Kalina D, Grupa-Urbańska A, Lebecka R, Tallant M, Kellenberger I, Dupuis B, 2023. Increase of Glycoalkaloid Content in Potato Tubers by Greening as a Method to Reduce the Spread of *Pectobacterium* and *Dickeya* spp. in Seed Production Systems. *Microorganisms* 11, 605.

## **P5 - Dual-RNaseq reveals the molecular mechanisms for unripe strawberry fruits withstand *Botrytis cinerea* infection**

Molecular plant-microbe interactions

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The necrotrophic fungal pathogen *Botrytis cinerea* is the etiological agent of grey mould (GM) disease. GM is a major threat to strawberry (*Fragaria x ananassa*) production, leading to severe fruit losses, at both pre- and post-harvest phases. In unripe fruits *B. cinerea* remains in a symptomless quiescent phase, while in ripe fruits it induces a rapid soft tissue decay. However, the mechanisms by which unripe fruits are more tolerant to GM remain poorly understood. This study aims to elucidate the molecular processes enabling unripe strawberries to resist infection. To investigate the strawberry-*B. cinerea* interaction, unripe and ripe strawberry fruits were infected with *B. cinerea* spores ( $10^5 \text{ mL}^{-1}$ ), and a control mock-infected group was included. Coding RNA (mRNA) and long non-coding RNA (lncRNA) were sequenced in both mock- and *B. cinerea*-infected fruits at 0, 3, 24 and 72 hours post inoculation (hpi). Based on differential gene expression (DGE) and gene ontology (GO) analyses, we found that unripe fruits had an early (3 hpi) upregulation of genes related to plant hormone signalling, particularly abscisic acid (ABA) biosynthesis, and plant defence, sterol synthesis, and cell wall organization pathways. On the other hand, in ripe fruits we observed an upregulation of some of these pathways, related to plant defences against pathogens, at late stages of infection (72 hpi). Moreover, we found that lncRNA response was similar between unripe- and ripe-infected fruits. Interestingly, *B. cinerea* infecting unripe fruits activates lipoic acid metabolism pathway (24 hpi) increasing oxidative stress tolerance, and down regulated linoleic acid pathway impairing the fungal growth and virulence. This study is pioneer in revealing the intricate molecular mechanisms involved in the unripe strawberry fruit tolerance to *B. cinerea*, which might to contribute to establish new sustainable strategies to control GM disease.

## **P6 - Fungal growth kinetics in stem tissues correlate with quantitative resistance to blackleg of canola**

Molecular plant-microbe interactions

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Quantitative resistance (QR) plays a crucial role in managing blackleg (*Leptosphaeria maculans*) in Canadian canola. Traditionally, field studies are used to assess QR in breeding materials, but these are time-consuming and often yield variable results due to environmental factors. This study presents a novel protocol using fungal growth kinetics in stem tissues to identify strong QR candidates among canola breeding lines and hybrids. We first evaluated 54 commercial lines with varying field resistance ratings by quantifying *L. maculans* DNA during fungal colonization of stem tissues using droplet-digital PCR (ddPCR) after cotyledon or petiole inoculation under greenhouse conditions. The *L. maculans* isolates carried a defined set of avirulence (*Avr*) genes, bypassing major R-gene resistance and enabling QR assessment. Both inoculation methods successfully established stem infections, and ddPCR measurements of fungal DNA amounts generally correlated with the earlier field resistance ratings. Strong correlations were also observed between cotyledon and petiole inoculation results and the resistance data from multi-year inoculated field trials (2019–2022). Subsequently, 40+ commercial canola hybrids from Canadian breeding programs were assessed using petiole inoculation and ddPCR. Over 90% exhibited strong QR, reducing fungal growth by >70% compared to a susceptible control. These findings highlight the effectiveness of this method for QR quantification and discovery while confirming that most commercial canola hybrids in Canada possess strong QR against blackleg, alongside specific R-gene resistance.

## **P7 - Genes and metabolic pathways activated in oilseed rape in response to *Plasmodiophora brassicae* (clubroot)**

Molecular plant-microbe interactions

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Clubroot is an economically important disease of oilseed rape in all countries where this crop is intensively cultivated. The disease is caused by the protist *Plasmodiophora brassicae*, a pathogen belonging to the infrakingdom Rhizaria. The lack of effective chemical control measures and no social consent for soil fumigation make genetic resistance the only effective method for protecting plants against clubroot. The aim of the study was to identify differentially expressed genes and find out the metabolic pathways related to the response of oilseed rape to infection with a pathogenic strain of *P. brassicae*.

Both healthy and heavily infected plants were found in cv. Crossfit at two fields located 100 km apart. To comprehensively characterize the gene expression profiles associated with immune response of plants, studies were conducted using RNA-seq. RNA transcriptome sequencing was performed following the published protocol, HISAT2 was used for alignments to the reference sequence.

Differential gene expression analysis was conducted on normalised count data using DESeq2. Functional annotation of candidate genes was used to elucidate differentially triggered metabolic pathways and processes (Kyoto Encyclopaedia of Genes and Genomes terms, Gene Ontology terms).

When comparing healthy roots of oilseed rape vs. infected roots, genes from six major metabolic pathways were identified as differentially expressed. The most important was the metabolism of branched-chain amino acids. Their degradation provides energy for the plant, especially under stress conditions such as pathogen infection. The other metabolic pathways activated in infected roots also play critical roles in energy metabolism, plant defense, immune responses and stress adaptation.

The results of our transcriptomic inquiry facilitates tracing differences in metabolic processes that distinguish healthy and infected plants. We have also studied differences occurring simultaneously in leaves of healthy and infected plants.

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## **P8 - Identifying candidate genes for bird-cherry oat aphid resistance in wild barley using targeted R gene sequencing**

Molecular plant-microbe interactions

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The bird-cherry oat aphid (BCA, *Rhopalosiphum padi*) poses a significant threat to barley production through direct feeding and as a vector for barley yellow dwarf virus and other viruses. Despite its impact, no commercial barley cultivars exhibit adequate resistance to BCA. Partial resistance (reduced nymph growth both under laboratory and field conditions) has been identified in the wild barley (*Hordeum spontaneum*) accession Hsp5 and introduced into modern barley germplasm via backcrossing. The genomic region conferring resistance was located to an 11 Mbp region on chromosome 2H.

Using the ResCap pipeline (Kushwaha et al., 2021), we enriched RNA for putative resistance (R) genes based on conserved motifs and quantified their expression in two susceptible cultivars, the resistant Hsp5, and four BC<sub>4</sub>F<sub>3</sub> lines, at six and 30 hours post-infestation. Seven differentially expressed R genes were identified, four within the QTL region on chromosome 2H and one 5 Mbp downstream. Three of these genes exhibited higher expression in Hsp5 and the BC lines, while two were upregulated in the susceptible parents. These genes are strong candidates for mediating BCA resistance.

Given the complexity of resistance mechanisms, we plan to broaden the search for candidate genes using transcriptome-wide analysis of aphid-induced responses in the selected genotypes. To address structural variations in *H. spontaneum* genomes, we will develop an annotated reference genome for Hsp5. Promising candidate genes will be validated through knockout and overexpression studies in Hsp5 and the cultivar Golden Promise. The resulting transgenic lines will be evaluated for resistance to BCA and other aphid species, including the English grain aphid (*Sitobion avenae*) and green peach aphid (*Myzus persicae*).

This research aims to elucidate aphid-host interactions and provide genetic markers to accelerate the breeding of BCA-resistant barley, contributing to more sustainable crop protection strategies.

## **P9 – Interaction between HCPro and ARGONAUTE1 contributes to vertical and horizontal transmission of turnip mosaic virus**

Molecular plant-microbe interactions

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Potyvirus are the largest group of positive-stranded RNA viruses in plants and the source of infections affecting the production of economically valuable crops worldwide. While potyviruses are mainly transmitted by aphids in a non-persistent manner, recent research suggests that the frequency and biological relevance of vertical transmission via seeds has been hitherto underestimated.

Turnip mosaic virus (*Potyvirus rapae*, TuMV) is seed-transmitted via direct embryo invasion in many *Brassicaceae* hosts. In a TuMV infection virion stability and optimal long-distance movement require an interaction between the potyviral helper component proteinase (HCPro) and host Argonaute 1 (AGO1). We investigated the effects of the HCPro-AGO1 interaction on aphid, seed, and mechanical transmission using a previously constructed TuMV mutant (TuMV<sup>AG</sup>) where HCPro's capacity to interact with AGO1 is disrupted.

Both horizontal and vertical transmission was reduced in TuMV<sup>AG</sup> compared to the control virus. In *Nicotiana benthamiana* the aphid-transmission rate of TuMV<sup>AG</sup> was 50 % of the control. While grow-out tests combined with detection by RT-qPCR revealed that TuMV was not seed-transmissible in *Nicotiana benthamiana*, similar experiments in *Arabidopsis thaliana* showed that the seed transmission rate of TuMV<sup>AG</sup> was five-fold lower than that of the control. Furthermore, mechanical inoculation experiments showed TuMV<sup>AG</sup> infections were significantly delayed.

The study suggests that the HCPro-AGO1 interaction is necessary for efficient TuMV transmission, likely because of low virion stability and slow systemic movement. Overall, the findings enhance our understanding of the dynamic relationships between molecular virus-host interactions, virus movement and transmission.

## **P10 - Interrelationship and impact of two bacterial pathogens on growth, reproduction and crude protein content of four crops**

Molecular plant-microbe interactions

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Tomato, eggplant, potato, and chili pepper are key Solanaceae crops valued for their nutritional and health benefits. However, these crops are susceptible to various biotic stress factors, particularly pathogens like *Pectobacterium carotovorum* subsp. *carotovorum* (*Pcc*) and *Pseudomonas syringae* (*Pss*). Studies on the effect of these pathogens on plant growth are commonplace but intricacies such as effects on reproduction and production of crude proteins remain unknown. This study aimed to investigate the effects of *Pcc* and *Pss* on these crops, and we revealed that both pathogens negatively influence several growth parameters, including stem diameter, plant height, number of leaves and fruits per plant, chlorophyll content, and biomass. Correlation analysis highlighted that leaf chlorophyll content, closely linked with growth and yield, was a critical factor affected by these pathogens. The analysis also indicated that changes in one parameter due to pathogen infection could lead to corresponding changes in others, underscoring the interrelationship between growth and physiological parameters. The study suggests that although pathogens like *Pcc* and *Pss* reduce crop performance, variations in crop responses point to potential resistance mechanisms that could inform strategies to enhance resilience.

## **P11 - Investigating *Plenodomus tracheiphilus* infection mechanisms in citrus lemon.**

Molecular plant-microbe interactions

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“Mal secco” is a severe vascular citrus disease (MSD) caused by the mitosporic fungus *Plenodomus tracheiphilus*. It predominantly affects lemon but also impacts citron, bergamot, lime, sour orange, and rough lemon to a lesser extent. This disease is widespread across the Mediterranean region, including the Black Sea area. The symptoms typically begin with chlorosis of the leaf veins and premature leaf drop. The pathogen then progresses to infect twigs and branches, leading to the formation of red discolored strands in the xylem of affected stems. This is followed by the dieback of twigs and branches, eventually resulting in the tree's death. A critical goal for lemon growers and breeders is the identification of resistant or field-tolerant clones and hybrids. To investigate how *P. tracheiphilus* infects and spreads within its host, a GFP (Green Fluorescent Protein)-labeled isolate of the pathogen was developed. This labeling allows direct visualization of fungal invasion and growth within living tissues at fluorescence and/or confocal microscope. Using *Agrobacterium tumefaciens*-mediated transformation, GFP was introduced into a virulent *P. tracheiphilus* isolate. The transformed isolates are tested for their virulence, as determined by their ability to induce symptoms in susceptible lemon plants, compared to the non-transformed isolate. Selected GFP-labeled isolates will be used to analyze the infection processes in both susceptible and tolerant Lemon cultivars. This approach deepens our understanding of host-pathogen interactions but also supports the development of innovative strategies for selecting Citrus cultivars with durable resistance to “Mal secco” disease.

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## **P12 - Investigating the role of secreted phospholipases in fungal virulence; the case of *Verticillium longisporum***

Molecular plant-microbe interactions

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Phospholipases are common enzymes able to hydrolyze phospholipids. They have a crucial role in several biological processes but, their precise role in fungal-plant interactions remain understudied. In this study, the role of a secreted phospholipase A<sub>2</sub> (VIPLA<sub>2</sub>) has been investigated in the soilborne plant pathogenic fungus *Verticillium longisporum*, a global threat for rapeseed cultivation worldwide. The *VIPLA<sub>2</sub>* gene encodes an active phospholipase A<sub>2</sub> and transient expression of this gene in *Nicotiana benthamiana* plants, led to an increased production of certain phospholipids. Further, VIPLA<sub>2</sub> was able to suppress chitin-induced ROS burst and hypersensitive response (HR), triggered by a PRR complex. In addition, fungal *VlsPLA<sub>2</sub>* overexpression strains showed increased virulence, and induction of fungal genes with a confirmed role in pathogenicity. Confocal microscopy showed that VIPLA<sub>2</sub> is initially localized to the host nucleus and was translocated to the chloroplasts at the later time points. Furthermore, our results showed that VIPLA<sub>2</sub> could bind to vesicle associated membrane proteins A (VAMP-A) and transported to the nuclear membrane, where it causes major alterations in genes known for their role in plant immunity. In conclusion, we showed that this phospholipase, is a virulence factor, co-regulating induction of pathogenicity factors in fungal cells. It hijacks the host VAMP-A proteins in order to facilitate entry to the nucleus, where it hydrolyzes phospholipids from the nuclear membrane. This action might act as a signaling cascade, suppressing basal plant immunity responses, such as the Pattern Triggered Immunity (PTI), indicating a crucial role in early stages of the infection.

### **P13 - Investigate the tolerance of commercial oilseed rape cultivars to *Verticillium* stem striping disease**

Molecular plant-microbe interactions

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*Verticillium longisporum*, a soilborne fungal species, is the causative agent of *Verticillium* stripe disease in *Brassica* species and represents a notable threat to oilseed rape (*B. napus*). To date, there are no effective control strategies available to combat this pathogen. Consequently, management of this disease is based on breeding. However, contemporary oilseed rape cultivars exhibit varying degrees of tolerance to this disease, though none manifest substantial resistance. In the present study, the degree of tolerance was investigated in 13 winter-type commercial cultivars that are widely utilized in Sweden. An attempt was also made to correlate any degree of tolerance to plant root architecture, given that previous data demonstrated a possible correlation of *B. napus* tolerance to other soilborne pathogens. To this end, greenhouse experiments were conducted using two *V. longisporum* strains (S22 and S31), isolated from infested soils in Östergötland. The results demonstrated that S22 exhibited a significantly higher virulence compared to S31 in all the tested cultivars. In the case of the S22 infection, only the cultivar "Helypse" exhibited a reduced biomass compared to the other cultivars. In contrast, the spring-type cultivar "Hannah" exhibited heightened susceptibility to the winter-type cultivars in milder infections. The cultivars "Commodore," "Credo," and "Janosh" demonstrated no significant yield reduction. Furthermore, a potential correlation between root size and tolerance was observed, indicating that cultivars with longer roots exhibited enhanced tolerance to *Verticillium* stem stripe disease, at least in milder infections. The experimental findings are currently undergoing further analysis and will be confirmed through field experiments.

## **P14 - Overexpression of a necrosis and ethylene-inducing peptide in *Verticillium dahliae* enhances virulence**

Molecular plant-microbe interactions

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*Verticillium dahliae*, a cosmopolitan fungus that invades the xylem of the host plant is the causal agent of vascular wilts in several plant species worldwide. The fungus is known to possess an arsenal of virulence factors, which are utilized for successful colonization and subsequent development of the disease. The present study aimed to investigate the functional role of VdNEP, an NLP-family protein, as a pathogenicity factor. Initially, eggplant leaves when soaked in a purified protein solution, showed necrosis, simulating vascular wilt symptoms. In order to dissect into the role of the VdNEP in the pathogen's virulence, extra gene copies were incorporated in the fungus. Greenhouse pathogenicity trials in eggplant, cotton and tomato plants, revealed that the overexpressing mutants were more virulent compared to the wild type parental strains. Interestingly, an overexpressing mutant derived from a defoliating strain with high VdNEP transcript levels, successfully managed to cause symptoms in tomato, as opposed to the wild-type defoliating strains that in practice do not infect tomato. More specifically, the pathogen's biomass in greenhouse-grown tomato plants inoculated with this VdNEP-overexpressing mutant, was significantly higher (22 times), compared to the wild type strain. Furthermore, a VdNEP-EGFP fusion protein was constructed, aiming to visualize VdNEP localization in the host cells during infection. A TRV vector was employed to harbor the *VdNEP-GFP* construct that was subsequently infiltrated into the mesophyll of Arabidopsis, cotton and tobacco. The VdNEP protein was able to reproduce necrosis and wilting symptoms without the presence of the pathogen, while EGFP facilitated tracking in the host cells. Confocal microscopy revealed that the VdNEP protein was localized at the subcellular space and attached to the cell membranes, resulting in cell death. These findings further support the critical role of VdNEP in *V. dahliae* infection and host colonization.

## **P15 - Screening *Luffa aegyptiaca* genotypes for resistance to the fungus *Verticillium dahliae***

Molecular plant-microbe interactions

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*Luffa aegyptiaca*, a valuable tropical plant belonging to the family Cucurbitaceae, is susceptible to *Verticillium dahliae*, a soil-borne pathogen that causes significant wilt disease in various host plants. This study evaluated the resistance of several indigenous and non-indigenous *Luffa* genotypes to *Verticillium* wilt. The disease severity index (DI) and the area under the disease progression curve (AUDPC) were assessed for each genotype following infection. A plating assay on PDA culture was conducted to confirm proper colonization of the plants, while real-time PCR and specific primers were utilized to validate fungal colonization and quantify the fungus in plant tissues. The results classified the *Luffa* genotypes into two groups, highly susceptible and susceptible, based on the severity of disease progression. Data analysis revealed that the Angled genotype exhibited the lowest sensitivity to *V. dahliae*, with a disease index of 18%. Both the re-isolation assay on PDA plates and real-time PCR confirmed the colonization and pathogenic effects of *V. dahliae* on *Luffa* genotypes. However, due to limited studies on the genetics of resistance to *Verticillium* in *Luffa*, further research is required to investigate the plant defense mechanisms, particularly in the less susceptible genotype identified in this study.

## **P16 - Phytohormonal signaling in resistant and susceptible apricot varieties in response to plum pox virus**

Molecular plant-microbe interactions

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Understanding the molecular mechanisms underlying disease resistance is crucial for improving the resilience of fruit crops. This study investigates the hormonal responses in two apricot varieties, Rojo Pasi3n (resistant) and Z506-7 (susceptible), following infection with *Plum Pox Virus* (PPV). We focused on the levels of key stress hormones (ACC, ABA, JA, and SA) and growth regulators (tZ, GA3, GA4) in both varieties, comparing their regulation before and after infection. The results revealed significant differences in hormonal regulation between the resistant and susceptible varieties. In Rojo Pasi3n, the resistant variety, we observed a marked increase in ACC levels, accompanied by a significant decrease in ABA, suggesting the activation of ethylene-dependent defense pathways and a shift in hormonal balance toward defense mechanisms. In contrast, the susceptible Z506-7 variety exhibited a reduction in ACC and only a moderate decrease in ABA, indicating impaired defense responses and greater susceptibility to viral infection. Both varieties showed a reduction in JA levels, but the decline was more pronounced in the susceptible variety, further supporting the hypothesis of compromised defense in Z506-7. Interestingly, the growth hormones tZ and GA3 exhibited a less dramatic decrease in Rojo Pasi3n, suggesting a better balance between growth and defense responses. However, in the susceptible variety, the increase in GA3 was more pronounced, reflecting a misregulation of growth under viral stress. These findings highlight the importance of hormonal regulation, particularly the modulation of ABA and ACC, in the resistance of apricot varieties to PPV. Our study provides insights into the molecular mechanisms of disease resistance and offers potential strategies for enhancing PPV resistance in apricot cultivars.

## **P17 - Phytohormone signaling in GF305 peach rootstock infected with PPV, HSVd, and ACLSV**

Molecular plant-microbe interactions

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This study investigates the effects of *Plum Pox Virus* (PPV), *Apple Chlorotic Leaf Spot Virus* (ACLSV), and *Hop Stunt Viroid* (HSVd) on the phytohormonal profiles of GF305 peach rootstock. The hormones considered include those involved in stress responses (ACC, ABA, JA, SA) and growth regulation (tZ, GA3, GA4). The results indicate that viral infections significantly alter the levels of phytohormones, with distinct patterns observed for each pathogen. In control plants, cytokinin (tZ) and gibberellins (GA3 and GA4) are present at high levels, suggesting a predominant role in growth and development. In contrast, the infected plants exhibited considerable variation in hormone concentrations. PPV infection notably increased the levels of stress-related hormones ABA and SA, while significantly reducing cytokinin (tZ) and gibberellins. ACLSV-infected plants showed elevated levels of JA and SA, with a substantial reduction in GA3 and GA4, indicating a possible shift towards stress resistance. HSVd infection had a moderate effect, with slight increases in tZ and JA but no notable change in growth-related hormones. These results suggest that viral infections modify the hormonal balance in GF305 rootstocks, emphasizing the role of ABA, JA, and SA in stress responses. The alteration of growth-related hormones, such as tZ and gibberellins, further supports the impact of viruses on plant development. Understanding these hormonal shifts can inform future breeding programs aimed at enhancing resistance to these viruses. By selecting or engineering rootstocks with optimal hormonal profiles, it may be possible to develop peach cultivars with improved resistance to viral infections, contributing to sustainable agricultural practices.

## **P18 - Screening baby leaf salad greens for host resistance to downy mildew isolates across the central coast of California**

Molecular plant-microbe interactions

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Downy mildew poses a significant threat to baby leaf salad green production in California. Baby kale, a key component of these salad mixes, holds substantial economic value, with a crop worth over \$12 million in 2022. Among the various baby kale species, *Brassica oleracea* is the most widely grown. The disease, caused by *Hyaloperonospora brassicae*, results in chlorosis, necrosis, and sporulation, making leaves unmarketable. Developing resistant varieties can offer a sustainable approach to disease management by reducing pesticide reliance. This study evaluated baby kale accessions for resistance to downy mildew isolates across California. Plants were inoculated with an *H. brassicae* sporangia suspension and incubated under high humidity. Disease severity was assessed based on chlorotic and necrotic symptoms and sporulation on a scale of 0 to 100%. An initial screening of 212 baby kale accessions using an isolate collected from a breeding field in Gilroy showed an average disease severity of 28%, with values ranging from 0 to 100%. Further subset screenings revealed severity averages between 2.2% and 9.4% inoculated with isolates collected from various fields. The final subset screening demonstrated disease severity ranging from 0.003% to 0.072% across four isolates. Notably, 13 accessions exhibited 100% estimated resistance probability, 11 ranged between 99.0% and 99.9%, and one fell below 99.0%. These results suggested that breeding resistant baby kale varieties is a promising strategy for managing downy mildew, minimizing crop losses, and supporting sustainable production.

## **P19 - Screening for genetic variation against the major causes of legume fatigue in a broad panel of *Pisum sativum***

Molecular plant-microbe interactions

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Legume fatigue is considered as one of the main reasons for the lack of yield stability and profitability in grain legume crops. It is characterized by general growth depression, which can lead to yield reduction or even total crop failure. A complex interplay of factors leads to legume fatigue, with fungal pathogens such as *Fusarium* species, *Didymella* species and *Phoma* species being the primary causes in peas. The problem of legume fatigue increases with the expansion of legume production. Both organic and conventional farming needs cultivars with high tolerance or resistance.

To support breeding efforts, a standardized method for testing susceptibility to the major legume fatigue pathogens is being developed and standardised. This method will form the basis for testing per year up to 250 genetic resources and cultivars. At the same time, provocation sites for legume fatigue are being established, where selected genotypes will be tested for resistance to legume fatigue under field conditions. In addition to assessing phenotypic traits, symptoms will also be recorded using drone-based multispectral reflectance measurements. The species composition of the pathogen complex will be characterised and analysed using DNA sequencing methods from root samples in order to make more precise statements about the complexes responsible. In addition, the soils of the provocation sites will be sampled throughout the trial to characterize the soil microbiome and identify other possible influencing factors.

This work is part of the DiPisum project, which aims to strengthen and establish pea as an important and promising crop. The ultimate goal of this work package is to gain a deeper insight into the complexity of legume fatigue and to identify potential sources of tolerance or resistance. Using phenotypic and genomic data, genome-wide prediction models will be developed and providing valuable tools to support both pea breeding and cultivation.

## **P20 - Screening of kikuyu yellows and black spot resistance in kikuyu (*Pennisetum clandestinum*) cultivars**

Molecular plant-microbe interactions

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Kikuyu (*Pennisetum clandestinum*) is a perennial grass widely used in grazing systems for beef and dairy production worldwide due to its rapid growth and adaptation to harsh conditions. However, the main Australian cultivar, Whittet, is extremely susceptible to the fungal diseases, kikuyu yellows caused by *Verrucalvus flavofaciens*, and black spot caused by *Bipolaris* spp. There are currently no commercial fungicides to control kikuyu yellows and, although there are fungicides that are effective against black spot, fungicides are probably uneconomic for broadacre use. Therefore, breeding for resistance to these two diseases has been a focus when selecting for new kikuyu cultivars. A study was conducted to examine variation in resistance to black spot and kikuyu yellows in three promising kikuyu selections with superior dry matter production and nutritive value in comparison with a newly-released cultivar, Fulkerson, which has some resistance to both diseases and Whittet, the susceptible common cultivar. Cultures of *Bipolaris* sp. and *Verrucalvus flavofaciens* were isolated from diseased kikuyu leaves. For black spot, a spore suspension of *Bipolaris* sp. was inoculated onto kikuyu leaves of four replicate pots of each kikuyu cultivar or selection. Negative controls were inoculated with sterile distilled water. The pots were placed in a humid chamber for 48 hours to encourage infection. Black spot was assessed at 3, 5 and 8 days after inoculation. For kikuyu yellows, agar inoculum of the fungus was used to inoculate the root zone of kikuyu plants of four replicate pots of each of the above cultivars and selections. Controls were not inoculated with the pathogen. The kikuyu plants were rated for disease symptoms after 4 and 8 weeks. Results from these pot experiments could assist in deciding whether to further test the promising selections for disease resistance in the field before considering them for commercialisation.

## P21 - Secreted proteases of *Fusarium graminearum*: What are their roles in fungal virulence?

Molecular plant-microbe interactions

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Fungal pathogens secrete a wide range of proteases during plant infection. Some of these proteins facilitate pathogen colonisation in the host plant and contribute to disease development. Fusarium head blight is one of the significant wheat diseases caused by *Fusarium graminearum*.

Understanding the disease mechanism caused by this fungus is crucial for designing new disease resistance strategies. However, many of the underlying disease-causing mechanisms in this interaction, including the function of *F. graminearum* secreted proteases, remain poorly understood. To better understand the role of those secreted proteases during fungal infection, protease genes expressed during *in planta* growth were identified from the available RNA-seq dataset and selected for further characterisation. One of the most highly expressed proteases during *in planta* growth is a predicted trypsin protease, *FgTPP1*. FgTPP1 is a secreted protease containing a functional signal peptide, which has been validated using a yeast secretion system. The  $\Delta Fgtpp1$  mutants were further generated to assess the role of this protease in fungal virulence. In a bottom spike infection assay,  $\Delta Fgtpp1$  mutants had a significantly lower percentage of fully bleached spikes than wild-type *F. graminearum*. Additionally, the FgTPP1 fused with GFP was transiently expressed in *Nicotiana benthamiana* leaves to analyse its subcellular localisation in plant cells and its involvement in plant immunity. This analysis revealed that FgTPP1 localised to the plant chloroplast and suppressed both the activation of the mitogen-activated protein kinase signalling pathway and the reactive oxygen species burst induced by the chitin elicitor in *N. benthamiana* leaves. Apart from FgTPP1, several other secreted proteases of *F. graminearum* are also expressed during spike infections. The role of these other secreted proteases is currently being investigated to define their overall contribution to *F. graminearum* virulence during wheat spike infection.

## **P22 - Study on the resistance of the new breeding lines of oilseed rape (*Brassica napus* L.) to turnip yellows virus (TuYV)**

Molecular plant-microbe interactions

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Turnip yellows virus (TuYV) is one of the most common viruses. This species belongs to the genus *Polerovirus*, family *Luteoviridae*. It has a wide host range and infects over 150 species from 23 families of dicotyledonous plants, including important crops such as lettuce, spinach, faba bean, radish, cabbage and oilseed rap (Graichen, 1996). TuYV occurs mainly in the phloem tissue of host plants and is transmitted by the following aphids: peach-potato aphid (*Myzus persicae*), potato aphid (*Macrosiphum euphorbiae*) and cabbage aphid (*Brevicoryne brassicae*). The limitations associated with the protection of *Brassica napus* plants against aphids are often related to the increased activity of TuYV. Therefore, all breeding work aimed at obtaining oilseed rape plants resistant to turnip yellows virus is very important.

For resistance studies and alternating crosses, *B. napus* genotypes with the cytoplasm of cabbages (kale, fodder cabbage - Chorinska, brussels sprouts, *B. taurica*) obtained in 2023 and 2024 were used. These plants are characterized by significant resistance to *Plenodomus* spp. (formerly *Leptosphaeria* spp.) and *Alternaria* spp. Varieties with declared resistance to TuYV were also used for the studies. New lines were sown in greenhouse conditions. In the 5-leaf phase, leaf fragments were taken to isolate genomic DNA and inoculation with insect vectors (aphids) was carried out. Before leaf fragments were collected for ELISA tests, observations of characteristic symptoms of turnip yellows virus were made one month after inoculation. On the other hand, reactions with two SCAR markers adapted from the literature (Juergens et al., 2010) were performed from isolated DNA. Although for some genotypes the PCR products were nonspecific (combination of the banding pattern determined for both susceptible and resistant plants), it was possible to select new oilseed rape lines with increased resistance to TuYV.

## **P23 - Susceptibility of nine Greek olive cultivars to *Colletotrichum* spp. strains causing olive anthracnose disease in Greece**

Molecular plant-microbe interactions

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Olive anthracnose, caused by various *Colletotrichum* spp., is a major fungal disease affecting olive production, leading to significant yield losses and deterioration of olive oil quality. In Greece, different *Colletotrichum* species, including *C. acutatum*, *C. gloeosporioides*, and *C. nymphaeae*, have been identified as the primary causal agents. This study aimed to evaluate the susceptibility of nine Greek olive cultivars to five *Colletotrichum* spp. strains. The selected cultivars included six oil-producing ('Mavroelia Messinias,' 'Koroneiki,' 'Ntopia Atsicholou,' 'Mastoides,' 'Koutsourelia,' and 'Myrtolia'), one dual-purpose ('Manaki'), and two table olive cultivars ('Karydolia Chalkidikis' and 'Amygdalolia'). As part of the above objective, detached olive drupes at a comparable ripening stage were collected and artificially inoculated using a standardized wound inoculation method. Disease severity was assessed at 4, 6, and 8 days post-inoculation using a commonly applied 0–5 scale. The results revealed significant differences in cultivar susceptibility. 'Ntopia Atsicholou' and 'Koroneiki' exhibited the highest tolerance to all tested strains, followed by 'Manaki,' which showed moderate resistance. 'Mastoides' demonstrated strong resistance to a *C. acutatum* strain, previously reported as the first *C. acutatum* strain causing olive anthracnose in Greece but had variable responses to other strains. In contrast, 'Koutsourelia,' 'Myrtolia,' 'Mavroelia Messinias,' and 'Amygdalolia' were the most susceptible, displaying severe symptoms across multiple pathogen strains. These findings provide critical insights into the selection of resistant cultivars for integrated disease management strategies, optimizing olive production and reducing the need for chemical control measures.

## **P26 - On the hunt for ecological effectors in major wheat pathogens.**

Molecular plant-microbe interactions

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Wheat powdery mildew, caused by the biotrophic fungus *Blumeria graminis* f. sp. *tritici*, is one of the most important pathogens of wheat. Like other biotrophic pathogens, *B.g. tritici* infection is strongly driven by its effectors and their interactions with the host, and probably also by other pathogens and microbes co-occurring in the same ecological niches. The latter hypothesis prompted us to study the possible ecological roles of mildew effectors with the hypothesis that some effectors might have specifically evolved to counteract challenge from other microbes. We generated large RNA-sequencing datasets from different field environment (i.e. natural pathobiomes) and from artificial co-infections with *Zymoseptoria tritici* (a hemibiotrophic foliar pathogen of wheat). In addition, we have also analyzed RNA-seq data originating from coinfection experiments with *Goluebia* sp., a biocontrol agent antagonizing mildew. Our results suggest there is a conserved, environmentally stable induction of numerous effectors irrespective of the microbial challenge impose on mildew, with noticeable differences between highly diversified effector families vs. single copy effectors. Altogether, our results suggest that large arrays of mildew effectors (including putative antimicrobial factors) are probably employed to create a stable ecological niche associated with sporulation.

## **P28 - Unveiling cryptic bioactive metabolites in tomato-*Serendipita* interactions: a multimodel approach**

Molecular plant-microbe interactions

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*Serendipita* is an endophytic fungus with plant growth promoting properties. During colonization, *Serendipita* circumvents the host immune response to prevent formation of extensive necrotic zones. Therefore it is hypothesized that *Serendipita* produces a variety of cryptic bioactive metabolites that contribute in altering the host's metabolism. The majority of these metabolites still await discovery as they remain hidden under laboratory growth conditions. For this project, a pipeline was created to discover and characterize metabolic interactions between fungi and host, facilitating the identification and production of yet undiscovered metabolites.

In this pipeline, tomato (*Solanum lycopersicum*) wild type and mutant lines compromised in salicylic acid (SA) and jasmonic acid (JA) dependent signalling pathways will be exposed to *Serendipita* spp., to stimulate the production of cryptic secondary metabolites. Two complementary experimental systems will be employed: a plant- and a hairy root-based system and a hairy root system. In the plant-based system, tomato responses to *Serendipita* colonization are monitored using hyperspectral and multispectral imaging techniques, while fungal colonization will be determined using microscopic imaging and qPCR. In the hairy root system, growth and phenotypic changes will be analysed using a root scanner. Insights gained on tomato-*Serendipita* interactions will be the groundwork for subsequent metabolic profiling, gaining a deeper understanding of the *Serendipita* metabolome and biochemical crosstalk between *Serendipita* and tomato.

## **P29 - *Phytophthora cactorum* RXLR effectors RXLR11, RXLR14 and RXLR17 regulate plant immunity and are essential for infection**

Molecular plant-microbe interactions

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*Phytophthora cactorum* causes devastating diseases in many fruit crops, ornamental and landscape plants. The pathogen harbours hundreds of RXLR effector genes encoding secreted proteins that are considered important for virulence as some *Phytophthora* RXLRs have been shown to manipulate host plant cells and suppress immunity. The exact role of most *P. cactorum* RXLR effectors during plant infection is unknown. We have studied twenty such effectors from *P. cactorum* that are expressed during infection of strawberry crowns. Some of the RXLR effectors induced cell death and enhanced callose deposition when transiently expressed in *Nicotiana benthamiana* leaves, indicating that these effectors activate the plant immune system. Of the 13 RXLR effectors that did not induce cell death, PcRXLR11 and PcRXLR14 suppressed cell death induced by the cell death-inducing protein INF1 from *P. infestans* and PcRXLR17 enhanced cell death when co-infiltrated with INF1. *In planta* expression of these effectors showed that the cell death suppressing effector PcRXLR11 promoted virulence of *P. cactorum*. N-terminal tagging of the PcRXLR11, PcRXLR14 and PcRXLR17 effectors with YFP revealed diverse subcellular localisations including chloroplast, cytoplasm and/or possibly plasma membrane. These three effector genes were found to be upregulated during pathogen infection in strawberry leaves compared to the mycelial stage. An initial experiment using fluorescein-labelled double-stranded RNA (dsRNA) showed that the dsRNA was taken up by mycelium, sporangia and oospores of *P. cactorum*. Silencing of these three effector genes using dsRNA targeting the effector's mRNA resulted in reduced infection in the strawberry leaves, suggesting that these effectors are essential for *P. cactorum* infection. These findings highlight the role of RXLR effectors of *P. cactorum* in plant immune suppression and virulence as well as a potential strategy to control important *P. cactorum* diseases in plants.

### **P93 - Adhesins and oxylipins influence the lifestyle of *Xylella fastidiosa***

Molecular plant-microbe interactions

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Lipids are crucial in shaping the lifestyle of the xylem-limited pathogen *Xylella fastidiosa*, which ranges reversibly from planktonic to twitching to biofilm. Notably, Diffusible Signalling Factors (DSF) (i.e. cis-2-enoic fatty acids) are implicated in regulating the lifestyle of this dangerous pathogen. It is believed that the symptoms caused in diverse hosts are driven by the formation of biofilm within the xylem vessels. The adhesion to xylem cell walls appears crucial in determining biofilm formation. Recently, we demonstrated that the oleic acid-derived oxylipin 7,10-DiHOME could be a crucial signalling molecule to promote biofilm formation and twitching motility, whose synthesis likely depends on a signal transduction requiring the presence of the afimbrial adhesin XadA2, and thus not working if this protein is depleted. Outcomes obtained provide a hypothesis on the mechanism that can control these fundamental switches *in vitro*. But what occurs within the host? Considering the case of OQDS (olive rapid decline syndrome), plant oxylipins, especially those derived from the 13-lipoxygenase (13-LOX) pathway, may help bacterial oxylipins in controlling lifestyle transitions in *X. fastidiosa*. In the plant model organism *Arabidopsis*, a deletion of the LOX2 gene, a 13-LOX, involved in jasmonate synthesis, reduced 13-oxylipins, paving the way for the growth and movement of the pathogen in the plant. This result was also validated under natural infection conditions in olive. The oxylipin 13-HODE, whose biosynthesis is catalysed by plant 13-LOX, accumulates in susceptible olive varieties compared to resistant ones. Apparently, a circuit composed by oxylipins and adhesins is involved in the lifestyle transitions of *X. fastidiosa*. Deciphering the relationship between these will provide insights into finding a tailored solution to control the *X. fastidiosa*

## **P117 - How pulsed electric fields modulate the transcriptomic response of malting barley to *Fusarium* infection**

Molecular plant-microbe interactions

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Barley (*Hordeum vulgare*) is a key raw material for brewing and food industries, but its quality can be compromised by *Fusarium* pathogens responsible for Fusarium Head Blight. Malting is a crucial step in processing barley, yet *Fusarium* infection during this phase can negatively impact grain quality and safety. Pulsed electric field (PEF) treatment is a non-thermal microbial control methods used in food and crop processing, with potential applications in improving malting efficiency. However, the molecular mechanisms underlying barley's response to both *Fusarium* infection and PEF treatment during malting remain largely unexplored.

To address this, we investigated transcriptomic changes in *Fusarium*-infected barley subjected to malting, with and without PEF treatment. RNA sequencing revealed over 12,000 differentially expressed genes (DEGs) across four malting stages, with the most significant transcriptional shifts occurring after 24 hours of germination. Functional enrichment analyses indicated that malting induces strong stress responses, including oxidative stress mitigation and hormone-regulated defense mechanisms.

PEF-treated barley exhibited fewer DEGs in later stages compared to untreated samples, suggesting that PEF modulates stress pathways and enhances stability during malting. Key upregulated genes in PEF-treated barley included those linked to abiotic stress resistance, while downregulated genes were associated with photosynthesis and cell wall remodeling. These findings provide new insights into the interplay between malting, pathogen stress, and PEF treatment, highlighting its potential to improve malting efficiency and barley resilience.

### **P130 - Decoding KVDS: Integrating microbial, genetic, and hormonal signatures in a multifactorial framework**

Molecular plant-microbe interactions

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Since its emergence in 2012, Kiwifruit Vine Decline Syndrome (KVDS) has posed a significant threat to the Italian kiwifruit industry. This complex and multifactorial syndrome presents substantial challenges for growers and production systems. Affected plants exhibit symptoms including root browning, loss of feeding roots, cortical layer hypertrophy, and detachment, followed by rapid whole decline, particularly during increased summer temperature. While several biotic stressors have been implicated as potential contributors to KVDS, its multifactorial nature necessitates an integrated management approach. To gain a deeper understanding of this complex disease, this study employs a multi-omics approach, to uncover novel insights into its etiology and to address proper control strategies. Root samples were collected from symptomatic and asymptomatic kiwifruit plants at two key phenological stages: end of flowering (June) and at fruit set (July). Metagenomic analysis revealed significant shifts in the microbial community composition of KVDS-affected roots, confirming the involvement of previously implicated genera. Gene expression analysis identified significant alterations in the expression of 17 genes associated with plant defence responses, including PRRs, ROS detoxification, PR and R proteins synthesis, and hormone signalling pathways. Furthermore, HPLC analysis revealed distinct phytohormone profiles in KVDS-affected plants compared to asymptomatic controls, suggesting perturbations in hormone homeostasis. These findings provide novel insights into the complex etiology of KVDS and could provide a foundation for developing effective management strategies.

## **P131 - Extracellular vesicles of *Dickeya* spp.: Characterization and predicted role in plant interactions**

Molecular plant-microbe interactions

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Bacteria of the *Dickeya* genus can secrete plant cell wall degrading enzymes (PCWDEs), which contribute to the maceration of plant tissue and the development of soft rot disease in economically important crops. These enzymes and other virulence factors, are released outside the bacterial cell via various secretion systems, including extracellular vesicles (EVs). However, the secretion via EVs by bacteria of the genus *Dickeya* has not yet been described. Our study aimed to isolate and characterize the EVs secreted by the *Dickeya dadantii* 3937 and *Dickeya solani* IPO 2222 strains, and to explore their role in pathogenicity. After isolating the EVs, we employed nanoparticle tracking analysis and transmission electron microscopy to visualize and characterize them. We conducted a proteomic analysis of the proteins present in the EVs using MALDI TOF/TOF mass spectrometry. Additionally, we performed *in silico* analysis to identify potential interactions between the bacterial proteins secreted via vesicles and the proteins of *Arabidopsis thaliana*. Finally, we established the effects of direct interactions between the bacteria and plant through the secreted EVs *in vitro*. All strains tested produced EVs of similar size; however, we observed changes in the number of secreted vesicles when mutations were introduced. The proteomic analysis indicated that EVs secreted by all tested bacterial strains contained virulence factors, which may potentially interact with proteins of *A. thaliana*, regardless of the tested mutations. *In vitro* analyses showed that substances secreted by bacteria into the medium could alter the root morphology of *A. thaliana*. These results suggest that EVs secreted by *Dickeya* spp. play a significant role in the pathogenicity of these bacteria towards plants.

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## **P132 - Genome-wide association studies identify the mating-type locus and avr candidate genes in grapevine downy mildew**

Molecular plant-microbe interactions

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The availability of high-quality diploid reference assemblies for oomycetes enables genome-wide association studies (GWAS) to link genotype and phenotype variation, identifying the genomic architecture of pathogens' life-history traits. In *P. viticola*, mating can occur only between individuals of different mating types (heterothallism). Using GWAS, we identified a genomic region of 570 kb associated with the mating-type phenotype (Dussert et al.). P2 individuals were homozygous for the MAT-a allele at the mating-type locus, whereas P1 individuals were heterozygous, carrying the MAT-a and MAT-b alleles. The mating-type region features a gene that encodes a transmembrane protein that might act as a hormone receptor; this is noteworthy since hormones have previously been identified as mating-type factors in *Phytophthora* spp. Our subsequent research delved into the genomic factors that drive the breakdown of grapevine's partial resistance to downy mildew, specifically focusing on Rpv3 (Paineau et al.). Using GWAS, we discovered a distinct structural variation exclusively present in the genomes of strains that are virulent on grapevines carrying the Rpv3 locus. The structural variation consisted of a deletion of 30 kb encompassing two closely-related genes that encode proteins of 800-900 amino acids with a signal peptide. The predicted structures of both proteins contain repeats that form structural elements typical of the LWY-fold, a conserved structural module in oomycete effectors. While our current knowledge of the molecular interactions between downy mildew and its host is still in its early stages, our research contributes to expanding this knowledge and opens avenues for further exploration of gene-for-gene interaction in this pathosystem. The recent discoveries regarding the breakdown of other partial resistances in grapevine (Rpv1, Rpv10, Rpv12) pave the way for future exploration of new effectors in *P. viticola* using similar approaches.

Dussert et al. 2020. Current Biology, 30.20: 3897-3907

Paineau et al. 2024. New Phytologist, 243.4: 1490-1505

### **P133 - Methyl jasmonate seed treatment enhances Norway spruce seedling resistance to grey mold via various defense responses**

Molecular plant-microbe interactions

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Millions of conifer seedlings are grown annually in forest nurseries to support of forest production. *Botrytis cinerea* is a significant pathogen in forest nurseries, causing high mortality in Norway spruce (*Picea abies*) seedlings. Our study assessed the efficacy of seed treatments with MeJA, pipelicolic acid (PipA), lignan (Li), and chitosan (Chi) to enhance Norway spruce seedling resistance against *B. cinerea*. MeJA treatment significantly reduced seedling mortality. This enhanced resistance correlated with increased chitinase activity and accumulation of flavan-3-ols (catechin and proanthocyanidin B1). Transcriptomic data indicated an increase in oxidative stress protection, cell wall reinforcement, and pathogenesis-related protein production in seedlings from MeJA-treated seeds. Our data also suggested an antagonistic relationship between abscisic acid (ABA) and jasmonic acid (JA)/ethylene (ET) hormonal signaling. Overall, our findings showed that MeJA seed treatment enhances seedling resistance against *B. cinerea* through a complex interplay of defense responses and regulatory systems. These insights highlight the potential of MeJA as a valuable tool in forest nursery integrated pest management.

## **P134 - Natural existing arbuscular mycorrhizal-bacterial biofilm associations and their functional behavior**

Molecular plant-microbe interactions

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In symbiosis with plant, arbuscular mycorrhizal fungi (AMF) access the carbon stored in the roots in exchange for increased uptake of nutrients and water from the soil. In the mycorrhizal-plant symbiosis, AMF-associated bacteria (AAB) serve as a third partner and are tightly linked to AMF. AAB are involved in mycorrhizal activity and nutrient uptake enhancement and have impact on plant development. In order to create innovative biofertilizer for sustainable crop production, it is important to understand the function and process of this inter-kingdom natural coexistence. In our research, we used in vitro and in situ co-cultures to screen 33 different AMF species, and we characterized 231 AAB using 16S rDNA analysis. 109 selected AABs were examined for ten functional qualities that promote plant growth, and it was found that different bacterial strains had a variety of advantageous traits. The association of AAB was seen as biofilm and endobacteria using microscopic methods. Further, by using an in vitro assay system, an association recreation of 12 AAB-*Rhizophagus irregularis* was investigated to look at the impact on mycorrhization and functional capabilities. It was observed that AABs moved along the developing *R. irregularis* hyphae and spores. Different AAB had an impact on AMF development as well as its capacity to solubilize phosphate and potassium, and fix nitrogen. We discovered both the synergistic interactions and partnerships between the two cross-kingdom microbial partners. Understanding the molecular elements of these fungal-bacterial connections, which will enable their later use and modification for sustainable agriculture practices, is another area of focus.

## **P135 - The SWEET solution: Role of OsSWEETs in conferring broad spectrum resistance against drought and blast disease**

Molecular plant-microbe interactions

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**Background:** To feed the projected human population of about 9.3 billion, the food production needs to be tripled by 2050. Rice is an important staple crop for almost half of the existing human population of Earth. Blast disease, caused by the hemibiotrophic ascomycete fungus *Magnaporthe oryzae* poses a serious threat to rice production with a reported loss of 30% annually. We lose 50% of global rice production every year due to drought and additionally, it makes the plants more susceptible to blast disease.

**Significance of the study:** It becomes essential to analyse plants under combinatorial stresses. Choosing the correct gene is essential due to the antagonistic mechanisms implied by biotic and abiotic stresses.

**Hypothesis:** One such gene family can be *OsSWEETs* which encode bidirectional sugar transporters in rice. Their involvement in disease susceptibility has only been recently demonstrated in bacterial blight of rice. Their role in imparting drought tolerance has also been elucidated.

**Objectives:** To study the role of rice *SWEETs* in nutrient acquisition mechanism of *Magnaporthe oryzae* while colonising rice plant cells.

**Major Findings:** We found 22 *OsSWEET* genes in the indica subspecies of rice. The 11 newly identified *SWEET* genes of *O. sativa* indica group were revalidated by using different bioinformatic platforms. The differential expression pattern of the genes under biotic stress (Blast disease), abiotic stress (intermittent drought) and also in combination of both the stress conditions. The expression of *SWEET* genes is significantly increased in drought conditions when compared to pathogen challenge alone. The expression pattern in the combinational stresses of both drought and blast disease gives similar pattern irrespective of the variety type. The differential expression pattern signifies the probable involvement of R gene Pi-54 for *OsSWEETs* mediated tolerance.

**Conclusion:** The differential expression pattern highlighted the involvement of *OsSWEET* genes in response to both blast and drought stress.

## **P136 - Uncovering the role of *Serendipita willimasii* volatiles in establishing its own plant beneficial endophytic colonization**

Molecular plant-microbe interactions

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There has been a growing interest in plant responses upon exposure to the volatilome of different fungi. However, knowledge of the composition of these fungal volatilomes and how they affect the plant's physiology is scarce.

The genus of *Serendipita* is an endophytic fungus who can increase the plant's resilience upon abiotic and biotic stress through its volatilome. A combination of better nutrient uptake through beneficial root colonization and protection against stresses with its volatilome makes *Serendipita* of high potential for advancing sustainable crop production.

To understand how *Serendipita* establishes symbiosis with plants, we developed an innovative approach to assess the content and bioactivity of the *Serendipita* volatilome. First, we identified the most abundant volatile organic compounds (VOCs) in the headspace of in vitro grown *Serendipita williamsii* using TD-GC-MS. In parallel, we investigated the role of *Serendipita*'s volatilome in the root colonization in tomato using an experimental setup that eliminates CO<sub>2</sub> fertilization effects. Strikingly, a higher susceptibility of tomato plants exposed to VOCs was observed to *Botrytis cinerea* compared to unexposed plants. A cutting-edge technology, hyperspectral imaging was applied for phenotyping studies. We will present insights into key volatiles, innovative experimental setups, and proposed modes of action.

## **P152 - Sources of resistance to wheat stem rust from the Danish breeding company Agrologica and gene banks.**

Molecular plant-microbe interactions

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In a project supported by the Danish Foundation for Organic Farming, we tested the resistance against wheat stem rust in the Danish plant breeding company Agrologica's wheat breeding program. Additionally, we investigated potential sources of new resistance in plant material sourced from international gene banks.

Wheat stem rust is potentially the most yield-reducing disease in wheat, with losses of up to 100% in case of severe outbreaks. For many years, wheat stem rust was of minor importance in Europe, but since 2016, it has made a comeback as a significant disease.

We tested approximately 600 wheat lines from Agrologica and found that 22 lines had a high level of resistance against five recent and highly virulent stem rust strains, including those responsible for recent epidemics in Europe. Genetic markers indicated that 14 of these lines most likely contain resistance genes already known to confer resistance to the tested strains. The remaining lines may contain previously undiscovered resistance, but further testing is necessary to confirm this.

In addition, we tested 508 lines of various wheat species, including durum wheat and emmer wheat, from the IPK and Czech gene banks. 136 lines showed effective resistance and in a selection of 26 lines, genetic markers revealed that 13 most likely contain known resistance genes. The remaining lines may be potential sources of new resistance.

In selected lines from both Agrologica and the gene banks, we also found indications of adult plant resistance, including detection of the Sr2 gene. Our discovery of broad-spectrum resistance in Agrologica's breeding program and the gene bank material is encouraging for the protection of European wheat production against stem rust.

## **Biocontrol, microbiomes and biostimulants**

## O1.1 - *Heterodera schachtii* biocontrol using *Pleurotus ostreatus* mycelium – practical solutions for field applications

Biocontrol, microbiomes and biostimulants

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*Heterodera schachtii* is a cyst nematode pathogenic to sugar beet crops. Its control is complicated. Chemical control always brings some environmental costs, thus, biological methods are demanded. One of the biological ways of the possible control of *H. schachtii* is the usage of fungal mycelia that are antagonistic or pathogenic to nematodes. One of this type of fungi is *Pleurotus ostreatus*. *P. ostreatus* (oyster mushroom) is one of the most widespread mushrooms in the world. The vegetative mycelium of this mushroom produces hyphal knobs with toxic droplets called toxocysts, that paralyze nematodes. Mycelium uses their bodies as a source of nutrients. This feature gives a great practical weapon against pathogenic nematodes. In our research, we collected wild strains of *P. ostreatus* mycelia, grew them to obtain homokaryotic hyphae, and crossed those hyphae to obtain new combinations of heterokaryons. The maternal mycelia and a sort of homokaryotic and heterokaryotic progenies were tested against a model nematode *Caenorhabditis elegans* and *H. schachtii*. Tests were carried out as laboratory and pot experiments, and also as field research. As a result of these tests and field experiments, we found differences in the nematicidal properties of tested mycelia. We proposed more effective strains for further field applications. The applications of tested mycelia gave the effectiveness at the level of 40 to 60% of reductions of the *H. schachtii* soil populations depending on the strain and the test type (pot or field). We also found various ways of action depending on the nematode type. *C. elegans* were mostly paralyzed by toxocysts but *H. schachtii* preferably were coiled by mycelium. One of the active compounds secreted to toxocysts is 3-octanone, a widespread fungal compound. HS-SPME-GC-MS analyses resulted in differences in 3-octanone production by tested mycelia, however, it was not directly correlated with the effectiveness of mycelia.

## **O1.2 - Deciphering the durum wheat kernels microbiome and metabolome to enhance its resilience to stresses**

Biocontrol, microbiomes and biostimulants

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Durum wheat (*Triticum turgidum* L. subsp. *durum* (Desf.) Husn.) is among the most cultivated cereal crop in the Mediterranean basin area. This cultivar represents a fundamental resource due to its significant role in human nutrition and its importance in the livestock industry. Climate and environmental change endanger the cultivation of key crops, such as durum wheat by influencing their growth and yield, which in turn are also related to the endophytic composition and the plant metabolome. Plant microbiome influencing the host's performance, it is crucial to gain a comprehensive understanding of its composition. By examining the kernel's microbiome composition, it is possible to assess the quality of the kernel before planting, identify the presence of species that produce toxic compounds, such as mycotoxins, or, on the other side, of beneficial microorganisms that can limit pathogen growth. In this way, new strategies to protect both the agricultural market and consumer health could be formulated. To conduct this research a metagenomic analysis through metabarcoding sequencing using Oxford Nanopore Technology was performed. The metagenomic data was matched with metabolomic and agronomic data to find possible correlations among these that would drive us in choosing the best combination of cultivation practices, pedoclimatic conditions and varieties to enhance the presence of biocontrol agents. Regarding this, we performed co-culture assays to individuate potential biocontrol agents within the cultivable microorganisms isolated from durum wheat kernel which can be employed at field level for bio augmenting the capacity of durum wheat to challenge pathogens such as *Aspergillus flavus*, *Fusarium* from the FHB complex and *Tilletia spp*, *inter alia*

### **O1.3 - Dynamics of microbiota and *Fusarium* spp. responsible for Fusarium head blight and implications for biocontrol strategies**

Biocontrol, microbiomes and biostimulants

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Fusarium Head Blight (FHB) is a devastating cereal disease, primarily caused by *Fusarium* spp. (Fsp). While soil and crop residues are recognized as primary inoculum sources, *Fusarium* diversity and population dynamics in these substrates remain less studied compared to grains. Increasing attention has been directed toward understanding the interactions within *Fusarium* communities and with the phytomicrobiome, given their crucial role in disease onset and development. Understanding microbiome diversity and dynamics across Fsp-colonized substrates offers valuable insights into FHB epidemiology and ecology, and may lead to the discovery of potential biocontrol solutions or bioindicators of crop health. Six min-till wheat fields, following maize, were monitored for two years, with soil, maize residues, and wheat grains collected at four key stages, before metabarcoding (16S, ITS2, EF1 $\alpha$ ), and Fsp-specific qPCR. The results revealed that substrate type significantly influenced *Fusarium*, bacterial, and fungal community composition, explaining 36%, 44%, and 57% of the variance, respectively. Among the 31 identified *Fusarium* species, *F. graminearum* and *F. avenaceum* dominated both grains and residues, while *F. poae* was primarily found in grains. In 2021, under high FHB pressure, *F. graminearum* prevailed in grains, whereas *F. poae* became dominant in 2022 under low disease pressure, consistent with its weaker pathogenicity. SourceTracker analysis showed that residues were major source of wheat contamination in 2021, although their contribution was less pronounced in 2022. Furthermore, metabarcoding-based comparative analyses and co-occurrence networks revealed significant negative correlations between FHB disease parameters and several microbial genera, including *Pseudomonas*, *Curtobacterium*, *Epicoccum*, *Pyrenochaetopsis*, and *Pantoea* (also identified as a bioindicator of fusariotoxin-free grains). These taxa also exhibited antagonistic activity in screening assay on wheat grain-based medium and surface-disinfected wheat grains. Collectively, our findings provide insights into the interplay between *Fusarium* populations, microbial communities, and environmental factors, highlighting the potential for integrated strategies to manage FHB more effectively.

## O1.5 - Priming-inducing beta-amino acids trigger metabolomic reprogramming and increase resistance to grey mould in strawberry

Biocontrol, microbiomes and biostimulants

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Strawberry is highly susceptible to the necrotrophic fungus *Botrytis cinerea* causing significant yield losses and leading to frequent chemical fungicide applications, raising concerns about human health and environmental impact. As a result, there is growing interest in natural-derived compounds as priming agents to strengthen plant defences. *B. cinerea* infection can lead to complex plant metabolomic reprogramming, which could likely be modified by priming agents. However, few studies have taken a metabolomics approach to understand changes triggered by priming agents. Therefore, this study employed a LC-MS/MS-based untargeted metabolomics approach to elucidate the metabolic changes in mock- and *B. cinerea*-infected strawberry leaves upon repeated applications with  $\beta$ -aminobutyric-acid (BABA) and (R)- $\beta$ -homoserine (RBH). Four applications of treatments were conducted in strawberry cultivars ‘Durban’, ‘Rowena’, and ‘Soraya’. Firstly, our results demonstrated that, whereas no induced resistance was found in ‘Durban’, BABA and RBH significantly reduced *B. cinerea* lesion area in ‘Rowena’ (by 41% and 67%, respectively) and ‘Soraya’ (by 48% and 60%, respectively). A long-lasting effect was also found for both cultivars in BABA- and RBH-treated plants. Untargeted metabolomic analysis of mock- and *B. cinerea*-inoculated leaves, collected 24 hours post-infection, revealed that signatures seem to be cultivar-dependent. Interestingly, we found that RBH induced a stronger metabolomic reprogramming in ‘Rowena’ (upregulation of 375 metabolites, versus 136 in BABA), while BABA was more efficient in ‘Soraya’ (upregulation of 190 metabolites, versus 74 in RBH). Enrichment analysis revealed that metabolomic reprogramming induced by RBH is dependent on flavonoids, riboflavin and purine metabolism, stilbenoid, phenylpropanoid, amino acids, and monoterpene biosynthesis. On the contrary, priming by BABA seems to be linked mainly to flavonoid biosynthesis. Our results highlight metabolomics as a valuable tool for understanding priming of defences induced by BABA and RBH. Current analysis will putatively identify metabolites triggering enhanced resistance, which could aid in establishing innovative mitigation strategies.

## **O1.6 - The inner fungal microbiome of Scots pine branches influences necrosis formation by *Diplodia sapinea* in Southern Sweden**

Biocontrol, microbiomes and biostimulants

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Scots pine (*Pinus sylvestris* L.), a species of critical importance to forestry and the wood industry, is increasingly threatened by fungal pathogens, such as *Diplodia (Sphaeropsis) sapinea*. Like plants in general, Scots pines live together with fungal endophytes that may influence the defensive capabilities and potentially reduce the rate and extent of pathogen colonization.

Our research focuses on the fungal community within the inner branch tissues of young Scots pine trees and the potential roles these inhabitants play in combating fungal pathogens. We hypothesize that the fungal community in Scots pine trees influences the extent of necrotic lesions caused by *D. sapinea* and that a well-established endophytic fungal community results in smaller necrotic lesions.

In a field test conducted in Southern Sweden, we partially excluded fungal air-borne propagules from young branches of Scots pine saplings using mesh bags and compared their fungal endophytic microbiome with the control branches that we expected to possess a better-established inner community. We inoculated the branches with *D. sapinea* and measured the necrosis sizes after 21 days. We also checked the physiological/morphological changes of the branches with a multispectral 3D scanner. The fungal microbiome in the necrosis and healthy parts of branches with the presumably differently established communities were examined by ITS metabarcoding using Oxford nanopore sequencing.

We found no detectable difference between the control branches and those with partial inoculum exclusion based on multispectral and morphological measurements, indicating that the mesh bags did not cause major phenotypic changes in the branches. However, the control branches possessed a higher number of fungal OTUs and showed higher fungal diversity. The size of the necrosis caused by *D. sapinea* was significantly smaller on control branches with no inoculum exclusion, proving our hypothesis.

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## **O7.1 - Breeding for better biocontrol symbiosis of *Trichoderma afroharzianum* against *Aphanomyces cochlioides* in sugar beet.**

Biocontrol, microbiomes and biostimulants

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The oomycete genus *Aphanomyces* causes several plant diseases, particularly affecting crops in the *Amaranthaceae* family, such as beets, spinach, and quinoa. *Aphanomyces cochlioides* is harmful to *Beta vulgaris* subsp. *vulgaris* (sugar beet), causing two distinct diseases at different life stages, both of which lead to significant yield losses. Natural parasites of oomycetes, such as fungi from the *Trichoderma* genus, have been used to manage various crop diseases. However, the relationship between host plant disease resistance and the biocontrol and biostimulatory effects of beneficial microbes like members of the genus *Trichoderma* remains poorly understood. In this study, we evaluated the effectiveness of *Trichoderma afroharzianum* T22, a biocontrol agent and plant symbiont, against *A. cochlioides* in inbred sugar beet breeding lines. While *T. afroharzianum* successfully reduced disease symptoms in some lines, surprisingly, it worsened symptoms in others. Our findings suggest that these genotype-dependent responses extend to other biocontrol agents and that *T. afroharzianum*-related effects persist under semi-field conditions. Additionally, we observed significant variation in the growth-promoting effects of *T. afroharzianum* among elite sugar beet breeding lines that are independent of biocontrol traits. These results have led us to hypothesize that independent host plant genetic factors may be important for the success of microbial symbionts in biocontrol, biostimulation, and plant-microbe interactions. Future research will focus on identifying and incorporating these genetic factors into breeding programs to enhance crop resilience and improve symbiotic interactions.

## **O7.2 - Combining wheat and diverse perennial species: What is the impact on *Z. tritici* development and the wheat microbiome?**

Biocontrol, microbiomes and biostimulants

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Increasing biodiversity in agroecosystems, is desirable for maintaining ecosystem functions, including disease regulation. Strip-intercropping is one of the most effective crop diversification strategy used to limit disease development. However, it generally consists of the association of two crops, whereas several ecological studies recommend highest levels of diversity. In this context, a new cropping system called “Agroprairie” has been designed resulting from the association of alternating strips of wheat and diverse prairies (>10 plant species). The aim of this study is to explore the effect of this new cropping system on the regulation of *Zymoseptoria tritici* the causal agent of Septoria tritici blotch (STB), one of the most damaging leaf diseases of wheat.

To achieve this, four wheat cultivars with contrasting susceptibilities to STB, grown in “agroprairies” associated with four types of prairies (with different growth strategies and legume proportions) were investigated to explore the influence of intra- and interspecific diversity on the development of STB. In addition, a reduced number of plots with the most STB-susceptible cultivar was inoculated with *Z. tritici* to test the resistance of the associated wheat to inoculation. In parallel, to explore the role of microbiome in disease development, rhizosphere and roots DNA from associated wheat and prairies samples was extracted and sequenced using 16S rDNA. Four different STB incidence and severity notations were recorded between flowering and ripening stage.

The results showed that agroprairies with the highest biomass and proportion of legumes reduce the incidence of STB compared to wheat monoculture. However, the protection afforded to wheat by prairie strips is lost when *Z. tritici* is inoculated, which means that this protection may mainly come from a barrier effect. In parallel, preliminary results show that agroprairies modify the composition of the bacterial rhizospheric and root microbiome of wheat, and increase the abundance of known PGPRs.

### **O7.3 - Combining *Trichoderma* and *Clonostachys* to improve fusarium head blight control on wheat: from lab to greenhouse scale.**

Biocontrol, microbiomes and biostimulants

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*Fusarium graminearum* (*Fg*) is the primary causal agent of Fusarium head blight (FHB) in Central and North Europe, producing harmful mycotoxins. Biocontrol agents (BCAs) offer a sustainable alternative to controlling this pathogen. *Trichoderma gamsii* T6085 (*Tg*) and *Clonostachys rosea* IK726 (*Cr*) are two BCAs known to control FHB. This study aims to improve efficacy and stability by combining them.

We assessed the compatibility of *Tg* and *Cr* *in vitro* and their effectiveness *in planta* during increasing environmental stress (growth chamber vs. greenhouse). Compatibility assays showed no growth reduction associated with volatile or diffusible compounds when the strains were co-inoculated as mycelia. Nevertheless, *Tg* spore germination was completely inhibited in *Cr* filtered spent media. Similarly, the co-inoculation of spore suspensions, resulted in a drastic reduction in *Tg* biomass production. Nevertheless, in the growth chamber experiment treatments induced defence related (DR) genes being upregulated by the co-inoculum at 96 hpi in the absence of the pathogen. Similarly, some DR genes were upregulated by the co-inoculum 24 h after the *Fg* infection (48 hpi with the BCAs). Furthermore, the co-inoculation strategy reduced the percentage of symptomatic spikes by 93%.

To improve the co-inoculation strategy, we analysed the germination process of *Tg* and *Cr* when co-inoculated. Fluorescent microscopy showed *Cr* advancing to the mycelial growth stage by 12 hpi, while *Tg* spores were still in the early germination (polar) stage. To enhance effectiveness at a greenhouse scale, we increased spore suspension concentration and pre-hydrated *Tg* spores for 12 hours before application.

Greenhouse trials confirmed that the co-inoculation strategy improved *Fg* management and enhanced treatment stability in terms of disease index, *Fg* biomass as well as grain yield and quality. These findings, coupled with further exploration of co-inoculum spore germination process, provide a foundation for developing more robust and scalable field treatments.

## O7.4 - Host plant switching and viral infections reshape the microbiome of the aphid vector *Aphis gossypii*

Biocontrol, microbiomes and biostimulants

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Insect-associated microbiomes constitute a crucial component of the physiological fitness of insects, including aspects such as reproduction, development, immunity, and behavior as well as pathogen transmission. While there is growing interest in understanding how microbiomes influence insect pest populations, the relative effects of the transition between host plants and viral infections remain unclear. In this study, we examined the bacterial community composition of the cotton-melon aphid, *Aphis gossypii* Glover, as a global agricultural insect pest that can damage cultivated plants directly via sap-sucking and indirectly by transmitting viral diseases, in response to cucurbit plant transitions and viral infections by persistent (Cucurbit aphid-borne yellows virus, CABYV) and non-persistent (Watermelon mosaic virus, WMV) plant viruses in single and mixed infections. Our 16S rRNA gene sequencing analysis revealed that the transition between plants significantly influenced aphid microbiome composition, with greater bacterial diversity in aphids feeding on melon than those feeding on cucumber. This disruption in the microbiome structure was mainly due to shifts in the relative abundance of facultative symbionts, such as *Arsenophonus* and *Sphingomonas*. Furthermore, viral infections significantly reduced microbiome diversity compared to that in healthy plants, with the lowest diversity observed in aphids feeding on mixed-infection plants with CABYV and WMV. A notable negative correlation between *Buchnera* and *Arsenophonus* abundance was identified in these viruliferous aphids, suggesting that viral infections may modulate endosymbiont interactions. This study underlines shifts in the aphid microbiome, which could provide insights for future pest-integrated management strategies, thus emphasizing the need for further investigation of microbial resource-based solutions to sustainably control aphid pests and associated viral diseases in agriculture.

## **O7.5 - Polycyclic tetramate macrolactams from *Lysobacter capsici* AZ78 boost defence mechanisms against *Plasmopara viticola***

Biocontrol, microbiomes and biostimulants

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*Plasmopara viticola*, the causal agent of grapevine downy mildew, is a significant threat to winemakers globally as it causes relevant annual losses in both the quality and quantity of grape production. Traditional control strategies mainly rely on copper-based and synthetic fungicides and have negative environmental impacts. Therefore, there is a pressing need to develop new alternatives to promote more sustainable grapevine production. In this regard, *Lysobacter capsici* AZ78 is emerging as a new and potential biocontrol agent which is seen to be effective against broad range of organisms.

In this study, we investigated the mode of action of *Lysobacter capsici* AZ78 (AZ78), an effective biocontrol agent of *P. viticola*. To do that, we evaluated the ability of live and heat-treated AZ78 cells to control *P. viticola* *in vitro* and *in planta*. Results showed that heat-treated AZ78 cells had the same efficacy as live cells, reducing disease severity by >95%, comparable to commercially available copper-based fungicides. Heat-treated and live AZ78 cells stimulated callose deposition and reactive oxygen production (ROS) in grapevine leaf disks, similar to acibenzolar-S-methyl and laminarin. This further underscores their role in plant defense activation, in addition to direct plant protection. Subsequently, polycyclic tetramate macrolactams (PTM) were extracted from AZ78 cells, and heat-stable antifungal factor (HSAF)/ dihydromaltophilin (DMP) and maltophilin (MP) were identified. Applying a mixture of DMP and MP (as low as 2.5 mg L<sup>-1</sup>) on grapevine leaf disks drastically reduced *P. viticola* infection. The mixture of these PTMs also stimulated callose deposition and ROS production in grapevine leaf disks.

Overall, this comprehensive approach sheds light on how AZ78 effectively controls *P. viticola*, paving the way for its possible development as a commercial biocontrol agent.

## O7.6 - When the pathogen becomes the prey: responses of *Phytophthora infestans* and *Botrytis cinerea* to mycoparasitism

Biocontrol, microbiomes and biostimulants

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The ability of an organism to sense potential microbial threats and mount an immune response is one of the hallmarks of all life on earth. Innate immunity has been well studied in humans, animals and plants however, it remains underexplored in other Eukaryotic lineages. Plant pathogens share their ecological niche with a multitude of other microorganisms that compose the plant holobiont including mycoparasite species that represent an important resource to control plant diseases. However, the mechanisms involved in these interactions are not well studied. This study examines how two well-known phytopathogens with differing life histories respond to attack by the mycoparasitic oomycetes *Pythium oligandrum* and *Pythium periplocum*. *Phytophthora infestans* is a hemibiotrophic oomycete that causes late blight in potato and tomato and infection mechanisms via the secretion of effectors, molecules that modulate host plant defences and facilitate plant infection have been extensively characterised. *Botrytis cinerea* is a fungal necrotrophic phytopathogen characterized by its broad host range and the secretion of an arsenal of toxins, cell wall degrading enzymes and effectors that induce plant cell death. The molecular components underlying the responses of these two Eukaryotic microbes to mycoparasitism by the *Pythium* species were investigated *in vitro* using RNAseq. Taking a biologically oriented gene mining approach, we mapped the processes involved in the responses to antagonism in the two prey species, covering recognition of the antagonist through putative NLRs, RLKs and GPCRs, signal transduction, gene regulation, detoxification and secreted defence or counter-attack molecules, such as specialised metabolites and effectors. Our data suggest effectors may have more diverse functions than previously thought, including roles in generalised stress responses or microbe-microbe interactions. Taken together, our results shed light on a poorly understood part of the biology of filamentous Eukaryotic microbes and represent a valuable resource to understand their responses within the plant holobiont.

## **P25 - Biological control of *Colletotrichum acutatum* with endophytic bacteria from olive drupes**

Biocontrol, microbiomes and biostimulants

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Olive anthracnose (OA), caused by fungi of the *Colletotrichum* species, poses a significant threat to olive cultivation worldwide, leading to severe yield losses and degradation of olive oil quality. Among the *Colletotrichum* species, strains belonging to the *C. acutatum* species complex are dominant in Greece. The continuing withdrawal of active substances, combined with the adverse effects of chemical fungicides on the environment, underscores the need to explore more sustainable ways of managing OA. This study aimed to explore potential biological control agents and assess their impact on the severity of OA. As part of this objective, 210 endophytic bacterial isolates were tested for their antagonistic activity against *C. acutatum* through multiple and dual culture assays. Several isolates showed satisfactory inhibition of mycelial growth and were further tested in situ for their effect on disease severity and conidial production on detached olive drupes (*cv.* Kalamon). Statistically significant differences were observed in all bacterial treatments compared to the control, and the isolates identified through 16S rRNA gene sequencing. The four most effective isolates were further evaluated in artificially inoculated olive trees at the fruit ripening stage, under field conditions, for two consecutive years. Notably, isolate II8 (*Serratia* spp.) reduced disease severity by 89.31%, while the other three isolates showed promising results. Field applications were also conducted during the flowering stage in olive cultivars *cv.* Kalamon and *cv.* Amfissis, where the bacterial isolates significantly reduced anthracnose severity in young fruits, with reductions up to 50% in *cv.* Kalamon and 36.8% in *cv.* Amfissis.

### **P35 - Utilizing *Lumbricus terrestris* to mitigate clubroot in oilseed rape farming**

Biocontrol, microbiomes and biostimulants

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Clubroot disease, caused by *Plasmodiophora brassicae*, poses a major economic threat to oilseed rape cultivation. Various species of earthworms have been explored as potential biological agents for reducing plant disease severity and improving overall agricultural productivity. This study examined the role of the earthworm *Lumbricus terrestris* in suppressing clubroot development under controlled greenhouse conditions using a food choice arena. The experimental design employed plastic pots and a factorial approach, with eight treatments involving soil inoculated or not with *P. brassicae*, with or without the addition of *L. terrestris*, and feeding periods of either 7 or 35 days. Clubroot disease development was assessed using a bioassay method. The results demonstrated that *L. terrestris* effectively suppressed clubroot disease in a susceptible oilseed rape cultivar, with the suppression being more pronounced over longer feeding periods. The highest incidence of clubroot (70.4%) occurred with a 7-day feeding period, while a 35-day feeding period resulted in a significantly lower incidence (27.5%). The effect of earthworm activity was particularly evident after 35 days, where the incidence of *P. brassicae* infestation was five times higher in pots without earthworms compared to those with earthworms. Additionally, the severity index of clubroot was significantly higher in pots without earthworms. In conclusion, the activity of earthworms plays a significant role in the natural self-regulation of agricultural soils, making them a promising candidate for biological control in the management of clubroot disease in practical farming scenarios.

## **P68 - Abiotic stress & needle pathogen infection: Which spruce families are struggling the most?**

Biocontrol, microbiomes and biostimulants

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Climate change is accelerating stress on forest ecosystems, increasing trees' vulnerability to pathogens and pests. To ensure the resilience of forests, breeding programs should focus on developing trees that are tolerant to abiotic stress and biotic damages. In Norway spruce (*Picea abies*) breeding programs, needle pathogen susceptibility is often overlooked, despite evidence that needle endophyte communities may include opportunistic pathogens<sup>1,2</sup>. The composition of the fungal community in needles is also influenced by host genotypes

and reflects the aerial fungal composition at the time of bud flush, suggesting this stage is when infections occur.

Our study explores the relationship between timing of budflush, fungal colonization and tree vitality at the family level. Hypothesizing that the inoculum available in the air at the time of budflush determine the fungal community in needles and the risk of pathogen infections. We also hypothesize that trees experiencing an abiotic stress like drought will have a larger pathogen load in the communities.

To test these hypotheses, 200 clones from 30 families were selected at two field-sites, one drought-prone and the one not. Tree and needle vitality were assessed, bud samples at different developmental stages and spore traps were collected weekly during two seasons. Air and needle communities were analysed using ITS2 metabarcoding and PacBio sequencing.

Preliminary results shows that the clones and families differ in vitality across sites and response differently to drought. These findings will be integrated with the fungal community composition data to analyse interactions between environmental stress, fungal colonization and host genetics.

## **P69 - Applications of yeast exudates improved strawberry antioxidant responses leading to lower incidence of fungal diseases**

Biocontrol, microbiomes and biostimulants

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Strawberries are highly susceptible to fungal diseases, leading to intensive applications of chemical fungicides with downsides on the environment and human health. Biological control agents are being increasingly explored as a sustainable strategy for controlling these diseases. However, more knowledge is needed to better understand their efficacy and their impact on plant fitness. Firstly, we have explored the effects of three endophytic yeasts' formulations (living cells, exudates and extracts) on *Botrytis cinerea* control in strawberry plants (*Fragaria x ananassa*, cv. 'San Andreas'). These formulations were applied every two weeks throughout strawberry cultivation period. It was found that the exudates were the most efficient treatment (leading to a 30-50% reduction of lesion area caused by *B. cinerea*, when compared to non-treated plants). Moreover, the photosynthetic rate was not significantly impacted by the multiple exudates' applications. In a second experiment, the two most efficient exudates from endophytic yeast species were leaf sprayed (independently or combined) in three cultivars ('Felicity', 'Fortuna' and 'Leticia'), following the procedure of the first experiment. Analysis of fungal diseases incidence during plant growth revealed that the application of yeast exudates, particularly when combined, reduced the powdery mildew incidence on leaves in about 4.9% and 19.7% in the cv. 'Felicity' and 'Fortuna', respectively, when compared with non-treated plants. Moreover, the results of an *in vivo* infection assay, using detached leaves from plants exposed to four applications with exudates, showed that the lesion area caused by *B. cinerea* was significantly reduced (51.6%) in the condition of combined exudates, when compared with leaves from non-sprayed plants. This differential response can be partly explained by the improved antioxidant response found in treated plants. This was the case of the activity of superoxide dismutase enzyme at leaf level (24h after the fourth application of exudates), which increased by 65.4%, as compared with control.

## **P70 - Almond microbiome of plants displaying symptoms of dieback and decline unfold the importance of soilborne pathogens**

Biocontrol, microbiomes and biostimulants

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Almond decline and dieback represent emerging threats to newly established intensive orchards in the Mediterranean region. Associated to this disease complex are the symptoms of trunk internal necrosis, cankers, and external gummosis that are responsible for significant production losses. This work represents the first attempt to explore the microbiome composition of almond trees using high-throughput DNA metabarcoding (16S and internal transcribed spacer ribosomal RNA). Most of the potential pathogens identified are primarily linked to root and crown rots, highlighting the crucial impact of these microorganisms. Specifically, *Neocosmospora rubicola*, a previous member of the *Fusarium solani* complex, *Dactylonectria estremocensis* and *Plectosphaerella niemeijerorum* were identified in the core microbiome and are suggested to participate in co-infections where their joined presence may escalate symptom development. Furthermore, *Fusarium* spp. was the third most abundant OTU, agreeing with previous studies that point it as an important player in the development of trunk diseases. Other highly abundant plant pathogens that are suggested to have similar effects include *Stemphylium majusculum*, *Cristulariella depraedans* and *Mycosphaerella tassiana*. These are known agents responsible for foliar diseases in many herbaceous and woody species that reduce overall plant vigor and may predispose plants to other opportunistic trunk pathogens that lead to dieback and additional decline. Most of the potential pathogens identified were also detected in the soil samples, suggesting that these or the nursery substrates might have been the source of inoculum. This study is crucial for the development of new approaches to manage this disease complex, and subsequent strategies will require a focus on soil health and pathogen interactions.

**P71 - Biocontrol of potato scab: Potential of antagonistic *Streptomyces* in managing *Streptomyces scabiei***

Biocontrol, microbiomes and biostimulants

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Potato scab, caused by *Streptomyces* spp., poses a significant global challenge, affecting both quality and yield. In Brazil, the disease has led to considerable economic losses. This study addresses the urgent need for effective and sustainable control methods, focusing on the biocontrol potential of non-pathogenic and antagonistic *Streptomyces* strains, specifically *Streptomyces* sp. IBSBF 3377 and *Streptomyces* sp. IBSBF 3379. The experiments included in vitro tests, greenhouse assays, and field evaluations. In vitro results demonstrated that both *Streptomyces* strains exhibited notable antagonistic activity, inhibiting the growth of six phytopathogenic *Streptomyces* species (*S. scabiei*, *S. europaeiscabiei*, *S. caviscabies*, *S. turgidiscabiei*, *S. reticuliscabiei*, and *S. brasiliscabiei*). Greenhouse and field trials were conducted against *S. scabiei*, the most commonly associated species with potato scab problems, to ensure relevance and practical application. In greenhouse trials, *Streptomyces* sp. IBSBF 3377 significantly reduced disease incidence by 75.5% and severity by 58.8%, while *Streptomyces* sp. IBSBF 3379 reduced incidence by 64.9% and severity by 62.6% compared to the positive control. Field trials confirmed these findings, with *Streptomyces* sp. IBSBF 3377 reducing disease incidence by 67.7% and severity by 61.3%, and *Streptomyces* sp. IBSBF 3379 reducing incidence by 67.9% and severity by 54.6%. This study underscores the importance of exploring interactions between organisms of the same genus for disease control, as they compete for the same ecological niche. Understanding the dynamics between antagonistic and pathogenic strains is crucial to uncovering novel biocontrol mechanisms. Future research will delve deeper into these interactions, investigating specific mechanisms such as antimicrobial production, resource competition, and induced plant resistance to optimize the use of these biocontrol agents.

## **P72 - Boosting the performance of *Pythium oligandrum* for biocontrol and biostimulation in potato**

Biocontrol, microbiomes and biostimulants

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Potato (*Solanum tuberosum*) ranks as the world's third most important food crop, following rice and wheat, with an annual production of 330 million tons, making it crucial for food security globally. However, pathogens like *Phytophthora infestans*, the causal agent of late blight, and *Alternaria solani* that causes early blight, pose significant threats to potato cultivation globally. These pathogens cause substantial annual losses and have developed highly aggressive strains that undermine current control strategies. The situation is exacerbated by climate change, which is expected to further reduce yields. The oomycete *Pythium oligandrum* is a mycoparasite with potential as a biocontrol agent and biostimulator in potato. Although commercial preparations of *P. oligandrum* are approved for use in potato and other crops within the EU, adoption of *P. oligandrum* remains limited due to challenges such as inconsistent standardization and limited shelf life, which deter farmers from opting for it over chemical alternatives. Potato crops are typically cultivated in sandy soils; however, much of the research on *P. oligandrum* has been conducted either *in vitro*, in compost or in nutrient-rich soils, leading to uncertainties when applied to field conditions. In this study, we examine the differences in root colonization by *P. oligandrum* across soils with varying sand concentrations. We are also investigating the effectiveness of *P. oligandrum* biocontrol when it is faced with both *P. infestans* and *A. solani*, as may be the case within the potato cropping system. Finally, we evaluate the growth-promoting properties of *P. oligandrum* across 96 potato cultivars and investigate correlations with specific genetic markers, as a prelude to breeding for potato cultivars that better host with biocontrol agent. Our findings aim to improve our understanding of *P. oligandrum* efficacy under crop production systems, with the goal being to enhance its effectiveness as a biocontrol and growth-promoting agent.

### **P73 - Collefree vector, a new tool for olive protection against anthracnose**

Biocontrol, microbiomes and biostimulants

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Olive anthracnose, the most significant and widespread disease affecting olive fruits, is caused by several fungal species, particularly from *Colletotrichum acutatum* complex. This disease has a large impact in Portugal, Spain, Italy and other Mediterranean countries as well as in Australia, and South Africa.

This disease is normally controlled through a combination of appropriate cultural practices and preventive applications of copper fungicides. However, when conditions are particularly favorable for anthracnose development, fungicide applications become insufficient or ineffective. In addition, European Community directives restrict the use of copper-based products, so there is a need to find alternatives to combat the disease.

The development of a modified virus-based vector carrying an antimicrobial peptide for plant protection against *Colletotrichum* complexes, will be a great innovative method to counteract the use of chemical products for the control of anthracnose, leading to a more sustainable olive production.

For that purpose, we have been developing the Collefree vector, a modified non-pathogenic version of Olive mild mosaic virus (OMMV) carrying the antimicrobial peptide S-carboxyamidomethylated Anafp from the fungi *Aspergillus niger*. This vector will replicate and accumulate in olive plants without being transmitted and not causing viral disease symptoms. The Collefree vector will also have the advantage of being a low cost and green alternative to chemical fungicides, with effective and quick results.

## **P74 - Environmentally friendly control of potato diseases with biofungicides**

Biocontrol, microbiomes and biostimulants

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Late blight, caused by *Phytophthora infestans*, and early blight, caused by *Alternaria spp.*, result in significant yield losses and are thus heavily sprayed against by growers. However, the excessive use of fungicides has negative impacts on the environment and human health. Biofungicides offer an eco-friendly alternative for controlling these diseases in potatoes. In this study, several biofungicides, including plant extracts, microorganisms, and parts of macroorganisms were explored. The results showed effective inhibition of both pathogens by biofungicides such as *Trichoderma asperellum*, *T. atroviride*, and *Bacillus amyloliquefaciens* for both late blight and early blight under in vitro conditions. Biofungicides such as chitosan and orange oil also proved very effective in suppressing *Alternaria spp.* Overall, our study has identified potent biofungicides for controlling late blight and early blight in potatoes.

## **P75 - Comparison of the endophytic mycobiome from grains of selected barley varieties grown in different regions of Poland.**

Biocontrol, microbiomes and biostimulants

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Barley (*Hordeum vulgare* L.) is among the top cereals cultivated worldwide. The main barley-producing countries are Russia, Canada, Australia, the European Union countries, and North America. In Poland annually, the fields of this cereal cover an area of about 760 thousand hectares. Spring barley is grown mainly for fodder and brewing purposes.

Barley is inhabited by the diversity of microorganisms whose mutual interactions influence its health, growth and development, grain yield and quality. Endophytic fungi are a key component of the microbial communities that inhabit the internal tissues of plants without causing visible symptoms of disease and play an important role in plant health and growth promotion.

The aim of the presented study was to compare the endophytic mycobiome of barley grains of three varieties ‘Aristelle’, ‘Flamenco’ and ‘Tilmor’ cultivated in different location in Poland. The research was carried out using traditional culture method and high-throughput Next Generation Sequencing (NGS) technology. The conducted analyses revealed endophytic colonization of barley grain tissues by fungi of the genera *Alternaria*, *Fusarium*, *Bipolaris*, *Epicoccum* and numerous cultures of non-sporulating species. The species composition, especially within the genus *Fusarium*, differed slightly depending on the barley variety studied.

The use of the culture method combined with Next Generation Sequencing allow for more precise and reliable results to be obtained.

## **P76 - Composition of plant pathogenic fungi in Pannonian forests are shaped by topography-driven changes in abiotic factors**

Biocontrol, microbiomes and biostimulants

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Plant pathogenic fungi are diverse and contribute maintaining high plant diversity in forest ecosystems. We carried out DNA metabarcoding of fungi from soil samples taken at 62 sites in the Bükk Mountains in northern Hungary. The selected sampling sites represent 11 characteristic Pannonian forest types distributed along elevation (i.e. temperature), soil moisture, and pH gradients. We compared genotypic richness and community composition of plant pathogenic fungi among forest types and explored relationships among environmental variables and fungal alpha and beta diversity. The soil DNA sequence data showed correlations between plant pathogenic fungal community composition and environmental variables, and, consequently among zonal, topographic or edaphic forest types. For example, forest type as a categorical variable was responsible for nearly 30% of the compositional differences between samples. Of the measured continuous variables, soil pH showed the highest correlation with community composition, explaining about 9% of the compositional variance, while elevation was responsible for 5%. The majority of the compositional differences (65%) remained unexplained, indicating that both niche-based and random (stochastic) processes influence the composition of fungal communities. Our work provides unprecedented insights into the diversity, landscape-level distribution, and habitat preferences of plant pathogenic fungi in the Pannonian forests of Northern Hungary.

## **P77 - Forestry treatments alter the plant pathogenic mycobiome in canopy and understory plants in an oak forest in Hungary**

Biocontrol, microbiomes and biostimulants

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The effects of different forest management approaches on vegetation have been extensively studied. However, little is known about how forest management affects plant-associated fungal communities. As part of the long-term Pilis Forestry Systems Experiment in northern Hungary, we generated DNA metabarcoding data from leaves of three plant species that are common throughout the study area and represent different growth forms and three major clades of plants: *Hypnum cupressiforme*, *Carex pilosa*, and *Quercus petraea*. Our goal was to characterize plant pathogenic fungal communities in each host species and to compare their responses to forestry treatments. We sampled six replicate plots of clear-cutting (CC), gap-cutting (G), preparation cutting (P), retention tree group (R), and control (C). DNA sequences were identified and assigned to functional categories using the UNITE and FungalTraits databases, respectively. We found 810 plant pathogenic fungal genotypes, of which 261 occurred in mosses, 551 in sedges, and 410 in oak leaves. We observed compositional differences of plant pathogenic fungi among plant species, particularly between the moss and the vascular plants. Several fungal genera preferred a specific host and we identified several indicator species for each host. Treatments accounted for 21.55% of fungal compositional variance in mosses, 42.98% in sedges and 34.75% in oak leaves. Compositional changes appeared to be proportional to the disturbance severity, with clear-cutting resulting in the greatest turnover of fungal species. Changes in plant pathogenic fungal communities correlated significantly with relative humidity, temperature measured at soil surface and at heights of 15 cm and 130 cm, and with vapor pressure deficit, indicating that a substantial effect of treatments on leaf fungal communities are mediated through changes in the microclimate. The observed compositional dynamics in leaf-associated fungal communities provide valuable insights into how different forest management activities shape plant pathogenic fungi, with possible implications for forest health.

## **P78 - Cyanobacterial bioextracts as biocontrol agents against plant pathogens**

Biocontrol, microbiomes and biostimulants

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The overuse of synthetic pesticides in agriculture poses a significant risk to human health, environmental sustainability and microbial ecosystems. Therefore, there is growing interest in sustainable alternatives, especially biopesticides derived from natural sources. Cyanobacteria, known for their ability to produce biologically active antimicrobial compounds, represent a promising option for plant protection. In this study, cyanobacteria from Lithuanian aquatic ecosystems are evaluated as potential biological control agents against plant pathogens, including bacteria, fungi and oomycetes.

The research includes biomass screening and the development of a methodology to evaluate the antagonistic activity of cyanobacterial extracts. Aqueous and methanolic extracts will be tested at different concentrations to determine their effect on 15 species of plant pathogens. Initial results indicate that the methanolic extract (44.45 g/L) has a strong inhibitory effect on fungal pathogens such as *Diaporthe* spp., *Fusarium* spp. and *Phytophthora* spp. Furthermore, a pronounced suppressive effect on the bacterial pathogen *Xanthomonas* spp. was observed. However, the aqueous extracts did not inhibit the growth of the pathogen, although slight morphological changes were observed in the cultures.

These results emphasise the potential of cyanobacterial bioextracts as biological control agents contributing to disease management and sustainable agriculture. Future research will further elucidate the mechanisms underlying their antimicrobial properties and pave the way for their application as environmentally friendly biopesticides.

## **P79 - Efficacy of phytosanitary products based on copper in the form of gluconate and elemental sulfur against plant pathogens**

Biocontrol, microbiomes and biostimulants

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Wheat is one of the most important crops in maintaining the world's food security. A factor that can significantly reduce the quantity and quality of winter wheat yield are fungal diseases. One of the basic methods of protecting wheat plants against pathogens is chemical treatment. Nowadays it is necessary to partially replace synthetic fungicides with alternative preparations. The use of hazardous chemical pesticides has severe effects on soil microbes and animal health. It is therefore worth looking for plant protection inputs of mineral origin, such as copper and sulphur. The experiment was done *in vitro* and in the field. The tested substances (copper gluconate and elemental sulfur) were added to sterile PDA medium to obtain the appropriate concentration of the active substance corresponding to the field. Discs (6mm) of fungal species (*Fusarium solani*, *F. commune*, *F. cerealis*, *F. avenaceum*, *F. culmorum*, *Alternaria alternata*) were placed in the center of Petri dish. The plates (5 replicates per species) were incubated at 20°C day/ 14°C night at 12 h/12 h photoperiod and the diameter of colony was measured daily. The field experiment was carried out in Urbanowice (Opole region) in 2022-2024 on Euforia variety. Experiment was done in 3 replicates (15 m<sup>2</sup> each plot). Disease severity was rated by determining the percentage of plants infected in each plot. All preparations containing copper gluconate and elemental sulfur contributed to a statistically significant reduction in the growth of mycelium of pathogens, even to the complete inhibition of mycelium development. On the field statistically significant decrease in the percentage of infection was observed even in comparison to routine used chemical treatment. This research brings promising findings on the fungistatic properties of sulfur and copper, opening up new perspectives in plant protection against fungal diseases.

## **P80 - Evaluation of potential biocontrol agents for mitigation of aflatoxin contamination in pistachios**

Biocontrol, microbiomes and biostimulants

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Pistachios that constitute one of the most important nuts in the world, are particularly vulnerable to infections by fungi that produce aflatoxins. Aflatoxins form one of the major groups of mycotoxins showing teratogenic and carcinogenic effects on humans and animals. Among the different numerous methods that have been evaluated for mitigating aflatoxins in pistachios, biological methods exhibit the highest efficacy. The aim of the study was to evaluate the capability of two yeast strains of *Aureobasidium pullulans* and *Meyerozyma caribbica*, one *Pantoea agglomerans* bacterial strain and one non-toxigenic strain of *Aspergillus flavus* to strongly reduce aflatoxin contamination in pistachios orchards in Greece. These microorganisms were isolated epiphytically from pistachios. Previous studies have demonstrated that these particular isolates effectively suppress aflatoxin biosynthesis in pistachio fields on the island of Aegina. For the achievement of the stated objective, the previously mentioned strains were applied to two distinct pistachio orchards: one commercial orchard located on Aegina Island and one experimental orchard at the Agricultural University of Athens. Analysis of the pistachio kernels revealed that aflatoxin formation was significantly inhibited by all four strains, resulting in reductions ranging from 86% to 99%. These results are consistent with findings from previous experiments and further support the development of biocontrol products for the effective management of aflatoxins in pistachios.

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## **P81 - Fungal communities in Swedish elms: Implications for Dutch elm disease resistance**

Biocontrol, microbiomes and biostimulants

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The native elm (*Ulmus* spp.) populations in Sweden, as well as larger parts of the elms' distribution area, are strongly threatened by the Dutch Elm Disease (DED). This wilt disease is caused by aggressive invasive fungi in the genus *Ophiostoma*. Although the disease is rapidly spreading, some elms show tolerance or even resistance to the disease. Previous research has shown that certain microbial communities have a positive effect on their host, for example, they may support disease resistance by stimulating tree resistance mechanisms. If similar communities could be found in the Swedish elms that show tolerance or resistance to DED, it could be possible to find new tools to push forward the breeding and support conservation of our native elms. The goal of our studies is to provide new knowledge about elm symbiotic microbial communities and their possible effects on the resistance of elms to DED. Using a crowdsourcing approach, we searched for elms that show tolerance to DED. Sampling efforts and inventories targeted *U. glabra* population in Southern Sweden where the disease has been active for decades. The endophyte communities were studied using metabarcoding and isolation technique. About 80 isolates were recovered from visually healthy trees, including several *Fusarium* species. The results of biotests showing the activity of selected isolates against DED pathogens are discussed.

Keywords: Dutch elm disease, *Ulmus* spp., *Ophiostoma*, microbial communities, fungi.

## **P82 - Impact of AgNPs obtained via plant extracts-mediated green synthesis on quarantine and regulated phytopathogens**

Biocontrol, microbiomes and biostimulants

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Quarantine and regulated non-quarantine phytopathogens, such as *Ralstonia solanacearum*, *Clavibacter sepedonicus*, *Xanthomonas campestris* and *Erwinia amylovora*, cause significant losses in agriculture and spread easily due to causing difficult to detect latent infections.

The aim of this study was to investigate the antimicrobial potential of silver nanoparticles (AgNPs) obtained via green synthesis driven by reduction of silver nitrate by aqueous extracts from common herbs, here *Artemisia absinthium*, *Artemisia vulgaris*, *Echium vulgare*, *Glechoma hederacea*, *Solidago canadensis* and *Urtica dioica*.

The synthesis of AgNPs was conducted at 100 °C in the presence of polyvinylpyrrolidone as a stabilizer. The exact size of AgNPs was determined by transmission electron microscopy to equal for *A. absinthium* – 26±10 nm, *A. vulgaris* – 30±8 nm, *E. vulgare* – 40±15 nm, *G. hederacea* – 22±8 nm, *S. canadensis* – 23±8 nm, *U. dioica* – 26±9 nm. The bactericidal properties of AgNPs were revealed by determination of minimal inhibitory concentrations (MIC) and minimal bactericidal concentrations (MBC). AgNPs of the highest antimicrobial properties were synthesized using *S. canadensis* extract and showed the lowest MIC of 1.3 ppm against *C. sepedonicus* and the highest MIC of 3.7 ppm towards *X. campestris*. The MIC and MBC values obtained in this study were lower than these reported in the literature; e.g. Gulamnabi et al. (2020) described a MIC of 12.5 ppm against *R. solanacearum*, while Johanna et al. (2024) mentioned a MIC of 50 ppm towards *E. amylovora*.

Furthermore, we tested the effects of herein revealed AgNPs on seedlings growth and observed 85.8% increase in median shoot length of *Solanum lycopersicum* exposed to *S. canadensis*-based AgNPs.

During further research, we hope to increase the efficiency of AgNPs synthesis, and study their ecotoxicity against naturally occurring soil organisms.

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### **P83 - Impact of soil microbiota and nitrogen fertilization on alder susceptibility to *Phytophthora x alni***

Biocontrol, microbiomes and biostimulants

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Alder (*Alnus glutinosa*) is a key species for European riparian forests due to its importance in maintaining soil fertility, riverbank stability, and ecosystem biodiversity. However, it is experiencing decline and mortality, primarily linked to the invasive pathogen *Phytophthora x alni*. The effect of biotic interactions on the relationship between alder and *P. x alni* remains unknown. The soil microbial community can influence both plants (e.g., mutualisms, pathogenicity) and pathogens (e.g., facilitation, antagonism). Symbiotic *Frankia* bacteria may also affect the plant-pathogen interaction through their indirect impact on plant health or the production of antimicrobial metabolites, but their nodulation is limited by high nitrogen concentrations in the soil.

Through a greenhouse experiment with alder seedlings, we evaluated plant responses to inoculation with *P. x alni* and the combined effects of soil communities (sterilized soil, soil with natural microbiota) and the inhibition of nodule formation through nitrogen fertilization. This study aims to clarify the impact of natural soil microbiota on alder decline and the effect of nitrogen fertilization on this interaction.

Natural microbiota and fertilization promoted larger seedlings through increased leaf area and chlorophyll content, although their combined effect reversed these benefits. Inoculation had limited impacts on ecophysiological measurements but led to a reduction in the seedling final weight. Inoculated plants also showed higher root necrosis and lower root/shoot ratio, which may hinder water and nutrient uptake, potentially limiting overall plant growth and development. Despite the benefits of natural microbiota and nitrogen fertilization on plant photosynthesis, they did not fully mitigate the impacts of *P. x alni* on the root system and overall plant growth.

## **P84 - Inhibitory effects of *Asparagus officinalis* L. Extracts on the growth of phytopathogenic *Fusarium* spp.**

Biocontrol, microbiomes and biostimulants

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*Fusarium* species are among the most widespread phytopathogens, causing significant agricultural losses and reduced crop yields globally. Under favorable environmental conditions, these pathogens produce toxic secondary metabolites, known as mycotoxins, which pose serious risks to public health and food safety. However, the growing resistance of *Fusarium* spp. to synthetic fungicides has necessitated the exploration of alternative, eco-friendly antifungal agents. This study evaluated the antifungal potential of asparagus extracts against the mycelial growth and mycotoxin biosynthesis of *Fusarium oxysporum* and *F. proliferatum* isolates under controlled *in vitro* conditions. Leafy asparagus extracts were obtained by immersing 15-week-old plant material in methanol (1:1 ratio), followed by solvent removal via a vacuum rotary evaporator. *In vitro* antifungal assays were conducted by growing a cylindrical piece of mycelium on potato dextrose agar (PDA) media supplemented with 10 % and 20 % extract fractions, with untreated media serving as the control. Mycelial growth inhibition was recorded on days 3, 6, 9, and 10 post-inoculation. Mycotoxin biosynthesis was analyzed using Ultra-High Performance Liquid Chromatography coupled with tandem mass spectrometry (UHPLC-MS/MS). The results demonstrated that all asparagus extract concentrations inhibited *Fusarium* growth compared to the control. The highest inhibitory effect was observed with the 20 % extract concentration. Furthermore, mycotoxin analysis revealed that *F. proliferatum* produced fumonisins B1–B3, beauvericin, and moniliformin, while *F. oxysporum* produced only moniliformin. Additionally, the mycotoxin content was highest in untreated samples, while asparagus extracts significantly reduced their levels. Moreover, the reduction in mycotoxin content was not concentration-dependent, and the 20 % extract did not affect moniliformin production in one of the *F. proliferatum* strains. This study highlights the potential of *A. officinalis* extracts as natural antifungal agents for managing *Fusarium*-induced plant diseases.

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## **P85 - Innovative and sustainable strategies large-scale production of dsRNAs in vivo: a case study on grapevine and *B. cinerea***

Biocontrol, microbiomes and biostimulants

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Grapevine is a globally significant fruit crop with substantial economic importance but also notable environmental impacts. Developing sustainable defense strategies against pathogens, particularly fungal diseases, is essential for reducing agriculture's ecological footprint. RNA interference (RNAi) has emerged as a promising alternative, allowing the direct application of exogenous double-stranded RNA (dsRNA) or small interfering RNA (siRNA) to plants, circumventing the limitations of genetic transformation. While studies have demonstrated the efficacy of synthetic dsRNA against several fungal pathogens including *Botrytis cinerea*, widespread application remains hindered by high production costs and the instability of naked dsRNA in the environment.

To overcome these challenges, our study focuses on developing cost-effective dsRNA production protocols using molecular farming and enhancing its stability through protein-based encapsulation. We designed a plasmid construct that links the dsRNA sequence to a viral-derived P0 capsid protein, from Turnip Crinkle Virus, which encapsulates and protects dsRNA molecules from degradation. Utilizing this plasmid, we are exploring various platforms for dsRNA-P0 expression, including *Nicotiana benthamiana* explants and cell suspension cultures such as BY-2 (*Nicotiana tabacum*) and MM2d (*Arabidopsis thaliana*).

By generating highly specific dsRNA sequences targeting fungal pathogens genes, such as *erg11*, *BcBmp1*, *BcRDR1* of *B. cinerea*, we aim to scale up production and improve environmental stability. This approach could offer a sustainable and scalable alternative to chemical fungicides, contributing to more eco-friendly crop protection strategies in viticulture and beyond.

## **P86 - Intraspecific variation in virulence of a mycoparasitic fungus suggests evolution of host specificity**

Biocontrol, microbiomes and biostimulants

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Microbial parasites of plants can have devastating effects on ecosystem stability, agricultural production and food security. However, parasites can in addition themselves be parasitized by other microorganisms, a phenomenon referred to as hyperparasitism. The ability of microbial hyperparasites to decrease plant pathogen populations through top-down cascades is the basis for their use as biological control agents (BCAs) in agriculture. However, different strains of the same species may display significant variation in biocontrol ability. Therefore, a deeper understanding of the evolutionary dynamics affecting hyperparasite populations and their genomic consequences is needed and may contribute to future biocontrol solutions. In this PhD project, important ecological, genomic and mechanistic questions in necrotrophic fungal parasites of plant pathogenic fungi (mycoparasitism) will be addressed. Initially, 63 strains of the BCA fungus *Clonostachys rosea* are assessed for *in vitro* antagonism and mycoparasitism of several host fungi and oomycetes, including *Rhizoctonia solani*, *Botrytis cinerea*, *Fusarium graminearum* and *Aphanomyces euteiches*. The data collected will be used to test the hypotheses that 1. There is variation between *C. rosea* strains in virulence towards different host fungi, and 2. Certain strains display high virulence towards a specific host (specialization) while others display intermediate virulence towards all hosts (generalism). Differences in virulence is further hypothesized to partially depend on different interaction mechanisms towards the intrinsically different host fungi, which include ascomycetes, a basidiomycete and an oomycete. A genome-wide association approach will be used to identify genomic regions, and thereby genes/alleles, contributing to high virulence towards specific plant pathogenic host fungi. Future work includes transcriptomics study of a limited number of strains selected based on their performance during *in vitro* interaction with plant pathogenic species to further support identification of virulence related genes/alleles.

## **P87 - Investigating barley phyllosphere microbiome to identify potential biological control targets for ramularia leaf spot**

Biocontrol, microbiomes and biostimulants

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Ramularia leaf spot (RLS) caused by *Ramularia collo-cygni* (Rcc) is an emerging disease of barley in temperate regions, including Europe. Although the disease can result in yield loss of between 5% to 25% control is solely reliant on the timely application of fungicides. However, due to environmental safety concerns surrounding the prophylactic use of pesticides in the wider environment alternative control methods are required. An in depth understanding of the barley microbiome and its interaction with Rcc could be helpful in identifying potential biological control targets for RLS. To address knowledge gaps about Rcc-barley phyllosphere microbiome and role of various factor in determining barley microbiome, spring barley field trials were conducted at two Irish locations and role of Rcc, fungicide, geographical location, barley variety and seed source was evaluated in modulating microbiome. Our data suggested a significant role of environment, fungicide treatment and Rcc in modulating barley phyllosphere microbiome.

## **P88 - Investigating the effect of temperature on conversion efficiency of hypovirulent isolates of *Cryphonectria parasitica***

Biocontrol, microbiomes and biostimulants

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The European chestnut (*Castanea sativa*) holds significant economic value for its nuts production, timber, and ornamental value. Its cultivation has faced major challenges due to the invasive fungal pathogen *Cryphonectria parasitica*, which causes chestnut blight. A prominent biocontrol approach relies on hypovirulence, mediated by the infection of the fungal host by the mycovirus *Cryphonectria hypovirus 1* (CHV-1). CHV-1 is horizontally transmitted by hyphal anastomosis among vegetatively compatible fungal individuals and vertically transmitted through conidia. With the progressive temperature shifts resulting from global warming, little is known about the influence of temperature on the conversion efficacy of hypovirulent strains, the molecular mechanisms underlying the hypovirus-fungal interaction, the viability of conidia and the mycoviral load, which were addressed as key parameters in the present study. Vegetatively compatible virulent and hypovirulent isolates of *C. parasitica* were grown *in vitro* at different temperatures to assess their growth pattern, quantity of produced pycnidia and conidia, and the viability and proportion of spores carrying CHV-1. Furthermore, assays on detached branches were conducted under varying temperature settings to determine the production of pycnidia and cankers on the infection sites of inoculated virulent and hypovirulent isolates. Both experiments proceeded with the investigation following the growth pattern, inhibition and conversion capacity of virus-containing strains by co-culturing them with compatible virulent strains. Lastly, temperature-induced molecular responses were determined in both virulent and hypovirulent strains, with a focus on key genes related to fungal growth, stress response, and hypovirus replication. Considering the warming effect due to climate change, a deeper understanding of the environmental and molecular factors affecting hypovirulence offer valuable perspective for improving disease management strategies based on biological control with hypovirulence.

## **P89 - In vitro antifungal activity of oregano and white thyme essential oils against *Plenodomus tracheiphilus***

Biocontrol, microbiomes and biostimulants

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*Plenodomus tracheiphilus*, the causal agent of “mal secco” disease, is a vascular pathogen severely affecting citrus health by causing wilting and dieback, representing a significant threat to citrus production. First reported in 1834 in the Aegean islands of Greece, “mal secco” is believed to have originated in Central Asia (Persia, Afghanistan, and northwestern India), where lemons were first cultivated. Over time, the disease has spread to citrus-growing regions across the Mediterranean basin, including Italy. Essential oils (EOs) are emerging as sustainable alternatives to conventional pesticides, aligning with global efforts to reduce chemical inputs in agriculture. This study evaluated the antifungal activity of commercial oregano (*Origanum vulgare*) and white thyme (*Thymus vulgaris*) essential oils against *P. tracheiphilus*. Oregano EO consisted primarily of carvacrol (72%), while white thyme EO was predominantly composed of thymol (50%), as determined by gas chromatography-mass spectrometry (GC-MS). The minimum inhibitory concentrations (MICs) were determined through *in vitro* assays, with oregano EO exhibiting a MIC of 300 mg/L and white thyme EO having a MIC of 500 mg/L. Both EOs demonstrated fungistatic activity at their respective MICs and exhibited stronger antifungal efficacy than copper-based fungicides used as controls.

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## P90 – Mitigation of drought in quinoa through rhizobacteria from the Bolivian highlands

Biocontrol, microbiomes and biostimulants

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Drought is one of the most destructive abiotic factors for agricultural production, causing considerable yield losses. Quinoa (*Chenopodium quinoa* Willd.) is cultivated worldwide in different environmental conditions due to its nutritional characteristics and ability to grow in harsh environments. This study aims to select drought stress tolerant rhizosphere bacteria from the Bolivian altiplano to evaluate their quinoa growth-promoting capacity, including *in vitro* germination, seedling growth under drought stress in greenhouse conditions and field studies. Rhizosphere soil from the southern highlands of Bolivia was collected to isolate 164 drought-stress tolerant bacteria. From these, 28 strains were shown to produce indole acetic acid, and/or to possess nitrogen-fixing or phosphate solubilizing capacity under *in vitro* conditions. Furthermore, all strains were evaluated for improvement of *in vitro* quinoa seed germination. Based on these properties, nine bacterial strains were formulated in three different matrixes and evaluated for quinoa seedling growth promotion during drought stress in a three-month greenhouse experiment. Three strains were shown to significantly ( $P < 0.05$ ) increase root length of the quinoa seedlings. One strain was selected and shown to significantly ( $P < 0.05$ ) increase leaf number in a field trial under semi-arid conditions in the southern altiplano in Bolivia. DNA sequencing and phylogenetic analyses of the 16S locus putatively identified the three strains with growth-promoting potential under drought stress as members of the genera *Bacillus*, *Pseudomonas* and *Serratia*. Microorganisms from the arid Bolivian altiplano constitute a potential biological source of bioinoculants to improve quinoa productivity and provide sustainable mitigation of climate change effects.

**Impact statement:** In this work, we have identified native bacterial strains from quinoa rhizosphere in the southern altiplano of Bolivia that can increase drought tolerance during the early stage of quinoa growth when applied as a formulated bio-input under greenhouse and field conditions.

**Keywords:** *Bacillus*, Drought tolerance, PGPR, *Pseudomonas*, Quinoa, *Serratia*.

## P91 - Mycobiome of wheat seeds and its association with germination capacity

Biocontrol, microbiomes and biostimulants

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To identify possible associations between germination capacity and fungal infestation of wheat seeds, we used seed lots of two spring wheat varieties (Mirakel and Zebra) grown in Norway in 2016 and 2017. The percentage of seed germination (determined by the ISTA germination method) differed between seed lots and they were grouped accordingly (<90% germinated seeds = low germination capacity, ≥90% germinated seeds = high germination capacity). For each seed lot the percentage of seeds infested by important wheat pathogenic fungi in Norway, namely *Microdochium* spp., *Fusarium* spp., and *Parastagonospora nodorum*, was determined using a plate-out test on PDA. In addition, the DNA-concentration of *F. avenaceum*, *F. culmorum*, *F. graminearum*, *F. poae*, *M. majus*, *M. nivale*, and *P. nodorum* was determined by species-specific qPCR, and the mycobiome was analysed by metabarcoding of ITS 1 and ITS 2.

Seed lots with a low germination capacity were highly infested with *Microdochium* spp. and had a high DNA-concentration of *M. majus*. Germination capacity was also negatively associated with the DNA concentration of *F. graminearum*, despite the low concentrations of *Fusarium* species observed in general. Metabarcoding additionally revealed a negative association between germination capacity and the relative presence of fungi within the genus *Neoscochyta*. *Parastagonospora nodorum* was present at high levels (% of seeds infested and fungal DNA) but seemed not to be associated with seed germination capacity. Our results also indicated some co-existence patterns between fungal species, including both pathogenic and non-pathogenic species, with some species combinations associated with the germination capacity of wheat seed.

## **P92 - Salinity stress on root-associated bacterial community: preliminary findings on antimicrobial activity of bioinoculants**

Biocontrol, microbiomes and biostimulants

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Salinity is one of the main abiotic stresses for global agricultural productivity, impacting plant growth and soil fertility. Microbial inoculants are widely accepted to promote plant tolerance towards salinity. However, there's a lack of knowledge regarding their application and effects under field conditions.

The purpose of this work was to evaluate a panel of selected bioinoculants from Excalibur H2020 project, to enhance the tolerance of tomato plants under salinity stress.

Before conducting the tomato trial with bioinoculants, preliminary analyses were carried out to assess: (i) the antimicrobial activity of bioinoculants through *in vitro* tests and (ii) the impact of salinity stress on the root-associated bacterial community and tomato oxylipin metabolites.

*In vitro* tests showed the antimicrobial activity of bioinoculants against 8 bacterial and 3 fungal tomato pathogens. *Trichoderma harzianum* inhibited the growth of all tested fungi, whereas the *Penicillium variable* inhibited the growth of two plant pathogenic bacteria.

Moreover, salinity stress trial showed how the stress induced a decrease of soil bacterial biodiversity on 7 and 14 days after salt stress imposed (dsi), with a change in microbial community structure from 7 dsi onwards. The higher abundance of potential plant growth promoting soil bacteria, in stressed plants, suggested the role of plants in recruiting selected taxa that might support their response to stress. Furthermore, from day 7, a more accumulation of 9-LOX oxylipins was observed in salt-stressed plants than in control once.

In conclusion, the biocontrol activity of two bioinoculants was assessed against tomato pathogens. Additionally, the rhizosphere bacterial communities and oxylipins profile of salt stressed plants were characterized.

These findings will allow us to optimize the next trial in which the selected bioinoculants will be employed to improve the plant response to salinity stress.

## P94 - Small RNAs mediated biocontrol fungus-plant interactions

Biocontrol, microbiomes and biostimulants

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Small RNA (sRNAs)- mediated RNA silencing is emerging as a key player in host-microbe interactions. However, its role in fungus-plant interactions relevant to biocontrol of plant diseases is yet to be explored. This study aimed to investigate Dicer (DCL)-mediated endogenous and cross-kingdom gene expression regulation in the biocontrol fungus *Clonostachys rosea* and wheat roots during interactions. *C. rosea*  $\Delta dcl2$  strain exhibited significantly higher root colonization than the WT, whereas no significant differences were observed for  $\Delta dcl1$  strains. Dual RNA-seq revealed the upregulation of CAZymes, membrane transporters and effectors coding genes in *C. rosea*, whereas wheat roots responded with the upregulation of stress-related genes and the downregulation of growth-related genes. The expression of many of these genes was downregulated in wheat during the interaction with DCL deletion strains, underscoring the influence of fungal DCL genes on wheat defense response. sRNA sequencing identified 18 wheat miRNAs responsive to *C. rosea*, and three were predicted to target the *C. rosea* polyketide synthase gene *pks29*. Two of these miRNAs (mir\_17532\_x1 and mir\_12061\_x13) were observed to enter *C. rosea* from wheat roots with fluorescence analyses and to downregulate the expression of *pks29*, showing plausible cross-kingdom RNA silencing of the *C. rosea* gene by wheat miRNAs. We provide insights into the mechanisms underlying the interaction between biocontrol fungi and plant roots. Moreover, the study sheds light on the role of sRNA-mediated gene expression regulation in *C. rosea*-wheat interactions and provides preliminary evidence of cross-kingdom RNA silencing between plants and biocontrol fungi.

## **P95 - The effect of mulberry (*Morus L.*) leaf extract on fumonisins biosynthesis and FUM genes expression in *F. proliferatum***

Biocontrol, microbiomes and biostimulants

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*Fusarium proliferatum* is widely recognized as a significant pathogen of a broad range of plants, including crops, ornamentals, and trees. *F. proliferatum* produces a range of secondary metabolites, e.g., fumonisins (FBs) and beauvericin (BEA). These mycotoxins are harmful to plants and animals and contribute to contamination of food and feed, posing a risk to human health. Its prevalence and pathogenicity make it a critical concern in agriculture and horticulture; therefore, new biocontrol solutions are needed.

Mulberry has a high potential as a source of pharmaceutical compounds, which, based on various investigations, exhibited antibacterial, antioxidant, anti-inflammatory, and antitumor effects. The beneficial effects of these bioactive compounds were studied in mulberry extracts containing flavonoids, alkaloids, and phenolic acids, which makes it a promising candidate for developing a biocontrol agent against *F. proliferatum*.

This study aimed to assess the potential of *Morus L.* leaf extracts, obtained using supercritical CO<sub>2</sub>, to analyze the changes in the FBs biosynthesis. Moreover, RT-qPCR analysis was used to examine the expression of two genes from the *FUM* gene cluster responsible for fumonisins biosynthesis in *F. proliferatum* after adding mulberry leaf extract. Samples of mycelia were collected after 0 h, 2 h, 24 h, 72 h, and 120 h of culturing and frozen in liquid nitrogen for RNA extraction. The transcript level of the glyceraldehyde-3-phosphate dehydrogenase gene (GAPDH) was used as an internal control, and the amounts of target transcripts were calculated. Liquid medium samples (in three replicates) were collected to compare the fumonisins (FB<sub>1</sub>-FB<sub>3</sub>) concentration in the medium using the LC/MS/MS method. Investigated mulberry extract strongly affected *Fusarium* metabolism by inhibiting mycotoxin biosynthesis. Moreover, adding leaf extract significantly influenced the expression of two genes from the *FUM* gene cluster.

## **P96 - The potential role of *Bacillus amyloliquefaciens* MBI600 against *P. ultimum* in cotton and its impact on plant defence**

Biocontrol, microbiomes and biostimulants

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Strains of the bacterial genus *Bacillus* spp. are well-known to protect plants against various phytopathogens. In our research, we evaluated the potential of *Bacillus amyloliquefaciens* strain MBI600 as a seed coat to protect cotton seedlings (*Gossypium hirsutum* L.) from the occurrence of damping off and root rot caused by the oomycete pathogen *Pythium ultimum*. For our experiment, cotton seeds were coated with different concentrations of the bacterium and plants were grown under controlled conditions. The pathogenicity metrics of seed rot, post-emergence damping off, and symptom severity were documented for two consecutive years. Comparison was done with seeds coated with conventional fungicides. Our results showed that *B. amyloliquefaciens* MBI600, at the concentration of 160 mL of bacterial inoculum per 100 kg of seeds, can significantly reduce the disease incidence and severity in cotton seedlings. In addition, gene markers associated with key plant defence pathways, such as JA/ET, SA, phenylpropanoid pathway, and oxidative stress, were analysed to capture the transcriptomic response of cotton plants when *B. amyloliquefaciens* MBI600 was applied, both in the absence and presence of *P. ultimum* pressure. Results highlighted increased relative expression of plant defence marker genes, indicating the activation of the studied pathways by *B. amyloliquefaciens* MBI600. To conclude, our results suggest that *B. amyloliquefaciens* MBI600 can be used as a seed coating to protect cotton seedlings from *P. ultimum*.

**P97 - Unravelling the tomato core microbiome for beneficial microbial consortium design:  
Swedish farms as a case study**

Biocontrol, microbiomes and biostimulants

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Tomato (*Solanum lycopersicum*) is a staple crop and economically important for many countries. However, tomato production is negatively affected by several detrimental diseases such as early blight (*Alternaria solani*) and late blight (*Phytophthora infestans*). The study of tomato microbiome plays a critical role in understanding plant health, growth, and resilience. In particular, understanding the interactions between different members of the plant microbiome, including the pathogens and beneficial microbes, can provide a more holistic view of microbiome-plant interactions useful for formulating effective disease control strategies. Hence, this project's objectives are to: i) characterize the core microbiome of tomato rhizosphere and phyllosphere from geographically distinct commercial farms in Sweden, to get a deeper understanding of the natural microbial structure of healthy tomato plants; ii) investigate whether tomato plants with aboveground *A. solani* or *P. infestans* infection can recruit beneficial rhizosphere microbes to resist subsequent pathogen infection. Through these combined lines of investigation, we aim to gain insight into the core microbiome of tomato rhizosphere and phyllosphere and will be able to assess rhizosphere microbiome shifts upon leaf pathogen attack. Studying the microbial community in the tomato phyllosphere and rhizosphere may pave the way for creating more efficient and sustainable disease management approaches, such as a microbial consortium.

## **P98 - The hidden microbiome of hop pellets: microbial communities and sensory-active metabolites in dry-hopped beers**

Biocontrol, microbiomes and biostimulants

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Hops (*Humulus lupulus*), a perennial climbing plant, are essential to beer production, providing secondary metabolites that contribute bitterness, aromatic qualities, and antimicrobial properties. However, despite these antimicrobial traits, hop pellets used in brewing are not sterile. Recent studies (e.g., Patakova et al., 2024) have confirmed bacterial presence in hop pellets, but comprehensive microbiome analyses, particularly of fungal communities, remain limited. Microbial communities introduced via hops may play a significant role in dry-hopped beers, as dry hopping involves adding hops to cooled, fermented beer rather than during boiling. This process allows alcohol- and acid-tolerant microorganisms to survive and potentially influence flavor and quality through their metabolic activity.

This study utilizes next-generation sequencing and metabolomic analyses to characterize microbial communities in hop samples and dry-hopped beers and assess their metabolic contributions. Samples were collected from various hop pellet types and beer styles before and after dry hopping. Fungal communities were analyzed using ITS2 metabarcoding, while gas chromatography-mass spectrometry (GC-MS) and liquid chromatography-mass spectrometry (LC-MS) identified sensory-active metabolites produced by microorganisms. These compounds, including both volatile and non-volatile metabolites, were evaluated for their potential impact on beer flavor and quality. By integrating microbiome and metabolomic data, this study provides new insights into how microbial communities introduced via hops contribute to the sensory characteristics of dry-hopped beers.

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## **P137 - Pathogens of *Dalbergia sissoo* (Shisham) and their management**

Biocontrol, microbiomes and biostimulants

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*Dalbergia sissoo* is a prominent hardwood species, is susceptible to various pathogens, posing significant challenges to its cultivation and management. This abstract highlights the pathogenicity of several common pathogens infecting Shisham, viz. *Alternaria*, *Cladosporium*, *Nigrospora*, *Periconia*, *Pestalotiopsis*, *Colletotrichum*, *Curvularia*, *Chaetomium* and *Fusarium* validated through pathogenicity testing via detached leaf assay soilborne pathogen bioassay.

Detached leaf assay served as a reliable method to assess the pathogenic potential of these organisms, providing insights into their virulence and host specificity. Results from these assays indicate varying degrees of necrosis, chlorosis, and lesion development on Shisham leaves, highlighting the pathogenic nature of the studied organisms. Furthermore, molecular techniques confirm the presence and identity of these pathogens, elucidating their roles in Shisham foliar diseases. Similarly, the pathogenicity of *Fusarium* spp. was assessed through direct soil inoculation using spore suspensions and fungal biomass to simulate natural infection conditions. Both methods induced characteristic disease symptoms in susceptible host plants, including wilting, chlorosis, necrosis, and vascular discoloration. Biomass inoculation caused faster disease onset, highlighting its higher inoculum potential. The persistence of *Fusarium* in soil was confirmed post-experiment, emphasizing its role as a soilborne pathogen.

This study provides valuable insights into *Fusarium* epidemiology and highlights the need for targeted management strategies such as biocontrol measures, to mitigate its impact on plant health. The antagonistic potential of beneficial microorganisms such as *Trichoderma* and *Rhizobium* against the aforementioned pathogens. These biocontrol agents offer promising strategies for managing Shisham diseases by inhibiting pathogen growth and mitigating disease progression. Through competitive exclusion, mycoparasitism, and induced systemic resistance, *Trichoderma* and *Rhizobium* demonstrated their efficacy in suppressing foliar pathogens and enhancing Shisham health. This research contributes to the development of sustainable disease management strategies for shisham cultivation, ensuring its long-term sustainability in forestry and agroforestry system.

### **P138 - *Achyranthes aspera* addition mitigated stress caused by *Fusarium* in chili plants by modulating physiological functions**

Biocontrol, microbiomes and biostimulants

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*Achyranthes aspera* L. has antifungal activity but its efficiency against *Fusarium* wilt of chilies caused by *Fusarium oxysporum* f. sp. *capsici*, (Fo) under *in vivo* conditions remains unknown. Herein, we evaluated the stress alleviating activity of *A. aspera* against Fo. Eleven treatments were assessed in pots; T1 as the negative control (C), consisting of only chili; T2 as the positive control (Fo1) at a conc. of  $2 \times 10^7$  CFU/mL; T3 as the positive control (Fo2) at a conc. of  $3 \times 10^7$  CFU/mL; T4 as deactivated Fo (DFo1); T5 as deactivated Fo 2 (DFo2); T6 as *A. aspera* weed (1%) (Aa1); T7 as weed (2%) (Aa2); T8 as Fo1+Aa1; T9 as Fo1+Aa2; T10 as Fo2+Aa1; and T11 as Fo2+Aa2. The DFo1, DFo2, Aa1, and Aa2 treatments were included expressly to investigate the individual effects of soil amendments with weed (Aa1 & Aa2) and DFo, not examined previously. Both Aa1 and Aa2 caused positive allelopathy, resulting in increases in morpho-biochemical attributes in chili plants. The DFo treatment caused negative allelopathy in chili plants. Among the treatments, soil amendment with Aa2 had the greatest alleviating efficacy against Fo1, Furthermore, in comparison to chili plants inoculated with Fo, the *A. aspera* amendment decreased the biotic stress by enhancing antioxidant activities in chili plants such as superoxide dismutase (SOD), peroxidase (POD), and catalase (CAT) by 20, 45, and 36%, respectively, and significantly increased photosynthetic rate ( $P_n$ ), transpiration rate ( $E$ ), stomatal conductance ( $g_s$ ), and intercellular CO<sub>2</sub> ( $C_i$ ) by 26, 61, 106, and 18%, respectively, thereby reduced disease incidence (DI), disease severity (DS), area under disease progress curve (AUDPC) DI, and AUDPC (DS) by 60, 82, 63, and 80%, respectively. The present study concluded that *A. aspera* amendment can appreciably mitigate the biotic stress caused by Fo in chili crop.

### **P139 - Biological control of basal rot of onion by soil amendment with *Sisymbrium irio* and *Trichoderma* species**

Biocontrol, microbiomes and biostimulants

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This study was undertaken to evaluate potential of five *Trichoderma* spp. and a weed *Sisymbrium irio* to control basal rot of onion caused by *Fusarium oxysporum* f. sp. *cepae* (FOC). *Trichoderma harzianum* caused the highest suppression (79%) in FOC radial growth in laboratory bioassays followed by *T. viride* (73%), *T. koningii* (71%), *T. pseudokoningii* (71%), and *T. hamatum* (69%). In pot experiment, FOC inoculated soil was amended with *T. viride*, *T. harzianum* and dry biomass of *S. irio* (DBS) for the control of basal rot of onion. The highest disease incidence (97%) was recorded in the positive control treatment (FOC inoculation only). Disease incidence was significantly reduced by 85–100% over positive control in different treatments of *Trichoderma* spp. and DBS. The disease was completely controlled in *T. viride*, and *T. harzianum* + DBS treatments. Application of different soil treatments enhanced fresh yield of onion from 2.93 g in positive control to 76.96–126.93 g and dry weight from 0.55 g to 7.75–16.14 g. The highest increase was due to 3% DBS followed by 3% DBS + *T. harzianum*. Soil amendments also significantly increased the chlorophyll, carotenoid and protein contents of the plants over positive control. The effect of soil amendments on activity of three defense related enzymes namely peroxidase (POX), polyphenol oxidase (PPO) and catalase (CAT) was also studied where all the treatments significantly enhanced PPO and CAT activities over positive control. Root and stem sections stained with FeCl<sub>3</sub> showed an increase in phenolic substances due to different soil amendments. Soil amendments enhanced the expression of POX and PAL (phenylalanin ammonia lyase) gene over positive control. On the other hand, expression of CHI (chitinase) gene was diminished due to *T. harzianum*, *T. viride* alone or together with DBS. It concludes that the highest yield of onion in FOC infected soils can be obtained either by sole application of 3% DBS or together with *T. harzianum*.

## **P140 - Exploring biocontrol efficacy of different *Trichoderma* isolates against three potato pathogens in vitro**

Biocontrol, microbiomes and biostimulants

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Early blight (*Alternaria solani*), late blight (*Phytophthora infestans*), and black scurf (*Rhizoctonia solani*) are economically significant diseases of potato in potato-producing countries worldwide, causing substantial yield losses. Currently, the primary method for controlling these diseases involves the use of synthetic fungicides throughout the growing season. The harmful effects of synthetic fungicides, combined with the rise in fungicide resistance and EU policies aimed at reducing their use, have created an urgent need for more effective alternatives. *Trichoderma* sp. is a widely recognized biocontrol agent due to its strong antagonistic properties, but its effectiveness in colder climates has not been well studied. In the present study, we investigated the biocontrol potential of several *Trichoderma* strains against three potato pathogens under different temperature conditions *in vitro*. Thirteen *Trichoderma* strains were evaluated in a dual culture assay against *A. solani*, *P. infestans*, and *R. solani* under two different temperature regimes: 14°C and 24°C for *A. solani* and *P. infestans* assay, and 10°C and 18°C for the *R. solani* assay. *In vitro* bioassays revealed a significant influence of both strain and temperature on the growth inhibition of the pathogens. *Trichoderma* isolates successfully colonized all three pathogens at higher temperatures; however, their growth inhibition ability varied among the pathogens at lower temperatures. Our study highlights the overall potential of *Trichoderma* spp. as biocontrol agents against potato pathogens, although their effectiveness in colder climates may be limited and should be considered in future research.

## **P141 - Harnessing antimicrobial volatiles of *Bacillus cabrialesii* FA26 for sustainable biocontrol of soilborne fungi**

Biocontrol, microbiomes and biostimulants

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The effectiveness of microbial volatiles as biocontrol agents is often studied under specific laboratory conditions, raising questions about their real-world applicability. This study explores how culture conditions, inoculum concentration, and application methods influence the antimicrobial volatile activity of *Bacillus cabrialesii* FA26 against key soilborne pathogens. FA26 exhibited significant suppression of *Fusarium oxysporum*, *Rhizoctonia solani*, and *Phytophthora capsici* on nutrient agar, while *Sclerotinia sclerotiorum* was better controlled on LB. A dose-dependent effect was observed, with spreading inoculation proving more effective than droplet inoculation. Headspace SPME/GC-MS analysis identified 26 volatiles, eight of which exhibited complete inhibition of at least one pathogen. These findings highlight a direct long-distance biocontrol mechanism and emphasize the importance of optimizing growth conditions to enhance microbial volatile efficacy. This research contributes to the development of sustainable disease management strategies, reducing reliance on chemical fungicides and improving plant health in agricultural systems.

**P142 - Nematicidal activity of pyrolisate of *Bambusa vulgaris* on root-knot nematode; *Meloidogyne incognita* of lettuce**

Biocontrol, microbiomes and biostimulants

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Pyroligneous Liquors are obtained from diverse biomasses and have been found to contain

important biologically active components with high phenolic, carbonyl and organic acid contents which have several pest control properties. Bamboo Pyroligneous Liquor (BPL) has proved effective in controlling several insect pests of many crops but its nematicidal potential is yet to be fully determined. We, therefore, investigated the nematicidal activities of Bamboo Pyroligneous Liquor in managing root-knot nematodes of *Lactuca sativa* in greenhouse trials. *Bambusa vulgaris* biomass was carbonised at 500 °C in a locally fabricated pyrolizer with automatic temperature control to obtain the Bamboo Pyroligneous Liquor (BPL). Two weeks old lettuce plants were inoculated with 1,000 juveniles of *Meloidogyne incognita*. BPL was applied at three concentrations of 100, 200 and 400 mg/ml. Results revealed that nematode infected lettuce plants treated with BPL recorded significantly higher ( $P < 0.05$ ) mean number of leaves, plant height and yield compared to the untreated control. In addition, BPL at 400 mg/ml had substantially higher nematicidal effects on both the soil and root population of nematodes of *L. sativa* and compared favourably with the carbofuran treatments. We concluded that BPL could serve as a very potent eco-friendly alternative to synthetic nematicides in managing *M. incognita* of lettuce.

**P143 - Phytopharmaceutical evaluation potential of extracts of *Thévétia peruviana* seeds against sclerotinia of common bean.**

Biocontrol, microbiomes and biostimulants

**ABENG NDOUMOU ELISEE JUNIOR<sup>1</sup>**

<sup>1</sup> ABENG NDOUMOU ELISEE JUNIOR

**TITLE :** évaluation of the phytopharmaceutical potential of extracts of *Thévétia peruviana* seeds against sclerotinia of common bean.

**Presented by :** ABENG NDOUMOU ELISEE, PhD student at the University of Yaoundé 1 in Cameroon.

**ABSTRACT**

We present here in summary a study that was done in Cameroon at the University of Yaoundé 1, Faculty of Sciences more precisely at the Department of Biology and Plant Physiology. This study was carried out within the framework of agricultural biology via the use of extracts (aqueous, acetone and methanol) of *Thévétia peruviana* seeds in the field and in the laboratory to reduce the development of *Sclerotinia sclerotiorum* causal agents of sclerotinia of common bean. These diseases mainly affect the leaves of common bean and can completely destroy sensitive cultivars in less than 10 days and cause yield losses of around 50%. As a general objective, it was a question of evaluating the biopesticide effect of *Thévétia peruviana* seeds. More specifically, the aim was to evaluate the in vitro and then field effect of extracts of *Thévétia peruviana* seeds on the growth, phytosanitary status and yield of common bean. From what results from this work, it appears that methanol extracts have shown a more significant effect than acetone and aqueous extracts in in vitro conditions while in the field acetone extracts have shown themselves to be more effective than methanol and aqueous extracts.

**P144 - Plant growth promoting and abiotic stress tolerant chickpea (*Cicer arietinum* L.) rhizobial isolates.**

Biocontrol, microbiomes and biostimulants

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Chickpea (*Cicer arietinum* L) is an important pulse crop prized for its high protein content and is grown as a food source worldwide, including Ethiopia. However, the yield of chickpea is low due to low soil fertility and the ever-changing abiotic stresses. Therefore, this study aimed at isolation, characterization, and selection of chickpea rhizobia effective in their nitrogen fixation and abiotic stress tolerance potential. As a result, 150 nodule samples were collected from three districts of chickpea-producing areas in South Wollo. The nodules were crushed, and the rhizobia were isolated and characterized by using standard microbiological procedures. Based on the presumptive tests conducted, 103 (68.7%) of the rhizobial isolates were tentatively categorized as *Rhizobium* species. Regarding phosphate solubilization, only 48 (46.7%) solubilized phosphate with a solubilization index ranging from 2.1 to 2.7 mm. Twenty-four (50%) of the isolates were found to be hydrogen cyanide producers. Among the rhizobial isolates tested under greenhouse conditions, 37 (77.1%) of them induced nodulation on their host plant (chickpea). Their symbiotic effectiveness evaluation test confirmed that 16(47.1%), 6(17.6%), 26.47%, and 3(8.8%) were highly effective, effective, low effective, and ineffective, respectively. Of the authenticated rhizobial isolates, 12 (35.5%) of them, including WuCR-15, 16, 17, 18, 19, 20, 23, 30, 31, 32, 36, 38, and 48, accumulated higher shoot dry matter than the positive control. Isolates WuCR- 11, 17, and 36 showed resistance to low and high extreme abiotic stresses of pH, temperature, and salt. Consequently, rhizobial isolates, WuCR- 11, 17, and 36, which were effective and competent in all the tested parameters, were recommended as good rhizobial candidates for applications under greenhouse and field conditions.

## **P145 - *Silybum marianum* incorporation in maize field overcome pathogen infection by improving physiological attributes**

Biocontrol, microbiomes and biostimulants

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In the present study, *in situ* preventive effect of *Silybum marianum* mulches on charcoal rot disease of maize caused by *Macrophomina phaseolina* was studied under field conditions for two consecutive years. The treatments included a negative and a positive control, 2 concentrations of *S. marianum* either alone or in combination with commercial fungicide. The treatment with dead inoculum (DIn) and *S. marianum* alone were included to investigate their effects on maize, not investigated earlier. The most potent concentration of *S. marianum* was 3 kg/m<sup>2</sup> in combination with half dose of fungicide, although *S. marianum* alone also showed significant stimulatory effects, while the addition of (DIn) was found inhibitory on the maize crop. There was 60.1 and 76.3% decrease in area under disease incidence progress curve (AUDIPC) and area under disease severity progress curve (AUDSPC) by the application of *S. marianum* (3kg/m<sup>2</sup>) with half dose of fungicide and *S. marianum* alone over infested control. Addition of *S. marianum* enhanced the biochemical, Chl *a*, Chl *b*, and carotenoids contents up to 55.2, 65.4 and 39.3%, respectively, over infested control. Whereas, physiological attributes such as carbon assimilation rate (*A*), stomatal conductance (*g<sub>s</sub>*), transpiration rate (*E*) and internal CO<sub>2</sub> conc. (*C<sub>i</sub>*) were increased up to 64.6, 46.5, 37.9 and 9.1%. Along with improvement in above said parameters, *S. marianum* (3kg/m<sup>2</sup>) in combination with half dose of fungicide increased the per hectare yield by 37.8% and 35.1%, over infested control, during years 1 and 2, respectively. These results suggest the use of *S. marianum* to manage the charcoal rot disease in maize crop as an ecofriendly plant disease control method. Moreover, the behavior of maize crop towards the application of *S. marianum* and (DIn) suggest that separate treatments are needed to include in soil amendment bioassays in order to avoid false positive or negative results.

# **Epidemiology and ecology of plant diseases**

### **O3.1 - Epidemiological and behavioural modelling of Integrated Pest Management (IPM) uptake in arable farming**

Epidemiology and ecology of plant diseases

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Reducing reliance on pesticides is an important global challenge. With increasing constraints on their use, in recent years there has been a declining trend in pesticide use for arable crops in the UK. But with increasing disease pressures and global demand for food, there is a greater need for effective measures of pest and disease control.

These circumstances highlight the need for widespread adoption of sustainable alternative control measures. Integrated Pest Management (IPM) is one such solution, comprising a set of management strategies which focus on the long-term prevention, detection and control of pests, weeds and diseases. While many of these methods are acknowledged to offer effective control, their implementation has thus far been limited in practice.

As a case study we consider *Septoria tritici*, an economically important disease of wheat. We used epidemiological modelling techniques to investigate the potential of different IPM control strategies. Combining existing data with a deterministic, compartmental infectious disease model of *S. tritici* transmission, we simulated the implementation of an IPM regime into the *S. tritici* disease system. We investigated the outcomes on disease prevalence and crop yield. Given the challenges with uptake of IPM, we also explored how grower behavioural dynamics influence the adoption of IPM practices. We found that IPM measures have the potential to offer individual growers comparable control to a standard fungicide regime, while also reducing the level of external infection incurred by the growers who maintain a fungicide regime.

### **O3.2 - Chocolate spot of faba bean – species complex identification, chemical control, and disease risk modelling**

Epidemiology and ecology of plant diseases

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Chocolate spot (CS) is one of the most destructive diseases affecting faba beans worldwide, leading to yield reductions of up to 90% in susceptible cultivars under conducive environmental conditions. Traditionally, the disease has been attributed to the fungal pathogens *Botrytis fabae* and *Botrytis cinerea*, however recent studies have identified three additional *Botrytis* species capable of causing the disease. Fungicide applications during flowering are commonly used to control the disease and limit damage to pod set, but this approach is not always effective. The reasons for this lack of control are not fully understood.

To increase our understanding of the CS species complex in Norway, we used species-specific PCR to identify different *Botrytis* species in symptomatic leaves collected at various locations and years. Some *Botrytis* species are known to be high-risk pathogens for fungicide resistance development, but resistance in Norwegian *Botrytis* populations in faba bean have not previously been studied. Therefore, we obtained *Botrytis* isolates from diseased leaves and used a mycelial growth assay to assess their response to the active ingredients (boscalid and pyraclostrobin) in the fungicide commonly used for CS control in Norway. Resistance to both boscalid and pyraclostrobin was detected among *B. cinerea* isolates, while only resistance to boscalid was detected among *B. fabae* isolates. To elucidate resistance mechanisms, we analyzed target gene sequences for the presence of mutations known to confer resistance to the two active ingredients. Field experiments were conducted to test the efficacy of various spray timings and fungicides in early and late faba bean varieties. Additionally, we are developing a disease risk model for CS to better understand the conditions that lead to disease and to improve the timing of fungicide applications.

### O3.3 - Epidemiological model of *Xylella fastidiosa* infection in vineyards

Epidemiology and ecology of plant diseases

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*Xylella fastidiosa* Wells (1987) (Proteobacteria: Xanthomonadaceae) is a xylem-limited, bacterium that infects over 600 plant species, including several economically important crops. Native to the Americas, *X. fastidiosa* subsp. *fastidiosa* (*Xff*) has caused severe losses in the U.S. grapevine industry as the causative agent of Pierce's Disease (PD). Since the detection of this subspecies in Mallorca Island (Spain) in 2016, concerns its impact on European viticulture have escalated, highlighting a critical gap in epidemiological tools needed for risk assessment and guide management strategies.

The *Xff* epidemiological system is complex, driven by interactions among the pathogen, insect vectors, and host plants, further modulated by abiotic factors such as temperature and water stress. To address this complexity, we developed an eco-epidemiological model to simulate *Xff* disease dynamics in vineyards by integrating physiological and ecological processes. The model incorporates key system components: herbaceous plants (hosts for vector immature stages), non-susceptible woody plants (vector food sources), reservoir plants (infection sources for vectors), grapevines (structured into compartments based on disease progression, including infectious asymptomatic plants), and vectors (categorized into immature, uninfected adult, and infected adult stages). Model parameters were derived from published laboratory and field data. Model calibration was performed based on a case study of an *Xff* outbreak in Southern Europe.

This model represents a significant step forward in understanding *Xff* epidemiology, providing valuable insights into the potential risk faced by wine-growing regions in Europe. Furthermore, it provides a critical foundation for designing effective strategies to prevent and mitigate the establishment and spread of this pathogen in vineyards.

### **O3.4 - Unveiling species-specific variability in the environmental needs of *Colletotrichum* spp. causing strawberry anthracnose**

Epidemiology and ecology of plant diseases

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Some species in the genus *Colletotrichum* are listed among the most important fungal pathogens causing plant diseases. *Colletotrichum* spp. are primarily reported as causal agents of anthracnose, occurring on plant leaves, stems, flowers, and fruits. These pathogens are able to cause serious pre- and post-harvest losses, thanks to the various lifestyles and the high variety of host plants. The complexity of species among the genus makes the epidemiological investigation a pivotal topic in agriculture and food security context. The current work comprises epidemiological studies on strawberry (*Fragaria x ananassa*) anthracnose to understand the effect of *Colletotrichum* species and environmental factors on the development of epidemics. Three *Colletotrichum* species, phylogenetically related and commonly isolated from strawberries, i.e., *C. nymphaeae*, *C. fiorinae*, and *C. godetiae*, were compared for their response to temperature and wetness for conidial germination and infection, length of latency period, and production of secondary inoculum. Experiments were performed under controlled environmental conditions to catch the pathogen response over the tested environmental variables, with minimal interference from uncontrolled factors. Conidial germination initiated after two hours of wetness at the optimum temperature (25°C) and showed a similar dynamic in response to the combined effect of temperature and wetness, irrespectively of the species. However, species were significantly ( $P < 0.05$ ) different in the ability to infect strawberries, with *C. nymphaeae* showing the highest disease severity. Although the response of latency period to temperature was similar among species, *in vivo* sporulation assessment revealed a higher capacity of *C. fiorinae* to produce spores compared to the other two species at all the tested temperatures. These findings bring new insights on strawberry anthracnose and may help to develop species-specific mathematical equations, which can be included in predictive models for disease outbreak and epidemic progress.

### **O3.5 - Engaging complexity of *Alternaria*-tomato pathosystem: from ecological studies to the development of a mechanistic model**

Epidemiology and ecology of plant diseases

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*Alternaria* spp. pose a threat to tomato production worldwide and to human health for their ability to produce mycotoxins. Although early blight and brown spot diseases have been known for many years, the undergoing taxonomical review of the genus *Alternaria* and the emerging of new species associated to tomato has led to a confusing and ineffective disease management. This research wants to review the current information on *Alternaria* spp. affecting tomato and address knowledge gaps to develop a mechanistic, weather-driven model to predict the occurrence of epidemics and mycotoxin contamination. About one hundred papers reporting results on *Alternaria* ecology were used to investigate the relationships between main pathogen biological processes and the environment. Furthermore, specific ecological studies were performed to address knowledge gaps, especially for poorly investigated processes as mycotoxin production or regarding recently identified fungal species like the *A. tenuissima*. New findings were achieved, revealing similarities in the environmental requirements of *Alternaria* spp., and very limited strain related variability tested for *A. tenuissima*. The overall knowledge retrieved was analyzed by means of system analysis and pooled together to develop the logical and mathematical framework of a mechanistic model, which incorporates key steps of the infection cycle in relation to the host state and the environment. The following compartments were fully characterized and implemented into the model structure: i) primary inoculum production, ii) spore borne infection, iii) incubation and latency period, iv) secondary inoculum production, and v) accumulation of mycotoxins into plant tissues. Model parametrization was carried out for the main *Alternaria* species affecting tomato, and model outputs embed the behavior of single species as well as their co-occurrence during an epidemic. The proposed framework therefore enables the representation of the complex *Alternaria*-tomato system and the prediction of risk in diverse epidemiological context.

### **O3.6 - Rising threats: The growing concern of ergot contamination in cereal grains**

Epidemiology and ecology of plant diseases

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Ergot, caused by *Claviceps purpurea*, is a disease with serious consequences for cereal production. The most problematic aspect of the disease is the production of toxic ergot alkaloids in affected plant tissue. Concerns about ergot have recently increased in Sweden, both because of the increased incidence of the disease and because of new, stricter European regulations on maximum levels of fungal sclerotia and toxins in cereal grain and products. At the same time, there are important gaps in our knowledge of the basic factors involved in the development of the disease and the toxin production, which limits our ability to make reliable recommendations for disease prevention and toxin management.

Considering the recent severe ergot outbreaks, we are investigating potential risk factors and management strategies to reduce the disease and its consequences. In a severe outbreak in 2022, regional effects, probably related to weather conditions at flowering, were pronounced, particularly in winter wheat, while problems were more widespread in rye. Weeds in the crop and an uneven crop development were also correlated with an increased risk of ergot, while the effect of variety choice was less pronounced. Toxin management is complicated by a poor correlation between the presence of sclerotia and toxin content, and by a pronounced variation in the type of toxin produced. Although ergot and its associated mycotoxins have been recognised as a threat since the Middle Ages, they continue to pose significant challenges in modern cereal production, affecting everyone from farmers to bread makers and consumers.

## **O6.6 - The big reset: redefining the nature of disease on the quest for novel resistance traits in cereals**

Epidemiology and ecology of plant diseases

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Plants are colonized by a very large diversity of microbes including both beneficial and pathogenic taxa. Recent discoveries in our field have blurred the lines between such dichotomy, while clearly pinpointing the role of the plant immune system as the major determinant of both beneficial and pathogenic interactions. We have been exploring novel routes to identify plant immunity traits acting as counterparts of such microbial complexity including (i) the genetic basis underlying the ability of different pathogens species to act in a complex, and (ii) the ability of plants to counteract such complex pathobiomes through the evolution of tradeoff-mitigation traits, and population-driven disease resistance traits. We will present our latest work in cereals, with a focus on Fusarium Head Blight, leaf blotch diseases [and to some extent stem rust] in wheat, emmer, and oats. Our results show there are unique genetic signatures differentiating the host responses to single vs. multispecies infections, some of which can be clearly linked to domestication and polyploidy. Furthermore, we were able to unravel a possible genetic model underlying the capacity of wheat to mitigate the tradeoffs resulting from conflicting plant immune response. Finally, we will discuss the evolutionary tractability, and amenability of these novel traits for breeding, and their practical implementation in resistance breeding in oats in the frame of a Nordic Oat Taskforce (NOaT) initiative, and other European projects in collaboration with CREA (Italy) and IPK (Germany).

## **O8.1 - Can temperature adaptation drive *Ralstonia solanacearum* strain K60 range expansion in the future?**

Epidemiology and ecology of plant diseases

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The *Ralstonia solanacearum* species complex (RSSC) consists of a group of phytopathogenic  $\beta$ -proteobacteria responsible for lethal wilts and rots in a wide variety of plants worldwide. The global distribution of RSSC strains is partly due to their ability to adapt and survive across different environments, resulting in spread to new locations in which they were not endemic before. In Europe, sporadic outbreaks of potato brown rot have been historically caused by cool-adapted RSSC strains. However, in the last years, tropical RSSC strains have been detected in cool-temperate regions of Europe, and a 2015 outbreak of warm-adapted *Ralstonia pseudosolanacearum* in greenhouse rose in the Netherlands has emphasized the potential risk of geographical range expansion of tropical RSSC strains into Europe. This study used experimental evolution to assess whether RSSC strain K60 isolated from tomato in the warm-temperate region of North Carolina (US) can adapt to colder climates. The ancestral K60 strain was experimentally evolved in rich liquid media at four different temperatures (15, 20, 28 and 34 °C) over 148 generations, after which phenotypic and genotypic adaptations were determined. The results showed that K60 can rapidly adapt to the 15 °C environment and that the improved growth at this temperature is especially clear in evolved non-mucoid colony types. In addition to identifying underlying genetic mechanisms of cool-temperature adaptation, *in planta* experiments comparing the virulence of ancestral and evolved populations in tomato model will be discussed.

## **O8.2 - Genomic complexity of *Verticillium longisporum*: virulence, genome rearrangements and evolutionary history**

Epidemiology and ecology of plant diseases

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*Verticillium longisporum* is an emerging soilborne fungal pathogen that poses a significant threat to oilseed rape (*Brassica napus*) and other cruciferous crops in the world. Like all its close relatives, *V. longisporum* is an asexual species, but unlike them, it is an allo-diploid species resulting from the hybridisation of at least two different haploid species. This genetic complexity presents challenges in understanding its evolution, host adaptation and virulence mechanisms.

Currently, there are no effective management strategies to control *V. longisporum* infections, making it a persistent problem for Swedish agriculture. With global warming, the pathogen's impact is expected to increase in the coming decades due to favourable climatic conditions that favour its survival and spread. Given the urgency of the problem, a comprehensive genomic approach is essential to develop sustainable disease management strategies.

With a large collection of samples (~100) from different regions of the Northern Hemisphere, we have combined different approaches to describe and understand the evolutionary history of this species. On the one hand, we have measured several phenotypic traits, such as virulence and fungicide tolerance. On the other hand, we are using the power of long-read sequencing technology (PacBio) to generate 100 high-quality genome assemblies of *V. longisporum* isolates. Through various genomic analyses, as SNP-based approaches and orthologous gene studies, we found a structure of three well-defined lineages, confirming the origin of three distinct hybridization events.

This extensive dataset allows us to dissect the evolutionary history of the pathogen, identify key virulence factors and characterise effectors responsible for host interactions. By integrating comparative genomics and functional analyses, we aim to unravel the molecular mechanisms underlying pathogenicity and adaptation.

### **O8.3 - Deciphering the emergence of phytopathogenicity in the *Sarocladium oryzae* species complex through pangenome analyses**

Epidemiology and ecology of plant diseases

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*Sarocladium* is a genus of filamentous fungi within the order *Hypocreales* (phylum *Ascomycota*, subdivision *Pezizomycotina*). Many species in this genus are known to behave as endophytes in *Poaceae* plants, where they often provide benefits such as alleviating abiotic and biotic stress and promoting plant growth. On the other hand, some *Sarocladium* species are associated with plant diseases, especially those within the “*Sarocladium oryzae* species complex” (SOSC), which includes *S. oryzae*, *S. sparsum*, and *S. attenuatum*. SOSC species cause rice sheath rot, a significant disease affecting rice paddies worldwide. Recent studies have revealed notable differences among SOSC species in terms of morphology, pathogenicity, and production of specialized metabolites. However, the evolutionary mechanisms underlying their transition from endophytes to plant pathogens remain poorly understood. To investigate this lifestyle shift, we sequenced the genomes of 21 SOSC strains from diverse locations and we performed several pangenome analyses, integrating 12 previously-published *Sarocladium* genomes. Our findings highlight a remarkable diversity of “biosynthetic gene clusters” involved in specialized metabolite production, including clusters associated with the phytotoxic compound helvolic acid and the antifungal compound cerulenin. We also identified variations in orthologs related to carbohydrate-active enzymes, peptidases, and putative effectors. Additionally, we provide the first characterization of “Starships”, a superfamily of giant transposable elements - in *Sarocladium*, and we discuss their potential role in the ecology of SOSC and related species. These results offer new insights into the genetic and biochemical factors that may have contributed to emergence of SOSC species as phytopathogens.

## **O8.4 - Parallel adaptation and admixture drive the evolution of virulence in the grapevine downy mildew pathogen**

Epidemiology and ecology of plant diseases

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*Plasmopara viticola* is a biotrophic oomycete responsible for grapevine downy mildew, one of the most destructive diseases in viticulture. Breeding for resistant varieties relies on the introgression of partial resistance factors from wild grapes, but virulent *P. viticola* strains are rapidly emerging. We combined quantitative genetics and population genomics to understand the determinants of this adaptation.

We carried out a QTL mapping study using two *P. viticola* F1 populations segregating for virulence towards grapevine resistance factors Rpv10 and Rpv12. For each R gene, one major QTL was detected. Additional virulent strains were also sequenced to allow for genomic comparisons at the identified loci.

For the AvrRpv12 locus, virulent strains consistently presented large deletions encompassing three RXLR genes that are prime candidate effectors. This points to a typical gene-for-gene interaction in which virulence is recessive and results from the loss of an avirulence gene. Population structure of *P. viticola* indicated that different virulent alleles emerged independently in several European winegrowing regions.

By contrast, virulence towards Rpv10 was determined by a dominant QTL enriched in putative effector genes. This mode of inheritance suggests that the locus could act as a suppressor of resistance. The region exhibits structural rearrangements and an extended effector repertoire in the virulent haplotype. Genetic structure analyses showed that it corresponds to an admixed segment which likely originated from a new introduction of the pathogen into Europe.

The identification of these loci will help monitor the evolution of *P. viticola* populations in order to preserve the efficacy of grapevine resistances.

## **O8.5 - Blackleg in Swedish winter oilseed rape - a study of the causal pathogens and race identification**

Epidemiology and ecology of plant diseases

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Blackleg is a serious disease of *Brassica napus* var. *oleifera* and is mainly caused by the fungus *Plenodomus lingam*, and *P. biglobosus*. The disease is of major economic importance worldwide and causes severe yield losses, especially in winter oil seed rape (OSR). It is, however, not fully explored which species that cause the symptoms in OSR in Sweden. *Plenodomus lingam* exhibits a complex race structure, which determine the pathogen's ability to infect different plant genotypes with specific resistance genes (R-genes). The aim of this study was to investigate the occurrence of these fungi and to identify the race composition of *P. lingam* in OSR.

Fresh plants from four OSR cultivars were collected from a field trial in Scania region in November 2022. Stem samples were collected in July 2023. Disease evaluation was performed by visual assessment of symptoms and quantification of pathogen's DNA using Droplet digital PCR (ddPCR). The occurrence of the species was also investigated in OSR fields in different regions. In addition, forty *P. lingam* isolates from five regions were studied in pathogenicity tests using a differential set consisting of 4 lines with R-genes and the susceptible cv. Westar.

The ddPCR result showed that both pathogens are present in the winter OSR field, and that the prevalence differ between cultivars. Further, the proportion between the two species changed between the autumn and summer, *P. biglobosus* was more abundant in the stems in summer than in the leaves previous autumn, while *P. lingam* was equally abundant in autumn as in the summer. We will present the distribution of the pathogens in different OSR fields. All *P. lingam* isolates were virulent on cv. Westar, but not on genotypes with R-genes *LepR1* and *LepR2*. Compatible interaction with differential lines carrying *Rlm3* and *Rlm7* were observed in 24 and 27 isolates, respectively.

## **O8.6 - European Wheat at Risk: Understanding the resurgence and genetic diversity of stem rust**

Epidemiology and ecology of plant diseases

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Wheat stem rust, caused by the fungal pathogen *Puccinia graminis* f. sp. *tritici* (*Pgt*), poses an ongoing threat to global wheat production. After being largely controlled in Europe for six decades, it has re-emerged since 2016, with epidemics affecting both durum and bread wheat in several European regions. This resurgence is attributed to the appearance of new virulent *Pgt* races through exotic incursions. From 2013 to 2024, approximately 600 stem rust isolates obtained from infected wheat, barley, and rye across Europe were studied at Global Rust Reference Center (GRRC), contributing to a deeper understanding of the pathogen's genetic diversity, virulence, and evolutionary dynamics. Wheat stem rust has been detected in 22 European countries in recent years. The Clades III-B (TTRTF), IV-B (TKTTF), and IV-F (TKKTF, TKFTF) were prevalent, indicating clonal reproduction. However, high genetic diversity and the ability to infect wheat was found in populations near *Berberis*, the alternate host, suggesting a potential role in the epidemiology of wheat stem rust in Europe. Localized field studies in Russia, Spain, and Sweden have further confirmed the genetic variability of *Pgt* populations. Seedling and adult plant assays using diverse stem rust isolates from distinct genetic groups revealed that most Scandinavian and European wheat cultivars exhibit susceptibility to the majority of tested races. Additionally, evaluations against two Ug99 races—currently undetected in Europe—showed that these races caused the highest levels of susceptibility across numerous cultivars. Most durum wheat cultivars were susceptible to TTRTF, with the exception of two varieties that demonstrated resistance to all tested races. The widespread susceptibility of commercially grown European wheat varieties underscores the urgent need to accelerate breeding programs focused on enhancing resistance to stem rust. Developing cultivars with broad-spectrum resistance is essential to mitigate the impact of evolving *Pgt* races and safeguard wheat production across Europe.

## **P24 - Molecular identification and fungicide resistance of *Colletotrichum* spp. strains causing olive anthracnose in Greece**

Epidemiology and ecology of plant diseases

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Olive anthracnose disease, caused by fungi belonging to *Colletotrichum* species, stands as a major global threat to olive drupes, leading to significant quantitative and qualitative deterioration of harvested products. This study aims to identify the *Colletotrichum* species associated with olive anthracnose disease in Greece and investigate the sensitivity of several *Colletotrichum* spp. strains to various chemical plant protection products (PPPs). As part of the above objectives, 110 *Colletotrichum* spp. strains were isolated from symptomatic olive drupes and leaves from different olive-producing areas of the country, while a multilocus analysis of ITS, TUB2, and GAPDH genes was conducted for their molecular identification. Selected strains were *in vitro* evaluated for their sensitivity/resistance to ten different fungicides, assessing the inhibitory effect on mycelial growth and conidial germination. Subsequently, detached olive drupes of Greek olive cultivars *cv.* Kalamon and *cv.* Koroneiki were artificially inoculated with five *Colletotrichum* spp. isolates and six different PPPs approved for olive cultivation in Greece were evaluated focusing on disease severity reduction and inhibition of fungal conidia production of each strain. The analysis found that the *C. acutatum* species complex is mainly responsible for the disease in Greece, with some of the isolates showing strong resistance profiles to several approved PPPs.

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### **P30 - Signals of adaption through positive selection among fungi causing Annosum root rot**

Epidemiology and ecology of plant diseases

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Annosum root rot disease causes severe losses to northern hemisphere forestry due to loss of timber, tree mortality and decreased carbon sequestration. The disease is caused by species from the *Heterobasidion annosum s.l.* species complex consisting of five species with different but overlapping conifer hosts. Two species, *H. annosum* and *H. irregulare*, prefer pine hosts, whereas *H. occidentale*, *H. parviporum* and *H. abietinum* prefer non-pine hosts (e.g. spruce and fir). In this study, we contrast the pine type species to the non-pine type species in a phylogenetic context with *Stereum hirsutum* as outgroup to infer evolutionary differences associated with host adaption. We sequenced, assembled and annotated the genomes of the *Heterobasidion* species. Initial modelling of gene family copy number evolution with CAFÉ 5 revealed that the spruce and pine type genomes are highly similar with only 20 gene families showing significant, but small changes in gene family size. A majority of the gene families with significant changes in size appears to be involved in secondary metabolism and/or cyclic compound metabolism based on InterProScan annotations. Similarly, single copy orthologous were highly conserved with a median non-synonymous to synonymous (dN/dS) ratio of 0.05 (PAML/codeml). Interestingly, 146 out of 1304 single copy orthologs show a significantly better fit for PAML models allowing sites with positive selection rather than only negative selection or neutral evolution. GeneOntology terms related to cyclic and aromatic compounds are overrepresented among these gene models. Our results indicate that adaption to different host types between the *Heterobasidion* species appears to be mostly a process of change-of-function among existing genes. Furthermore, most of the changes detected are related to secondary metabolism, enabling the fungus to cope with the varying biochemical environments and/or produce defence substances.

### **P31 - Distribution and genetic characterization of Flavescence dorée Phytoplasma in South Tyrol (northern Italy).**

Epidemiology and ecology of plant diseases

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The disease *Flavescence dorée* is caused by phytoplasmas of the 16SrV group (FDp, *Candidatus Phytoplasma vitis*) and represents a major threat to European viticulture. This phytoplasmosis was first detected in South Tyrol (northern Italy) in 2018. Since then, as part of a province-wide monitoring program, 4,890 samples have been analyzed for the presence of FDp according to Pelletier et al. (2009), which allows the simultaneous detection of grapevine phytoplasmas from the 16SrV and 16SrXII-A groups, enabling the parallel diagnosis of *Bois Noir* phytoplasma (BNp; stolbur group, 16SrXII, *Ca. P. solani*). Over the past six years, a continuous increase in FDp-positive samples has been observed, rising from 1.3% in 2018 to up to 29% of symptomatic grapevines testing positive for FDp in 2024. Meanwhile, the presence of BNp in symptomatic grapevines decreased from 89% in 2018 to 58% in 2024.

To genetically characterize the FDp population in South Tyrol, 87 FDp-positive samples collected between 2018 and 2022 were assessed according to Rossi et al. (2019), based on a segment of the *dnaK* gene. All samples tested so far were assigned to the *dnaK1* genotype, corresponding to FD-D as defined by Martini et al. (1999). These findings are consistent with observations from other Italian wine-growing regions. Future analyses are planned to include FDp-positive alternative host plants and to assess the distribution of genotypes within the South Tyrolean cultivation area.

### **P32 - A seven-year study on sensitivity shifts and target site alterations in *Zymoseptoria tritici* population**

Epidemiology and ecology of plant diseases

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Winter wheat is the main field crop in Estonia and farmer's income relies on the quality and yield of the crop. Therefore, diseases are managed by applying synthetic fungicides two to three times per season. It is crucial to monitor the local *Z. tritici* population for alterations in fungicide target sites to follow up on the adaptation and sensitivity shifts, allowing for early detection of any decline in fungicide efficacy.

Over the course of this study, more than 900 *Z. tritici* isolates were collected from Estonian commercial winter wheat fields in seven years and assessed for fungicide sensitivities of several DMIs, SDHs, and QoIs. Also, prevalent alterations in fungicide target sites (SDH, CYP51, CytB) were studied.

Throughout these seven years, the prevalence of G143A alterations in the *Z. tritici* population increased from 50% to full prevalence, significantly impacting the sensitivity to azoxystrobin and pyraclostrobin. In CYP51, the frequency of alterations V136A, A379G, and S524T steadily rose, with the most notable change being an increase in S524T alterations from 10% to 50% of the overall population. Before 2023, alterations such as B-N225I, B-T268I, C-W80S, C-N86S, or C-H152R in SDH subunits, which influence SDHI sensitivity, were rare, detected in less than 10% of the isolates. However, in 2023, there was a rapid increase of isolates with C-N86S alteration up to 25% of the population. By 2024, up to 50% of the isolates had a relevant alteration in the SDH-C subunit, with C-N86S and C-T79N being the most prevalent, followed by C-H152R. The decrease in sensitivity to fluxapyroxad and bixafen was in line with a higher rate of relevant alterations in the SDH molecule.

Overall, the *Z. tritici* field population remains under continuous selection pressure from various fungicide active ingredients used in commercial fields, indicating that the evolution of fungicide resistance is ongoing.

### **P33 - Emergence of new aggressive *Plasmodiophora brassicae* pathotypes causing clubroot in central Europe and Sweden**

Epidemiology and ecology of plant diseases

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*Plasmodiophora brassicae*, the causal agent of clubroot disease in *Brassica* plants, poses an increasing threat to European oilseed rape (*Brassica napus*) production. Regular monitoring of *P. brassicae*-virulence spectra and identifying new pathotypes are important for developing effective disease management strategies through resistance breeding. Additionally, characterizing virulent pathotypes is essential for understanding non-pathotype-specific resistance. In this study, 17 *Brassica* hosts—including the European Clubroot Differentials (ECD) and the Somé set—along with the clubroot-resistant oilseed rape cultivar *Mendel*, were used to assess the virulence patterns of 84 *P. brassicae* isolates collected from the Czech Republic (CZ), Germany (DE), Poland (PL), and Sweden (SW). Based on the Somé set, six *P. brassicae* pathotypes were identified. While only one pathotype was found in SW, four distinct pathotypes were detected across CZ, DE, and PL. Of the eight hypothetical pathotypes defined by the Somé system, P7 and P8 were absent, whereas P1–P6 were observed at varying frequencies among the countries, with P1 and P3 being the most prevalent. Additionally, the pathotypes P1(+), P2(+), and P3(+) were detected, with P1(+) being the most frequent. These virulent pathotypes were capable of overcoming the resistance of *B. napus* cv. *Mendel*. According to the ECD set, virulence analysis differentiated the 84 *P. brassicae* isolates from Central Europe and Sweden into 42 distinct pathotypes. The most prevalent were 16/31/31 (identified in DE, PL, and SW) and 16/06/12 (found in CZ, DE, and PL). Overall, analysis of all samples revealed significant negative correlations between clubroot incidence and both soil pH and the frequency of oilseed rape in crop rotation, highlighting key factors influencing disease prevalence.

### **P34 - Assessment of buried sclerotia germination and carpogenic variability among *Sclerotinia sclerotiorum* isolates**

Epidemiology and ecology of plant diseases

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*Sclerotinia sclerotiorum* (Lib.) de Bary is a major necrotrophic, soil-borne pathogen responsible for Sclerotinia stem rot in oilseed rape. The sclerotia of *S. sclerotiorum* serve as the primary inoculum for initiating infections. Between 2020 and 2022, a comprehensive national survey was conducted across major oilseed rape-growing regions in Germany. Numerous sclerotia samples were collected, leading to the isolation and purification of 62 *S. sclerotiorum* strains. Subsequent analyses revealed significant variability in cultural characteristics, and virulence among these isolates; however, differences in their carpogenic germination remain poorly understood. This study aimed to evaluate the impact of environmental factors on sclerotial germination under field conditions by establishing sclerotia depots and monitoring apothecia emergence over four consecutive years. Additionally, the effects of soil moisture (25%, 50%, 75%, and 95%), air temperature (10°C/10°C, 14°C/10°C, and 18°C/10°C), and light conditions (white and UV light) on sclerotial germination and apothecial formation were examined under controlled conditions using a 17-hour day/7-hour night cycle. Furthermore, variability in carpogenic germination among *S. sclerotiorum* isolates was investigated. Between 2021 and 2024, significant differences were observed in both the germination rate of sclerotia and the timing of germination throughout the season. Under field conditions, high soil moisture, particularly prolonged wetness, and soil temperatures ranging from 10°C to 14°C were critical for apothecial formation. Under controlled conditions, higher soil moisture levels (75% and 95%) accelerated sclerotial germination, with sclerotia incubated at 14°C/10°C germinating earlier (after 38 days) than those at 10°C/10°C or 18°C/10°C. Light conditions also played a crucial role, as apothecial formation occurred only in treatments exposed to a combination of white and UV light. Moreover, significant variations were observed in the time required for different *S. sclerotiorum* isolates to produce the first stipe and first apothecium, indicating that genetic differences among isolates influence carpogenic germination.

### **P37 - Identification, virulence, and fungicide sensitivity of *Sclerotinia* spp. isolates causing stem rot in oilseed rape**

Epidemiology and ecology of plant diseases

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*Sclerotinia sclerotiorum* causes Sclerotinia stem rot (SSR) in various Brassica species. Recently, the related pathogen *S. subarctica* has been reported on Brassica, causing symptoms identical to those of *S. sclerotiorum*. The significance of *S. subarctica* in infecting economically important crops, such as oilseed rape (*B. napus*), as well as the pathogen's virulence and susceptibility to fungicides, remains to be determined. In an ongoing study, *Sclerotinia*-infected oilseed rape plants were collected across Germany and *Sclerotinia* spp. were isolated. To identify whether *S. subarctica* is present among the isolates, the rRNA ITS region was amplified and sequenced. In parallel, the virulence and pathogenicity of the isolates were evaluated through their inoculation into five oilseed rape cultivars at the cotyledon stage. Of these, 17 had low (lesion area below 10%), 40 had moderate (lesion area between 10% and 30%), and seven showed high virulence (lesion area above 30%). Furthermore, all isolates were evaluated under *in vitro* conditions for their sensitivity against different groups of fungicides including azoxystrobin, boscalid, fludioxonil, mefentrifluconazole, metconazole and prothioconazole. The mycelial growth was measured and EC50 values were determined. The preliminary results demonstrated a significant difference in inhibitory efficacy among fungicides and among the isolates.

### **P38 - Maize-bean intercropping: Effects on plant development and Fusarium toxin accumulation**

Epidemiology and ecology of plant diseases

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Seed- and soil-borne pathogens of the genera *Fusarium*, *Rhizoctonia*, and *Pythium* that infect maize plants during the seedling stage pose a threat to optimal seedling emergence, plant development and yield stability. Additionally, *Fusarium* infections may result in the accumulation of mycotoxins in the harvested material. To mitigate the risk of infections, diversified crop rotations and integrated pest management strategies are often recommended. Maize-bean intercropping are known to improve biodiversity and pole beans have proven effective because they fix nitrogen for the maize plants. The maize-pole bean silage is used as feed for cattle and pigs as higher crude protein levels are achieved compared to pure maize silage. The extent to which *Fusarium* infection of the seeds affects the growth and yield of the maize-bean mixture, and whether it leads to an accumulation of mycotoxins in the silage maize, are the subjects of our investigations. Therefore, we cultivated two maize bean mixtures in 2024 on two field sites in Braunschweig. Seeds were either non- treated (control), or infected with *F. culmorum* spores. In addition, maize was grown alone. For all field trials, four plots per mixture and treatment were sown in a randomized block design. Emergence and early development of the plants were recorded at growth stages BBCH14 and BBCH32. Finally, silage maize was harvested at the respective maturity stages of the plants to collect yield data. Subsamples were further analyzed for *Fusarium* toxin contamination. In detail, we will provide data on zearalenone, deoxynivalenol and fumonisin contents in relation to the mixtures and seed treatments.

**P39 - Determining the identity and virulence of *Stemphylium* species on legumes in Saskatchewan, Canada.**

Epidemiology and ecology of plant diseases

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Field surveys of *Vicia faba* L. (faba bean) conducted in 2017 to 2021 in the Canadian province of Saskatchewan revealed that a large proportion of leaf samples were infected with isolates of *Stemphylium* species. In 2022, sequencing of diagnostic loci of a group of *Stemphylium* isolates causing severe leaf blight symptoms on faba beans identified *S. eturmiunum* as the causal pathogen. The recent phylogenetic revision of the genus *Stemphylium* led to an investigation of species composition of isolates collected from a range of legume host samples collected in Saskatchewan. A phylogenetic analysis was conducted using the internal transcribed spacers and the intervening 5.8S region of the nuclear ribosomal DNA, glyceraldehyde 3-phosphate dehydrogenase, and calmodulin sequences with comparisons to type-isolate sequences. Results indicated three distinct groups of isolates, delineating the species *S. beticola*, *S. eturmiunum*, and *S. vesicarium*. Two experiments were conducted to determine the virulence and host range of isolates of *S. eturmiunum*, *S. beticola*, *S. vesicarium* and *S. botryosum*, the first one on faba bean cultivar CDC 1310, lentil (*Lens culinaris* Medik.) cultivar CDC Robin and alfalfa (*Medicago sativum* L.) cultivar Rugged, and the second one on these three hosts as well as field pea (*Pisum sativum* L.) cultivars CDC Citrine and CDC Huskie, and chickpea (*Cicer arietinum* L.) cultivars CDC Lancer and CDC Vanguard. Although all isolates caused, at a minimum, trace levels of disease on at least one cultivar of each host species, results indicated host specificity of *S. eturmiunum* isolates to faba bean and *S. beticola* isolates to lentil, whereas *S. vesicarium* only produced trace amounts of disease on all hosts evaluated. Due to the high virulence of *S. eturmiunum* isolates on faba bean, incorporation of *Stemphylium* blight resistance in faba bean breeding programs and research into the epidemiology of the disease are warranted.

## **P40 - Distribution of *Cronartium pini* throughout northern Sweden and within individual forest stands**

Epidemiology and ecology of plant diseases

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*Cronartium pini* is a rust fungus which causes considerable damage in Sweden's Scots pine forests. It exists in a host alternating and a non-host alternating life-cycle form, called *Cronartium flaccidum* and *Peridermium pini* respectively. Although the distribution of the two life-cycle forms has been analyzed, more detailed information of the distribution of the two forms within individual stands and how they relate to each other is missing. In this project, the importance of site characteristics, weather conditions and plant material for the distribution of the fungus will be analyzed, as well as how they affect the risk of infection by either life-form.

Aeciospore samples were gathered from forests throughout northern Sweden, as well as information describing the stand characteristics around infected trees and the occurrence of *Melampyrum* spp., which is the most important alternate host in this area. Information gathered about the lesions from which the spore samples were taken, included lesion size and location on the tree, as well as the total number of lesions on the tree.

Life-cycle form will be determined by using SSR markers and digital PCR. The samples analyzed with microsatellites, will be used to study how unique multilocus genotypes are spread throughout individual stands and throughout northern Sweden. Preliminary results show that both forms can occur within the same stand more frequently than previously believed.

## **P41 - DMI fungicide selection in *Zymoseptoria tritici* is independent of geographical and genetic background of the pathogen**

Epidemiology and ecology of plant diseases

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*Zymoseptoria tritici* is an important plant pathogen responsible for septoria tritici blotch (STB) on wheat crops worldwide. Demethylation inhibitor (DMI) fungicides are commonly used to manage STB by targeting the synthesis of sterol 14- $\alpha$ -demethylase which is crucial for pathogen cell permeability and encoded by the CYP51 gene. However, prolonged DMI usage has led to *Z. tritici* populations developing reduced sensitivity to this fungicide group. In this study, 311 isolates were collected pre-treatment in nine wheat growing regions in Europe in 2019. High-throughput amplicon-based sequencing of nine housekeeping genes and the CYP51 gene was used to analyze these isolates. Minimum spanning network analysis of the housekeeping gene data showed no population structure among *Z. tritici* samples. We identified several mutations in the CYP51 that were independent of geographical origin. These mutations were combined in different haplotypes most of which had been previously reported as being linked to fungicide resistance. A clear clustering of CYP51 haplotypes was found, indicating selection pressure from DMI use. Overall, these findings show the potential of high-throughput sequencing in monitoring fungicide resistance to support the development of effective and sustainable anti-resistance strategies.

## **P42 - Annual legume crops intercropped with winter oil seed rape are infected by soil borne pathogens**

Epidemiology and ecology of plant diseases

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Winter oilseed rape (OSR) is a profitable branch of production with continuous increased acreage, also in organic production. Organic winter OSR is subject to a large pressure of insect pests and available soil mineral N in early growth stages is of great importance for high yields. Intercropping (IC) winter OSR and legume crops (LCs) may increase functional biodiversity as well as providing N in early spring. The LCs may, however, be sensitive to soilborne root pathogens that may be of disadvantage to LCs grown as cash crops later in the crop rotation.

The main objective of the project was to develop a concept to improve N management in organic winter OSR by IC annual LCs and to assess the influence of IC on the prevalence of insect pests and weeds. Here, we present additional findings from the field trials of the importance of choosing correct LC to avoid root diseases later in the crop rotation.

Two field trials were established in August 2022 in central Sweden and the LCs; faba bean (FB), blue lupin (BL), common vetch (CV) and a clover mix (CM; Egyptian clover, Persian clover and squarrose clover) were seeded simultaneous with winter OSR. The roots were visually assessed for discolouration in November and were molecularly analysed for the root pathogens *Aphanomyces eutheices*, causing pea root rot, and *Fusarium avenaceum*, *Phoma* spp and *Cylindrocarpon destructans*, causing root rot in clovers.

The combined results of the visual assessments of root discolouration and molecular analyses of fungal pathogen abundance in the roots provided an important indication of a potential build-up of inoculum in the soil. BL, FB and CM should be avoided as IC in rotations where clover leys are grown. The results also reflect the history of legume cultivation in the field, which is often intensive in organic fields

### **P43 - Fungal diversity of chestnut blight cankers in the Lake Garda area of San Zeno di Montagna (northern Italy)**

Epidemiology and ecology of plant diseases

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The European chestnut (*Castanea sativa*) is widely affected by the fungal pathogen *Cryphonectria parasitica*, which causes chestnut blight cankers. The cankers are also prone to invasion by other fungal species, which can interfere with the development of necrotic lesions. In Italy, the fungal community associated with chestnut blight cankers has been barely studied and its impact on the severity of chestnut blight cankers remains largely unknown. In February 2024, bark cankers from 89 chestnut trees in the area of San Zeno di Montagna (Province of Verona) in northern Italy were sampled. Fungal isolation on potato dextrose agar (PDA) primarily targeted *C. parasitica*, and 89 pure cultures from 87 cankers could be isolated. In addition, 42 pure isolates of other fungal species were obtained. In order to identify these cultures, they were grouped according to colony appearance and color. Twenty-six isolates, representative for each phenotype or phenotypic group, were finally selected for DNA sequence analysis of the internal transcribed spacer (ITS) region. In addition, 26 cultures of *C. parasitica* were included in ITS sequencing. Based on ITS sequences, 16 different fungal taxa, apart from *C. parasitica*, could be identified. Of these, *Gnomoniopsis smithogilyvi*, associated with brown rot of chestnut fruits, most commonly coexisted in chestnut-blight cankers as it was isolated from 20% of the samples. The study also identified four isolates producing abundant amounts of spores, which could be assigned to *Penicillium glandicola* and *Trichoderma* spp. The former species showed an inhibitory effect on co-cultures with *C. parasitica* and *G. smithogilyvi* and should be further investigated for its antagonistic capacity as a potential biological control agent. In order to identify all the fungal taxa to the species level, a multilocus sequence typing analysis is under way.

## **P46 - Genetic diversity in *Puccinia triticina* populations on wheat in Denmark**

Epidemiology and ecology of plant diseases

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Utilizing disease-resistant cereal varieties is a cost-effective and efficient approach to prevent diseases in wheat and is crucial for fulfilling the aim of increasing the area under organic cultivation to 25% by 2030. In 2024, leaf rust (*Puccinia triticina* Eriks) became extremely widespread in Denmark, and severe attacks were observed in several cereal species, highlighting the significance of this rust disease in a future climate characterized by milder winters and higher temperatures and humidity (precipitation) in the summer. This heightened occurrence served as the motivation for commencing genotyping and race typing at the Global Rust Reference Centre (GRRC) in Denmark. Consequently, a study of diversity in Danish populations of leaf rust was initiated with the goal of identifying representative "new" populations for future infection trials with leaf rust and sharing this information with plant breeders for variety selection. Field samples were collected in 2024 from diversified wheat host cultivars and locations in Denmark and elsewhere. Samples were processed for recovery and multiplication using standard GRRC procedures for development of single pustule isolates, which formed the bases for SSR genotyping using 19 primer pairs and virulence phenotyping using an international set of wheat lines representing diversified Lr-genes. The Danish results from 2024 were analyzed and compared with reference isolates from the past, as well as with isolates developed from incoming samples at GRRC targeting yellow and/or stem rust during the years.

## **P47 - Genetic diversity of new strains of begomoviruses infecting tomato in Oman**

Epidemiology and ecology of plant diseases

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In Oman, vegetable crops have severely been infected by geminivirus disease. In South-and North of Oman, obvious virus-like symptoms were observed particularly on tomato crop. The disease in the north of the country has previously been shown to be caused by different begomoviruses (family Geminiviridae). A comprehensive survey conducted in Oman confirmed the presence of tomato leaf curl diseases (ToLCD). In this study the analysis of diverse tomato infecting viruses isolated from tomato exhibiting ToLCD symptoms originating from south and north of Oman is reported. Forty-nine sequences of seven viruses (six monopartite and one bipartite begomoviruses) were characterized from tomato host plant. The viruses identified are TYLCV-OM, ToLCBrV, ToLCLwV, ToLCSDV-OM, CLCuGeV-AB, ChLCV-OM, and MYMIV. All of these clones show them to have highest nucleotide identity with the corresponding virus isolates reported earlier. However, the sequences of sixteen clones of ChLCV-OM exhibited 93.54 to 100 % nucleotide identity to each other presenting to belong the same begomovirus species. Among these, five isolates grouped separately as G1 share 93.54 to 94.08 % identity with the remaining eleven isolates grouped as G2. This indicates the occurrence of two strains of ChLCV-OM in tomato in Oman. Twenty-one potentially full-length clones of Tomato leaf curl betasatellite (ToLCB) were also characterized associated with different ToLCD infecting begomoviruses. The phylogenetic analysis results based on viruses and betasatellites formed closed clusters with the respective viruses and the betasatellites. The findings of this study confirm the significance of geminivirus diseases on tomato crop in Oman and highlight the latest complex association of ToLCD.

## **P48 - International monitoring and characterisation of *Fusarium* spp. causing root rot on asparagus and pea**

Epidemiology and ecology of plant diseases

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Germany has the largest area of asparagus (*Asparagus officinalis* L.) cultivation in Europe, followed by Spain and Italy. For pea (*Pisum sativum* L.) the three most important producers in the EU are France, Spain and Germany. For both crops, root rot caused by *Fusarium* spp. is of great importance. In asparagus; it is considered being the main component of asparagus decline syndrome and in pea, this can result in a 30 to 50% loss of yield. Several *Fusarium* species are present in fields, but not all of them are pathogenic. In cooperation with breeders, plant protection advisors and horticultural associations, a multi-year monitoring programme for the fungal pathogen *Fusarium* spp. on peas and asparagus is conducted.

*Fusarium* strains occurring in the major cultivation areas are isolated, determined at species level and their pathogenicity and virulence evaluated. For pea, 88 sites across Europe have been sampled over the past three years and 427 isolates have been produced. For asparagus, samples have so far only been taken in Germany at 140 locations over three years, with 429 isolates being taken in culture. Genetic uniformity can be ensured by generating single-spore lines from the various isolates. For pea, seven different *Fusarium* species have been detected on the infected roots, with *F. oxysporum*, *F. solani* and *F. redolens* being the most common. A large number of species were also found in asparagus, with *F. oxysporum* clearly dominating in frequency. In addition, the pathogenicity and virulence of the individual isolates is determined using diversity sets adapted to the crop species. Significant differences in the virulence of individual isolates have already been observed in both pea and asparagus. Early knowledge of changes in the pathogen spectrum is of great importance for breeding, as only the use of representative fungal strains leads to successful resistance breeding.

**P49 - Investigating genomic variability and molecular mechanisms of pathogenesis in ‘*Candidatus Phytoplasma solani*’**

Epidemiology and ecology of plant diseases

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‘*Candidatus Phytoplasma solani*’ (*CaPsol*) is a phloem-limited bacterium associated in Europe and the Mediterranean basin with bois noir (BN) disease of grapevine and with stolbur diseases of tomatoes, potatoes, and other solanaceous crops. Its ecology is highly intricate, involving various insect vectors, primarily the planthopper *Hyalesthes obsoletus*, and a diverse range of host plants within agroecosystems. Developing effective control measures requires understanding the complex interactions between the phytoplasma and its hosts. *CaPsol* pathogenicity relies on effector proteins that disrupt plant hormone signaling, cell cycle regulation, and immune defenses, facilitating colonization, spread, and symptom development. The main aim of this work is to (i) type *CaPsol* strains from BN-affected grapevines by multiple gene (*tufB*, *stamp*, *vmpI*) sequence analyses and to investigate differences in their virulence, studying their prevalence in grapevines exhibiting a range of symptom severity; and (ii) identify genes encoding for phytoplasma secreted effectors from the genomes of nine genetically distinct *CaPsol* strains to evaluate their expression variations in grapevine. *CaPsol* genome analysis was performed to identify protein-coding sequences (CDS) using PRODIGAL. Putative effector prediction was done using SignalP and TargetP, focusing on cleavage sites and signal peptides via neural networks and Markov models. InterProScan classified proteins into families and identified functional domains. Molecular typing results showed a high variability and diversity of *CaPsol* strains infecting grapevines, showing severe and mild symptoms. Genome analysis identified 30 effectors, categorized by localization within or outside sequence variable mosaic (SVM) regions. Primers for qPCR were designed to evaluate effector gene expression in grapevine. Findings would improve our understanding of *CaPsol*-plant interactions, aiding sustainable control strategies.

## **P50 - New insights into the life cycle of *Gemmamyces piceae***

Epidemiology and ecology of plant diseases

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*Gemmamyces* Bud Blight is a serious pathogen that targets the buds of spruce trees. In 2008, *Gemmamyces piceae* was rediscovered in the Czech Republic after being absent for half a century. Since then, this pathogen has destroyed forest stands of *Picea pungens* in the borderland mountains of the Czech Republic and continues to remain active in Europe.

Our study of the pathogen's life cycle is based on available literature and detailed monitoring conducted from 2021-2022. This monitoring took place in the Ore Mountains, where we sampled along a 30 km transect during the growing season (June-September). Samples were evaluated for the size and maturity of fruiting bodies and spores. Some infested buds were marked, and the development of fruiting bodies was observed in subsequent years. Germination was also assessed for some samples using Malt Extract Agar.

The results regarding the maturation of conidia indicate that peak maturation occurs in July of the first year of infestation, which aligns with current literature. However, monitoring the development of fruiting bodies on infected buds in subsequent years revealed that no new fruiting bodies were found on the monitored buds. This suggests that all ascocarps must have been present in the first year of infection, contradicting previously published literature.

Ascospores developed in ascocarps were most frequently observed in June and July of the second year, with rare occurrences in the first season at the end of August and the beginning of September. Sampling of older buds revealed the presence of viable ascospores even on two-year-old buds. These findings indicate that the pathogen's life cycle is more complex than previously understood. The presence of viable spores on two-year-old buds and the observation that all fruiting bodies are present in the first year of infestation should be considered when planning plant protection measures and monitoring efforts.

## **P51 - Phylogenetics meets history: Case studies of watermelon mosaic virus and Grapevine pinot gris virus**

Epidemiology and ecology of plant diseases

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Phylogenetics, the study of the evolutionary history of organisms, has advanced significantly since its origins in the insights of Charles Darwin and Alfred Wallace. Initially, evolutionary relationships were inferred from observing and comparing superficial characters of organisms, but with the advent of molecular biology, phylogenies could be reconstructed mathematically using gene sequences. The development of rapid and affordable sequencing technologies over the past few decades has enabled the reconstruction of viral phylogenies.

Dates can now be added mathematically to the phylogenies, especially if the viruses are evolving at measurable rates, and if their population has been sampled on different occasions. By combining knowledge of the likely dates of nodes in virus phylogenies with historical events affecting agriculture, researchers can explore factors influencing viral evolution, such as host shifts driven by seed trade or changes in agricultural practices triggered by pest invasions. However, high recombination frequencies in some viruses pose a challenge to phylogenetic inference and divergence time estimation, as they can disrupt genetic relationships and introduce conflicting evolutionary signals.

To illustrate these challenges, the evolutionary timescales of two plant viruses with differing recombination frequencies; watermelon mosaic virus (WMV) and grapevine pinot gris virus (GPGV) are reconstructed using two different dating approaches.

## **P52 – Population structure and genetic diversity of *Pyricularia oryzae* causing rice blast disease in Uruguay**

Epidemiology and ecology of plant diseases

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Rice blast, caused by *Pyricularia oryzae*, ranks among the most destructive diseases affecting rice globally. In Uruguay, rice cultivation spanned approximately 149,000 hectares in the 2023-2024 season, with around 40% of this area planted with susceptible varieties. In contrast, during the early 2000s, around 85% of the cultivated area consisted of susceptible cultivars. The blast fungus exhibits a high degree of variability in the field, yet limited information is available regarding its genetic diversity and population structure in Uruguay. To investigate the population structure and genetic diversity of the rice blast pathogen in Uruguay, polymorphic SSR markers were utilized. Isolates were collected from various cultivars and plant organs across the principal rice-growing regions of Uruguay, starting in 1993 and extending to the past season. Genetic diversity was assessed using common 11 SSR markers, 10 of which were polymorphic, resulting in a total of 52 alleles. The number of alleles per locus varied from 2 (pyrms37-38) to 16 (pyrms427-429). The population structure was analyzed using the STRUCTURE program alongside SSR genotypic data, revealing two main groups that were not directly correlated with plant organs or geographical regions. Cluster analysis of blast isolates demonstrated distinct groupings that also lacked correlation with geographical regions, plant organs, or plant subspecies. Nevertheless, some degree of distinctiveness was associated with Japonica cultivars, as indicated by the identification of minor clusters. Analysis of molecular variance (AMOVA) indicated that genetic variation was greater within populations than among them. This study's findings enhance understanding of the genetic diversity of *P. oryzae* in Uruguay over the past decades, considering the adoption of various management practices and cultivar usage. Furthermore, the generated information will contribute to more effective selection strategies that, in conjunction with marker-assisted breeding, can facilitate the development of new rice varieties resistant to blast disease.

## P53 - Prevalence of cereal cyst nematodes in Sweden

Epidemiology and ecology of plant diseases

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Cereals are crops grown worldwide, and they are an important component of food security. However, the production is threatened by yield loss due to infection of cereal cyst nematodes (CCNs). To date, the status of CCNs has not been fully investigated in all cereal-dominated areas in Sweden. Investigations carried out in oat fields during the 1980s in one region in western Sweden showed a wide occurrence of CCNs, especially *Heterodera avenae*. Follow-up surveys in 2018–2019 revealed high prevalence of CCNs, occurring in approx. 50% of the randomly sampled fields. The aim of this study was to close the knowledge gap on the status of CCNs in Swedish cereal production.

Cereal fields located throughout the cereal districts in south central Sweden were selected in consultation with crop advisors. Soil samples were collected in a W-pattern into one pooled sample per field. In total, 190 different fields were sampled in autumn 2023–2024. The nematodes in the soil samples were analysed morphologically for the occurrence of *H. avenae*, *H. filipjevi* and *H. hordecalis*. The cysts were extracted from soil using centrifugation floatation followed by enumeration under microscope. The cysts were then crushed, and the number of eggs and juveniles were counted. We will present the current distribution of CCNs in cereal districts in Sweden revealed through our sampling.

## **P55 - Selection in *Phytophthora infestans* driven by fungicide use**

Epidemiology and ecology of plant diseases

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*Phytophthora infestans* causing late blight is one of the main biotic constraints in potato production worldwide and its management largely depends on fungicides. In Sweden, potato production occupies only 1-2% of the arable land area, while 21% of all fungicides used are applied in potato crops. The combination of the high genetic variability of *P. infestans* and the intensive use of a limited number of fungicides indicates a high risk of fungicide resistance development in Sweden. In 2022 resistance to mandipropamid was confirmed in Denmark in the lineage EU43. In a survey carried out in Sweden in 2022, the lineage EU43 and isolates resistant to mandipropamid have also been found in low frequencies. Resistance to another recently introduced fungicide, oxathiapiprolin, has also been reported from Europe and in 2023 its presence was confirmed in Sweden in one isolate carrying total resistance to both fungicides. This project aims to determine the presence and spread of fungicide resistance in *P. infestans* in Sweden. Samples were collected in field trials with different spraying programs during season 2024 as live isolates and on FTA cards. DNA was extracted from FTA cards and Sanger sequenced to determine the presence of mutations linked to fungicide resistance in the target genes Cesa3 for mandipropamid and ORP1 for oxathiapiprolin. The samples were also genotyped using microsatellite markers. The preliminary results confirmed the presence of mutations for mandipropamid in samples collected from the plots treated with mandipropamid. The majority of these were confirmed as EU43 lineages. The mutation was also detected in an “other” genetically distinct lineage. In addition, the live isolates will be used for resistance phenotyping for both fungicides. Together, these results will allow us to quantify the fungicide resistance situation in Sweden on a population scale.

## **P57 - Species identification and pathogenicity of *Berkeleyomyces rouxiae* associated with black root rot of tobacco in Poland**

Epidemiology and ecology of plant diseases

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Black root rot is a disease affecting many crops such as cotton, carrot, lettuce, peanut and tobacco. The disease is caused by a soil-borne pathogen formerly known as *Thielaviopsis basicola*. A recent taxonomic revision based on DNA sequence data distinguished within *T. basicola* two cryptic species: *Berkeleyomyces basicola* and *B. rouxiae*. Here we aimed to perform species identification, morphological characterization and pathogenicity assays for fungal isolates obtained from tobacco roots with black root rot symptoms in Poland. Sequencing of the three DNA regions (ITS, ACT, MCM7) resulted in obtaining sequences with the highest similarity to *B. rouxiae* sequences deposited in NCBI database. Subsequent phylogenetic analysis confirmed this species assignment as our tobacco isolates (named OT2, OT3, WPT7, WPT8) were included in the same cluster of the phylogenetic tree with other *B. rouxiae* strains including the type species and one tobacco strain from Switzerland. Detailed morphological characterization revealed that the four representative isolates obtained in our study showed a similar structure and gray/brown color of the mycelium and sizes of endoconidia and chlamydospores, which fitted within the ranges previously described for *B. rouxiae*. The growth rate of the cultures varied from 3.8 to 5.1 mm/day depending on the isolate. Pathogenicity assay was performed on young tobacco plants grown in the inoculated peat substrate. All four tested isolates caused symptoms of the black root rot on the susceptible tobacco cv. Virginia Joyner, although no statistically significant difference was found in the disease severity index between plants inoculated with WPT7 and uninoculated control. The other three isolates caused severe disease symptoms and significantly reduced shoot and root dry weight of this cultivar. Therefore, we found strain-specific differences in virulence among the tested isolates. These isolates may be used in future comparative analyses aimed at identifying different pathogenicity genes.

## P58 - Species identification of fungi inhabiting soybean seeds in southern Poland

Epidemiology and ecology of plant diseases

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Fungi have the ability to colonize soybean seeds in the field, during their maturation in the pods and after harvest, during storage. Field fungi like *Alternaria*, *Cladosporium*, and some *Fusarium species* can invade and damage the seeds before harvest. Alternatively, if infected seeds are later sown in the field, they can reduce seedling emergence and cause diseases such as: leaf spot, pod necrosis, seed decay and Fusarium seed rot. On the other hand, fungi colonizing the seeds during storage (e.g. *Aspergillus* and *Penicillium*) may decrease seed vigour and germination percentage. Seed-borne fungi may also cause changes in the shape, colour and biochemical composition of seeds. Some of them have the ability to produce toxins that are harmful to humans and livestock.

The objective of this study was to identify the fungi inhabiting soybean seeds following storage and to determine their germination capacity and chemical composition. The research material constituted twelve soybean seed lots collected from the fields in southern Poland and stored over winter. The germination percentage of these lots ranged between 20.67% and 81.33%. Mycological analysis allowed for obtaining fungal isolates which were subjected to taxonomic identification using microscopic methods and DNA sequencing. A total of 353 fungal isolates from 16 genera were identified, with *Aspergillus*, *Fusarium* and *Alternaria* being the most prevalent. Species identification was successful for 94% of isolates. Twelve seed lots exhibited significant variation in the number and species composition of the isolated fungi. Statistically significant positive correlations were observed between the number of *Aspergillus* and *Penicillium* isolates and the content of free fatty acids. Conversely, the number of *Fusarium* isolates showed a negative correlation with protein and nitrogen content. The results obtained made it possible to identify the fungal species that may be responsible for the reduction in quality of seeds harvested in southern Poland.

## **P60 - Susceptibility of oilseed radish (*Raphanus sativus* supsp *oleiferus*) and some brassica crops to *Plasmodiophora brassicae***

Epidemiology and ecology of plant diseases

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Oilseed radish (OR; *Raphanus sativus* var. *oleiferus*) is grown as a cover crop (CC) in pure stands or in species mixtures. Oilseed radish can deliver multiple benefits and provide several ecosystem services for farmers in their crop production as preventing the leaching of nutrients, improving soil structure and carbon sequestration and reducing plant parasitic nematode populations. There are, however, potential disservices of cover crops, such as disease hosting, that must be taken into consideration. Clubroot, caused by the soilborne obligate pathogen *Plasmodiophora brassicae* is one of the most devastating diseases of brassica oilseed crops, capable of causing significant crop losses. In this study, the prerequisites for integrating OR as a CC in a crop rotation were evaluated using a mixture of Swedish field soils holding natural inoculum of *P. brassicae*. Twelve market OR cultivars (cvs.) were compared with clubroot-resistant (CR) winter oilseed rape (OSR; *Brassica napus*) and other selected species of the Brassicaceae family. The study was performed as a replicated bioassay in a growth chamber. The results show that the OR cultivars were infected, which implies that OR multiplies the pathogen. The susceptibility of the OR cultivars was not significantly different from that of the CR OSR cultivars Alister and Archimedes, but it was significantly different from that of the OSR cv. Mendel. The disease severity index (DSI) for OR cultivars ranged from 2.3 to 9.3, and disease incidence was 3–17%. The best performance was shown by black radish (*Raphanus sativus* var. *niger*) with a DSI of 0.3. For sustainable brassica crop production, we suggest avoiding OR as a cover crop in crop rotations including OSR or other brassica crops, since there is a risk of increasing inoculum in the soil.

## **P61 - Temporal and weather-driven variations in airborne pathogenic spores in forest landscapes**

Epidemiology and ecology of plant diseases

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Forest pathogens represent a significant threat to biodiversity and the stability of forest ecosystems, affecting tree health and altering ecological dynamics. Their spread can have severe ecological and economic consequences, especially in the context of climate change, which may modify their dispersal patterns and disease dynamics. In this regard, obtaining information on their presence at broad spatial and temporal scales is essential for understanding their ecology and managing their impacts.

Spore traps are key tools for detecting airborne propagules of pathogens in a given environment and analyzing their spatiotemporal dynamics. Here, we use a dataset on the presence of pathogen spores in the air to describe temporal and weather driven variations across landscapes. The dataset has been collected through weekly sampling with active and passive spore traps between 2013 and 2024 in three distinct ecosystems in Sweden, and fungal species identified through DNA metabarcoding.

The high temporal resolution of these data allow for the assessment of meteorological factors influencing spore availability in the air and, consequently, the potential dispersal of forest pathogens. In the future, this dataset will facilitate long-term studies on pathogen diversity, community composition, and pathogens interactions. Additionally, its use may contribute to the development of forest management strategies and policies aimed at mitigating the effects of these pathogens on ecosystems and anticipating the impacts of climate change on their dispersal and the damage they cause.

## **P62 - The effect of weather extremes on diseases and pests in Dutch field crops**

Epidemiology and ecology of plant diseases

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As a result of climate change, the Netherlands is increasingly facing extreme weather such as heat waves, drought and heavy rainfall, and climate models predict that such extremes will become more frequent in the future. Weather extremes are expected to lead to changes in the composition of disease and pest populations in several agricultural crops important to the Netherlands, but knowledge about their impact is currently limited. In a new project running from 2025-2028, an interdisciplinary group of researchers from Wageningen University and Research will approach this topic from different angles. First, multi-year data sets will be analysed to link the occurrence of extreme weather events to disease incidence and/or severity. Second, methodologies such as air sampling and metagenomics will be optimised for implementation in subsequent experiments. Third, potato, beetroot and onion will be exposed to different moisture regimes in the field, and the impact of these regimes on the dynamics of a broad range of diseases and pests will be monitored. Fourth, potato will be exposed to the same moisture regimes following inoculation with bacteria and fungi that are adapted to different climatic niches. These treatments will be combined with control measures to assess the extent to which these measures may mitigate the negative impacts of the diseases and pests under extreme conditions. Fifth, simple epidemiological models will be developed and parametrised to generate predictions about the effect of weather extremes on the occurrence of one of the main bacterial diseases of potato. Taken together, the results from these complementary research approaches will paint a first, multifaceted, picture of the effect of weather extremes on diseases and pests in Dutch field crops.

## P63 - The use of biological soil monitoring to track plant pathogenic fungal taxa

Epidemiology and ecology of plant diseases

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High throughput sequencing (HTS) has revolutionized biodiversity monitoring, enabling extensive data collection on various fungal taxa without targeted sampling. Our aim was to evaluate the use of monitoring data to track soil-borne plant pathogenic taxa, gain information about their spatial and temporal occurrence, and identify the abiotic and biotic factors that may influence their distributions. We compiled data on 11 fungal taxa that include soil-borne plant pathogenic fungi with broad host ranges (i.e., more than five host plant species) found in Switzerland. We investigated their occurrence at 30 sites included in the Swiss Soil Monitoring Network using amplicon sequence variants (ASVs) obtained from ITS-based metabarcoding. Samples were annually collected between 2012 and 2016 at ten sites each of the land use types (LUTs), forest, permanent grassland, and arable land. We investigated associations among ASVs, geographic location, and LUT preference. Category discrimination analyses were used to explore the relationships among the presence of these taxa and the abiotic and biotic factors at each site. Most shared ASVs among LUTs were found in both arable and grassland sites. *Verticillium* ASVs were only found at arable and grassland sites, while *Fusarium*, and *Rhizoctonia* ASVs were relatively more abundant at arable and grassland sites than in forests. Among all sites, these taxa showed negative associations with soil organic carbon, total carbon, and C:N ratio, but a positive association with bulk density. Inverse associations were found in ASVs belonging to the genus *Armillaria*, which were found exclusively in forest sites. These analyses highlight how biodiversity monitoring with HTS can contribute information on plant pathogen ecology, including their habitat breadth and the abiotic and biotic factors that drive their distribution. Ultimately, this information can contribute to the development of sustainable control strategies through the avoidance of infected sites and targeted interventions.

## **P64 - Uncovering legume soil fatigue for sustainable expansion of European grain legume cultivation**

Epidemiology and ecology of plant diseases

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Over the past decades, significant efforts have been made to promote the cultivation of legumes. Cultivation of legumes, particularly grain legumes, can reduce the use of mineral nitrogen fertilizers, enhance biodiversity, reduce dependence on imported feed proteins, and improve soil biological properties and humus content. Despite these efforts, grain legumes are still not widely grown.

One major obstacle to legume cultivation is "legume fatigue". Legume fatigue limits the expansion of legume cultivation in many European regions. The exact causes of legume fatigue are not fully understood, but soil-borne diseases interacting with abiotic factors are believed to play a key role. Recent findings suggest that the balance between pathogen load and soil suppressiveness is critical. Some farms and regions do not report legume fatigue as a problem, while others experience severe limitations in legume production. Identifying the causes of this variation is urgent and requires a collaborative effort that covers different environments and includes comprehensive assessments of both biotic and abiotic factors.

In a recently launched project, LeFaSus, a network of farms and long-term experiments has been established to identify the primary factors contributing to legume fatigue. This network spans a transect from southern to northern Europe, including Italy, Germany, Luxembourg, and Norway. The project aims to deliver a reliable set of indicators for both legume fatigue and disease-suppressive soils, linking these indicators to the management practices that likely influenced them. The background and plans for the project will be presented.

## **P66 - Understanding fungicide resistance mechanisms in *Monilinia fructicola***

Epidemiology and ecology of plant diseases

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Brown rot is a devastating disease of stone fruits caused by several *Monilinia* species. The development of resistance by *M. fructicola*, the main agent of the disease, to different chemical fungicides, may reduce their efficacy against the pathogen. The aim of this study was to determine the sensitivity levels of *M. fructicola* isolates from peach orchards in Greece and elucidate the molecular mechanisms of resistance. For this purpose, the sensitivity level of 137 isolates was determined for 7 different fungicides. Fungicides tested were: fenhexamid (HAs), cyprodinil (APs), tebuconazole (DMIs), fludioxonil (PPs), pyraclostrobin (Q<sub>o</sub>Is), boscalid and fluopyram (SDHIs). Each isolate was clustered based on the EC<sub>50</sub> and Resistance Factor values, as sensitive, low, moderate and highly resistant. Noteworthy, high resistance frequencies were observed for cyprodinil (21.8%), fenhexamid (29.9%), fludioxonil (30.65%) and tebuconazole (37.22%), while multiple resistance was conspicuous among those isolates. To investigate the molecular mechanism of resistance, sequences of the target genes *Erg27*, *Mdl*, *Os1*, *MfCyp51*, *Pos5* and subunits b, c, and d of the gene *Sdh* were analyzed to elucidate target site point mutations possibly correlated with the resistance phenotype. Overexpression of the gene *MfCyp51* and *ABC1* transporter was studied as a possible rationalization, too. This information provides an important advancement for risk assessment of chemical fungicide application to manage brown rot of stone fruit.

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”.

## **P146 - Biophysical and agronomical drivers of the spatio-temporal distribution of *Plasmopara viticola* oospores in vineyard soil**

Epidemiology and ecology of plant diseases

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Grapevine downy mildew (GDM), caused by the obligate biotroph oomycete *Plasmopara viticola*, is one of the most destructive diseases in viticulture. Its reproductive cycle is comprised of two parts: the asexual phase, which occurs during the grapevine's growing season and in which the zoospores propagate on green parts of the vine; and the sexual phase, in which the crossing of strains in the leaves leads to the formation of oospores that overwinter in grapevine leaf debris and the surrounding soil. Oospores constitute the primary inoculum for downy mildew epidemics during the following growing season. While conventional GDM management relies primarily on fungicides to prevent the asexual propagation of the pathogen, the primary inoculum is increasingly thought to play a major role in GDM epidemics. Therefore, measures based on reducing the primary inoculum or interrupting the pathogen's sexual cycle have the potential to significantly reduce epidemic pressure. But first, a more thorough understanding of the drivers of GDM primary inoculum spatio-temporal distribution in vineyard soils is needed.

To that end, we carried out an extensive study of *P. viticola* oospore concentrations in vineyard soils across Saint-Emilion (France). Soil samples were collected in November 2023 and February 2024 across 63 Bordeaux châteaux. The soils were analyzed for GDM oospore concentrations using droplet digital Polymerase Chain Reaction (ddPCR) and soil physio-chemical properties. We also took into account vineyard management practices and the disease history of each parcel.

We analyzed the effects of season, soil physio-chemical properties, cover cropping type and duration, vineyard treatments, soil management practices, and GDM sensitivity of the parcel on GDM oospore concentrations in the soil. This work thereby identifies key biophysical and agronomical drivers of *P. viticola* spatio-temporal distribution, contributing to a broader project to make GDM management more sustainable by finding ways to interrupt the pathogen's sexual cycle.

## **P147 - Hidden threats: Undercover viral pathogens of vegetables in Ukraine**

Epidemiology and ecology of plant diseases

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Intensification of agricultural production has led to dramatic changes in the biodiversity of plant viruses. Usually, the common viruses in a certain territory are pathogens having either several transmission routes or a wide host range. Thirty-year monitoring demonstrated that among vegetable crops (in particular, tomato, cucumber, zucchini, onion, garlic) in Ukraine, viruses such as CMV, ToMV, TMV, CGMMV, PVY, ZYMV, WMV-2, OYDV, LYSV circulated annually.

The last 5-year screening of vegetables identified viral pathogens with a limited distribution, which are of concern for producers due to their properties. In particular, the highly virulent tomato spotted wilt virus (TSWV) was found on tomato and pepper plants. Occurrence of TSWV was only 1%, but the virus was detected during one season in three regions of Ukraine, both in greenhouses and in open ground. Another pathogen that was characterized by narrow distribution was cucurbit aphid-borne yellows virus (CABYV). Its occurrence as evaluated was <1%. Nevertheless, the virus has a relatively wide host range, and its detection in Ukraine coincided with an outbreak of the same disease in Europe. Phylogenetic analysis of the part of the coat protein gene of two CABYV isolates from greenhouse cucumber and open-field zucchini showed that these Ukrainian isolates are 95% identical. BLAST search showed that the isolate from cucumber had 99% identity with Turkish isolates (MT219982.1, MT219978.1, MT219984.1), while the isolate from zucchini had 99% identity with isolates from Slovenia (MN145451.1) and Poland (MK059479.1).

These data indicate the emergence and probable spread of plant viruses which have either not been identified or studied in Ukraine yet. The situation with other viruses (for example, tomato torrado virus (ToTV), or latent viruses of garlic and onion) remains unknown. Given the exchange of seed and planting material and import/export of plant products to/from Ukraine, such ‘minor’ pathogens require more attention.

## **P148 – Molecular identification of Ukrainian isolates of Barley yellow dwarf virus-PAV in their vector *Rhopalosiphum padi***

Epidemiology and ecology of plant diseases

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Understanding the mechanisms of plant-virus-vector interaction is fundamental to comprehending their ecology and evolution, and crop protection. The impact of climate change on aphids' feeding behavior and virus transmission efficiency remains questionable. Such data must be considered when modeling virus epidemiology, especially for those persistently transmitted. In Ukraine, outbreaks of barley yellow dwarf virus-PAV (BYDV-PAV) occur periodically, causing significant yield losses of wheat and other cereals. To study the vectoring of BYDV-PAV in Ukraine, we identified virus in *Rhopalosiphum padi* (L.) aphids collected from symptomatic plants in the northern (Kyiv) and southern (Odessa) regions. Aphids were tested for virus occurrence using RT-PCR with primers BYcpF/R (Kundu et al., 2008), leading to the recovery of two BYDV-PAV isolates. Phylogenetic analysis of the 641 bp fragment including the coat protein gene region and the intergenic region revealed 93% identity among these BYDV-PAV isolates. At the same time they were 100% identical to BYDV-PAV-P3 (KU893147.1) and BYDV-PAS-P2 (KU893145.1) isolates from Poland, and to the isolate from the Czech Republic (MN780852.1). The lowest sequence identity (81%) was showed between the studied isolates and isolates from Korea (LC637409) and China (EU332330.1). This is the first report on the genetic diversity of BYDV-PAV isolates in different parts of Ukraine as detected in their vector.

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## **P149 - Testing of populations of selected fungal pathogens of cereals for resistance to fungicides**

Epidemiology and ecology of plant diseases

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### **Testing Fungal Pathogen Populations of Cereals for Fungicide Resistance**

Effective protection against fungal pathogens is critical to maintaining cereal crop yields and quality worldwide. Resistant pathogen populations pose a significant challenge, necessitating strategies to mitigate resistance and optimize crop protection. This study evaluated the susceptibility of fungal pathogens to various fungicidal agents, revealing variability in efficacy and emphasizing the need to reassess certain agents in crop management.

A total of 236 isolates from *Monographella nivalis*, *Oculimacula yallundae*, *Zymoseptoria tritici*, and *Ramularia collo-cygni* were tested against five fungicidal active ingredients (fluxapyroxad, azoxystrobin, fenpicoxamid, prothioconazole, and mefentrifluconazole) at different concentrations. Susceptibility was assessed using EC<sub>50</sub> values, representing the concentration inhibiting 50% of mycelial growth.

Results confirmed significant variability in fungicide susceptibility among isolates. Azoxystrobin exhibited the highest resistance rates, with 94.83% of *Monographella nivalis* isolates showing reduced susceptibility (EC<sub>50</sub> > 0.5 µg/ml), compared to none for prothioconazole and only 6.78% for fenpicoxamid. *Oculimacula yallundae* isolates showed no resistance to prothioconazole or fluxapyroxad. Conversely, *Zymoseptoria tritici* displayed notable variability; 85.51% of isolates were resistant to azoxystrobin, while none showed resistance to fenpicoxamid. *Ramularia collo-cygni* demonstrated resistance to all tested fungicides, highlighting a critical challenge.

The findings underscore the need for ongoing monitoring of resistance in pathogen populations and the development of adaptive management strategies. Integrating diverse control methods and aligning practices with evolving agricultural conditions are essential to sustainable cereal protection. This study contributes valuable insights for optimizing fungicide use and mitigating the risk of resistance development.

**Keywords:** wheat, barley, fungal pathogens, fungicides, resistance, crop protection, monitoring

## **P150 - The occurrence of bean yellow mosaic virus and cucumber mosaic virus in organic cultivation of yellow lupin in Poland**

Epidemiology and ecology of plant diseases

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Yellow lupin (*Lupinus luteus* L.) belongs to the Fabaceae family, and its seeds provide protein-rich animal feed. Recently, there has been growing interest in organic farming. Unfortunately, under such farming conditions, plants are more vulnerable to diseases. Of the viral diseases threatening yellow lupin crops, bean yellow mosaic virus (BYMV) and cucumber mosaic virus (CMV) are of greatest economic importance.

In the present study, the occurrence of BYMV and CMV was determined in 3 locations of organic yellow lupin cultivation in the eastern Poland, i.e. Osiny (Lublin voivodeship), Szepietowo (Podlaskie voivodeship) and in Grabów (Mazowieckie voivodeship). In 2022-2024, 6 varieties (Goldeneye, Salut, Diament, Puma, Mister, Bursztyn) were evaluated in late June and early July. The presence of viruses was detected by DAS-ELISA immunoassay with specific antibodies. Leaves from 40 randomly selected plants of each variety were collected for analysis.

A small number of BYMV-infected plants were observed in Szepietowo in 2022 and 2023, respectively: 42 and 24 plants. However, in 2024, there was a significant increase in BYMV infection in Szepietowo - 165 infected plants. In Grabów, 71 plants were infected in 2022, 106 in 2023, and 222 in 2024. In Osiny, 64 plants were infected in 2022, 79 in 2023, and only 59 in 2024.

In 2022, CMV was detected in 18 plants in Grabów and in a single plant in Szepietowo. In 2023, CMV infection was found in 56 plants in Szepietowo and 51 in Grabów and in only 5 plants in Osiny. An increase in infection was observed in 2024, when 130 CMV-infected plants were found in Grabów, followed by 59 plants in Osiny and 31 plants in Szepietowo. Among the varieties tested, Goldeneye seems to be the most susceptible to BYMV but not to CMV. The highest virus pressure was observed in 2024 in Grabów.

## **P151 - Twig canker and shoot blight of peach in Italy: the role of *Diaporthe amygdali* and a cultivars tolerance screening**

Epidemiology and ecology of plant diseases

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*Prunus persica* (peach) is a key fruit species in global agriculture, with Spain and Italy as the main producers in the European Union. As peach production faces increasing pressure to minimize the use of synthetic plant protection products, effective management of diseases such as twig canker and shoot blight (TCSB) becomes essential. *Diaporthe amygdali*, the main pathogen responsible for TCSB, causes severe dieback of shoots, flowers, leaves and branches, typically in late winter or early spring. Despite its significant impact, a comprehensive understanding of the biological parameters of the pathogen and the susceptibility of peach cultivars is still lacking. This study aimed to fill these gaps by characterizing *D. amygdali* from symptomatic trees in a productive region of the Emilia Romagna region. Morphological and molecular analyses of the isolates were performed, followed by assessments of mycelial growth and conidia germination at different temperatures to determine optimal growth conditions. Susceptibility tests were performed through *D. amygdali* artificial inoculation on 47 commercial and experimental peach genotypes, followed by symptoms monitoring and lesion measurement. The first results provide valuable insights on the thermal responses of *D. amygdali* for the development of predictive models for more precise and targeted control strategies. The susceptibility findings represent an initial screening of peach genotypes based on their response to TCSB, that could be further investigated to identify possible genetic tolerance traits for breeding programs and development of new disease-tolerant peach varieties.

# **Precision pathology, digitalization and AI tools**

## **O1.4 - Metabarcoding of beech microbial communities: inhibiting the co-amplification of host plant DNA**

Precision pathology, digitalization and AI tools

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Climate change is increasingly affecting forest ecosystems worldwide, posing significant challenges to the health and resilience of key species such as *Fagus sylvatica*, a deciduous tree of great ecological importance. A potential role for microbial endophytes in modulating plant health and resilience has been repeatedly suggested. Here, we aimed to assess the composition of the endophytic community living in beech leaves using amplicon sequencing with Illumina technology targeting the bacterial V3-V4 region on the 16S ribosomal RNA gene and the fungal internal transcribed spacer (ITS) regions 1 and 2. However, a standard approach to metagenomic analysis of these conserved regions can result in significant and unwanted amplification of DNA from the host plant (i.e. the ribosomal ITS region for fungi and chloroplast and mitochondrial DNA for bacteria), thereby compromising the targeted microbial resolution, as confirmed by preliminary sequencing results. This study investigates two alternative approaches to overcome this problem: peptide nucleic acid (PNA) clamps and selective primers. PNA clamps are oligomers with a peptide backbone designed to block PCR amplification of beech ITS and 16S regions. Moreover, for fungi, we chose primers reported in the literature that selectively target only their specific DNA of the ITS1 and ITS2 regions. This study contributes to the definition of an effective protocol, easily extendable to other species, that will allow a more accurate characterisation of the endophytic community and provide new relevant information on their role in plant health in the context of ongoing climate change scenarios.

#### **O4.1 - Multi-point detection and monitoring of *Plasmopara viticola*, *Erysiphe necator* and *Botrytis cinerea***

Precision pathology, digitalization and AI tools

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Internationally, *Plasmopara viticola*, causal agent of grapevine downy mildew (GDM) is known as a destructive obligate oomycete pathogen. GDM predominantly affects Europe and the eastern United States, but economic losses are also seen in other grape-growing regions. *P. viticola* has been identified as a species complex composed of five clades, three of which are economically relevant to grape growers (clades *aestivalis*, *riparia*, and *vinifera*). Previous work has identified cryptic species coinfecting with temporal variation, emphasizing the importance of accurate differentiation. In addition to *P. viticola*, grape disease management is impacted by the presence of two other major pathogens: *Erysiphe necator* (grapevine powdery mildew) and *Botrytis cinerea* (botrytis bunch rot). These pathogens are commonly found in the eastern United States and the management of all three simultaneously is critical for overall grapevine health. Simplified detection of the three pathogens in one assay would benefit monitoring efforts. This research developed two qPCR assay systems to detect three major pathogens and differentiate specific clades of downy mildew and improve monitoring efforts. First, three independent qPCR assays were optimized into a multiplex qPCR assay to detect *P. viticola* (general species) as well as two significant pathogens, *E. necator* and *B. cinerea*. This multiplexed assay was validated on vineyard samples to demonstrate its use. Second, a multiplex qPCR assay and digital PCR assay was designed to differentiate between the three *P. viticola* clades (cl. *aestivalis*, *riparia*, and *vinifera*) and proof of concept experiments using various sample types were performed to test the assay utility. The use of these tools could guide future management strategies as pathogen species and levels fluctuate. The molecular tools developed here hold an important role in answering both basic and applied grapevine downy mildew research questions as researchers comprehensively approach disease management.

## **O4.2 - Genomic insights and molecular detection of *Diaporthe amygdali*: advancing management of TCSB in Italian peach orchards**

Precision pathology, digitalization and AI tools

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In recent years, symptoms of Twig Cankers and Shoot Blight disease (TCSB) have re-emerged in several Italian peach orchards, particularly in key production districts of the Emilia-Romagna region. *Diaporthe amygdali*, recognized as the primary causal agent of TCSB, induces the rapid drying of shoots, flowers, leaves, and branches, often accompanied by resin exudation from cankers observed in late winter or early spring. Given Italy's status as the second-largest peach producer in Europe, ensuring sustainable production and maintaining high fruit quality require a deeper understanding of *D. amygdali* and the development of effective detection and management strategies. As part of the Italian IMPEACHMENT project (PRIN 2022), this study utilized a hybrid whole-genome sequencing approach, combining Illumina short-read and PacBio long-read technologies, to generate the first high-quality genome assembly of *D. amygdali* isolated from peach tree. This genomic resource facilitated the identification of candidate virulence-associated effectors and other pathogenicity-related genes, providing new insights into the infection mechanisms of this emerging pathogen. In parallel, a novel and highly specific molecular detection method was developed, based on a quantitative PCR (qPCR) assay targeting unique genomic markers. This assay enables rapid, sensitive, and reliable detection of *D. amygdali*, supporting early diagnosis and timely disease management in peach orchards. By combining pathogen genomics and molecular diagnostics, this research lays the foundation for more effective TCSB management strategies, ultimately contributing to the sustainability and resilience of peach production in Italy and beyond.

### **O4.3 - Development of novel ddPCR assays for detection and quantification of SDHI resistance in *Botrytis cinerea***

Precision pathology, digitalization and AI tools

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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 fungicides, it is vital that quick and efficient resistance monitoring programs be developed and implemented in the control strategies. Unfortunately, typical approaches of fungicide resistance identification take time and often lack the ability of sensitive detection 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 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. 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"

#### **O4.4 - One more step towards disease predictions: field data update model estimations, estimations drive field data collection**

Precision pathology, digitalization and AI tools

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Are mathematical models truly ready to predict plant epidemics, just after their validation? The answer to this question is generally “no”. The estimation of the initial conditions such as the day zero, the position, and the inoculum of the initial outbreak is one of the main factors that limits the implementation of validated models in decision support systems, to cite some examples. Moreover, the high variability of the biological and environmental conditions, and the presence of factors that are not accounted by models, decrease the reliability of open loop simulations. On the other hand, field data are noisy and expensive, in terms of time and money, and only track the history of the epidemics into the fields. In other fields of science, however, the combination between models and field data led to a substantial improvement of the model predictivity. The main question is if plant pathology could have the same benefits, as well. This study aimed to answering this question by revising the current workflow that considers only epidemiological models. We modified the modelling framework by introducing two new components: a sensing model, that describes the measurement process, and an estimator, that corrects the model estimations based on the field data. This scheme is extremely powerful, as it can either improve the model predictivity or can drive the data collection process. To fully show the potential of the estimators, we consider an application of high interest in plant pathology: the model-driven first detection of infected plants, with optimal allocation of the resources, e.g., limited number of tests or plant that can be inspected.

#### O4.5 - Are spore traps a useful tool for detection and biodiversity analyses in plant pathology?

Precision pathology, digitalization and AI tools

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Spore traps are commonly used for air quality control and studies on aerial dispersal of fungal pathogens. They can catch not only fungal and oomycete spores, but also pollen, smaller insects and a variety of inorganic particles. We designed a series of small-scale studies to estimate Burkard spore trap potential in surveillance of plant diseases in cultivation fields and urban spaces. The presence of DNA extraction inhibitors in the samples proved to be an important bottleneck, however we successfully developed a Chelex 100-based protocol. Samples collected in open fields, close to farm buildings and in the centre of a small city (Lund, Skåne) contained a large variety of biological material, including plant, fungal, oomycete, insect and even metazoan DNA. The composition of the samples varied throughout the plant cultivation season and pathogen spores specific to a particular site, such as *Alternaria solani* in a potato field, were detected. Furthermore, DNA metabarcoding studies showed very little diversity of fungal genera at different potato cultivation sites in two consecutive growing seasons. Comparison with disease progression curves revealed that spores were detected in the collected samples at around the time when first disease symptoms were observed in the plants, therefore too late to be used as an early detection method. To verify Burkard spore trap efficiency in *A. solani* spore entrapment, samples were collected inside a greenhouse chamber and in a small research garden trial where potato plants were artificially inoculated with *A. solani* spores. In both experiments *A. solani* DNA was identified in all of the collected samples, showing 100% detection efficiency in small, confined spaces with large density of the inoculum. Our findings suggest that although Burkard spore traps might not always be appropriate for early detection of plant pathogens they are a useful tool for surveillance and biodiversity studies.

#### **O4.6 - Advancing kauri dieback diagnostics: A multi-dye loop-mediated isothermal amplification assay for *Phytophthora* detection**

Precision pathology, digitalization and AI tools

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*Phytophthora* species are globally significant plant pathogens that cause severe disease outbreaks in forestry and agriculture. *Phytophthora agathidicida* and *Phytophthora cinnamomi* are the primary pathogens driving kauri dieback, posing a severe threat to the survival of New Zealand's iconic native kauri (*Agathis australis*). These trees are deeply valued by New Zealanders and hold cultural significance in Māori traditions. However, the rapid spread of kauri dieback and lack of a cure have made kauri a threatened species, leading to forest closures for preservation. Early and accessible detection is crucial for effective disease management, yet current molecular diagnostics lack portability and robustness. Loop-mediated isothermal amplification (LAMP) is a promising molecular tool for pathogen diagnostics due to its speed, simplicity, and suitability for in-field use.

This study presents an optimized LAMP assay with separately designed primer sets for *Phytophthora agathidicida* and *Phytophthora cinnamomi*, improving reaction efficiency and detection speed compared to the previous methods. The newly designed species-specific primers target conserved genomic regions, resulting in rapid and strong signal amplification within 15–25 minutes at 65°C, outperforming existing protocols. The significant improvement of this assay is the multi-dye compatibility, allowing detection through eight visualization methods, including pH-sensitive indicators, colourimetric dyes, and fluorescence-based probes. Unlike previous LAMP assays restricted to a single detection method, this assay offers greater versatility for various diagnostic settings, ensuring effective use in both well-equipped laboratories and low-resource field environments.

This optimized LAMP assay offers a valuable tool for kauri dieback management and contributes to improvements in *Phytophthora* diagnostics by enhancing reaction efficiency, detection sensitivity, and multi-dye compatibility. Future work will focus on integrating this assay with microfluidic platforms to enhance field usability and enable high-throughput screening. These advancements offer valuable potential for forestry and agriculture by enabling rapid pathogen detection and improved disease management in both laboratory and field settings.

## **P27 - Machine learning analysis to reveal new methods of gushing prediction**

Precision pathology, digitalization and AI tools

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Gushing, or over-foaming, is a phenomenon that causes severe economic losses in beer production. There is a lack of knowledge in the molecular mechanisms underlying this phenomenon, but it has been proposed that hydrophobins synthesized by fungal species, such as *Fusarium* spp., are the causal agents of this phenomenon. Current prediction tools of gushing include *Fusarium* detection based on the seed colour. It seems, however, that these analyses may lead to over-detection, with too many seed lots being downgraded consequently. To manage the problems of gushing, better information about causes and predictors are urgently needed.

We performed a study of Swedish barley grains, analyzing the fungal communities and selected characteristics of the seed such as water content, presence of kernels with pink coloration, kernel weight and ergosterol content. We developed a Random Forest algorithm to study the value as predictors of the different studied variables.

The results show a complex landscape where not only *Fusarium* spp., indeed also *Fusarium* spp. interactions with other fungal species, could lead to gushing. Moreover, the results suggest that additional predictors could be used for better precision in the selection of seed lots suitable for beer production.

### **P36 - Enhancing oilseed rape growth assessment: NDVI and NDYI analysis from UAV-based RGB Imagery**

Precision pathology, digitalization and AI tools

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This study evaluated the potential of the Normalized Difference Vegetation Index (NDVI) and the Normalized Difference Yellowness Index (NDYI), derived from Red-Green-Blue (RGB) imaging, to monitor the growth status of oilseed rape (*Brassica napus*). The collected index values were further analysed for their correlations with yield. Field trials were conducted over two years, incorporating different seed densities and nitrogen rates. A multispectral camera mounted on an unmanned aerial vehicle captured imagery. NDVI and NDYI values were calculated using reflectance data from the RGB and NIR bands in a reconstructed and segmented orthomosaic. Throughout both growing seasons, NDVI increased significantly from the seedling stage to early winter, followed by a slight decline post-winter. It then rose again, reaching a peak before flowering, before dropping sharply during the flowering stages. NDVI reached its highest peak at full pod development before declining steeply until maturity. In contrast, NDYI plateaued around flowering, decreased during pod development, showed a brief increase at the end of pod development, and then declined until maturity. NDVI measured at full leaf development before winter and at full pod development showed the strongest correlations with final yield. A significant correlation was found between NDYI at full flowering and final yield.

## **P44 - Decision support system for the diagnosis of post-harvest diseases of apple: an innovative teaching tool**

Precision pathology, digitalization and AI tools

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We developed a decision support system named ‘DSSApple’ to diagnose post-harvest diseases of apple fruit (*Malus × domestica*) based on observed macroscopic symptoms. The focus was put on ten post-harvest diseases of apple commonly occurring in South Tyrol (northern Italy), namely Alternaria fruit spot, Alternaria rot, Bitter rot, Black rot, Blue mold rot, Bull’s eye rot, Fusarium rot, Grey mold rot, Mucor rot and Side rot. The interactive, web-based system can be used by expert and non-expert users and consists of two streams. (I) The image-based stream is built on a database of pictures showing disease symptoms on various apple cultivars at different stages of development. The users interact with this stream by selecting pictures that are most similar to the target disease to be determined. (II) The expert-based stream asks consecutive questions about the observed appearance of external lesions and internal rot symptoms on a target apple. The knowledge base for each disease includes characters of each symptom. These were accurately described based on information provided in specialist literature and on the observation of symptoms on apple fruit, from which the exact diseases were determined by microbiological and/or molecular genetic assays. The knowledge base was modeled in a Bayesian network to develop the expert-based reasoning mechanism, and the system was complemented by a visual encyclopedia detailing the possible expression of each symptom. ‘DSSApple’ has been used in practical teaching of undergraduate classes of plant pathology to introduce students to post-harvest pathology but also to learn how to proceed in symptom-based diagnosis. It can be integrated in laboratory activities with naturally infected or inoculated apple fruit and combined with other experiments to introduce the concept of Koch's Postulates. The use of images makes the system also suitable to develop practical activities in circumstances where a laboratory is not accessible.

## **P45 - Differentiation and early detection of post-harvest apple pathogens using hyperspectral imaging**

Precision pathology, digitalization and AI tools

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Fungal post-harvest pathogens can lead to significant economic losses in apple production. *Colletotrichum godetiae* and *Penicillium expansum* causing bitter rot and blue mold of apple, respectively, are two major representatives of post-harvest pathogens. They are entirely distinct in their infection biology, the former being a latent and the latter a wound pathogen. The aim of the study was to investigate the growth dynamics and spectral differentiation of these two pathogens on apple fruit. Apple fruit of the cultivars ‘Granny Smith’ and ‘Cripps Red’ Joya® were wound-inoculated in the upper hemisphere at four points distributed at 0°, 90°, 180°, and 270° with 15 µL of spore suspension (10<sup>6</sup> spores/mL) and incubated in a moist chamber at room temperature to provide favorable conditions for fungal growth. Each experimental variant as well as the mock controls included 24 apple fruit. Pathogen development at each apple and inoculation point was assessed at 3, 5, 7, and 10 days post-inoculation (dpi) for *P. expansum* and 3, 5, 7, 10, 14, and 21 dpi for *C. godetiae*, using an RGB camera (24 MPix, three channels, spectral range of 400 to 700 nm, ca. 200 nm of spectral resolution/channel), a hyperspectral imager (1 MPix, 16 channels, spectral range from 506 to 902 nm, and 10 nm of spectral resolution/channel), and a spectroradiometer (spectral signature scanner, 1024 channels, spectral range of 350 to 2500 nm, and 1 nm of spectral resolution/channel). The image and spectral signature analysis is expected to provide evidence of the pathogen progression over time, scale and spectrum. These findings shall demonstrate the potential of hyperspectral imaging as a tool for early detection and discrimination of fungal pathogens, contributing to improved post-harvest disease management strategies.

## **P54 - *Aphanomyces euteiches* in Swedish pea production - soil analysis and impact of legume cover crops in crop rotation**

Precision pathology, digitalization and AI tools

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*Aphanomyces* root rot (ARR), caused by the oomycete *Aphanomyces euteiches* is a devastating disease of pea and other legumes. The disease can only be managed by avoiding cultivating pea and other host crops in infested fields. In Sweden, ARR prediction is mainly based on soil analysis using a commercial bioassay. The aim of this study was to provide pea growers with a reliable PCR-based soil analysis for precise prediction of ARR. In addition, we investigated susceptibility of legume cover crops and assessed their impact on ARR infection in successive pea plants.

Occurrence of *A. euteiches* was assessed in 24 soil samples by evaluating disease severity index (DSI) in susceptible pea cv. Linnea and quantification of gene copies in soil. Susceptibility of 14 legume cover crops was examined by artificial inoculation in vermiculite and the influence of selected species on ARR infection was investigated in naturally infested soil.

The number of *A. euteiches* gene copies varied across the tested soil samples and fields with high as well as low ARR risk could be identified. A significant correlation was found in one bioassay between DSI and gene copies g<sup>-1</sup> soil. Visual assessment and PCR analysis revealed varying susceptibility among the cover crops. Susceptible cover crops significantly increased DSI in the subsequent pea plants as well as number of *A. euteiches* gene copies in soil. These findings highlight the importance of molecular soil analysis in disease prediction and emphasize the need to avoid certain legume species in pea crop rotations.

## **P56 - Evidence-based disease control methods in potato production - a systematic map**

Precision pathology, digitalization and AI tools

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Potato is the world's third most important crop for food consumption and contributes to food security in many developing countries. However, potato is susceptible to a wide range of pathogens that cause diseases that lead to quality and yield losses. One of the main constraints to sustainable potato production is the dependence on the use of pesticides for disease control. Therefore, the aim of this systematic map is to provide a global overview of evidence-based disease control measures available, and to identify disease control practices that could enable more sustainable potato production. Searches were conducted in academic bibliographic databases and online sources of grey literature to identify all relevant literature published between 2000 and 2022. The screening process, which followed a protocol with eligibility criteria, was conducted at three levels: title, abstract, and full text. The systematic map is presented as a searchable database where the volume and main characteristics of the identified studies are described. According to preliminary results, 1245 eligible articles were included in the systematic map. *Phytophthora infestans*, a major constraint to global potato production, was the most studied disease accounting for 30% of the studies. Cultivar resistance, pesticides and biological control agents (BCA) were the most studied disease control methods. From 2017 and onwards, the number of studies using BCA increased dramatically, reaching higher numbers than pesticide control. In addition, 43 other disease control methods were studied and most of them focused on sustainable intervention. This demonstrates the growing interest in identifying alternative control strategies for a more sustainable potato production. The results of this systematic map provide valuable information and support for researchers and stakeholders, in particular authorities and advisory organizations, in providing scientifically relevant crop protection strategies for farmers.

## **P59 - Scientific evidence of sustainable disease protection strategies for oilseed rape in Sweden: a systematic map**

Precision pathology, digitalization and AI tools

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Oilseed rape (OSR; *Brassica napus* L.) is a highly valued crop for food, feed and industrial use, and during recent decades, its area of production and profitability have increased. Concurrently, several diseases negatively impact OSR production. The aim of the systematic map was to provide an overview of plant protection measures to be directly implemented in crop production management for OSR production and to identify knowledge gaps and areas where more research is needed. The search strategy was designed to cover available scientific evidence for OSR disease management in climate relevant for Scandinavian crop production. The systematic map provides an overview of research progress throughout the time chosen, and it identifies knowledge gaps regarding important diseases where only a few studies have been published, for example, diseases caused by viruses. Scientific studies on plant disease protection measures for OSR based on field trials where the results are intended to be directly implemented in crop production management are compiled and presented in a user-friendly set up. The findings highlight that research on disease control measures often is a need-driven processes, which is reflected in that the most important diseases have been primarily studied, whereas the less economically important diseases are under-studied. The map can be used as an important tool for field-based advisors to identify and provide scientifically based plant protection strategies for farmers.

## **P65 - Different SDHI fungicide treatments and the frequency of resistance markers in *Erysiphe necator* populations**

Precision pathology, digitalization and AI tools

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Succinate dehydrogenase inhibitor (SDHI) fungicides are widely used to control grapevine powdery mildew (*Erysiphe necator*). However, resistance to SDHIs has emerged due to mutations in genes (*sdhB*, *sdhC* and *sdhD*) encoding the subunits of the succinate dehydrogenase enzyme.

Understanding the distribution of resistance markers and their association with fungicide treatments is crucial for optimizing disease management strategies.

This study investigated mutations in the *sdhC* and *sdhD* genes and their correlation with SDHI applications. Powdery mildew-infected grapevine leaf samples were collected from untreated control plots and plots treated with boscalid, fluopyram, or fluxapyroxad in two Hungarian wine regions throughout 2022–2023. DNA was extracted from over 170 field samples. First, Sanger sequencing of the coding regions of *sdhC* and *sdhD* was performed to identify occurring markers. Then, two resistance markers were specifically screened: one using Sanger sequencing of a fragment of the *sdhD* gene and the other using a rapid, digestion-based diagnostic assay for a marker in the *sdhC* gene.

A mutation in *sdhD*, resulting in an amino acid substitution (*sdhD*-I71F), was detected in 22.1% of samples. This marker has not been previously reported in Europe, and it is associated with reduced SDHI sensitivity in other fungal pathogens according to published data. Its occurrence significantly correlated with fluxapyroxad treatment but not with boscalid or fluopyram.

Additionally, a mutation in *sdhC* (*sdhC*-G169D), also linked to SDHI resistance in previous publications, was detected in 65.5% of samples. It was more prevalent in fluxapyroxad-treated plots and less frequent in fluopyram-treated areas, while boscalid treatment had no effect on its distribution. Double mutants (*sdhD*-I71F + *sdhC*-G169D) were observed in 9.4% of samples and were significantly more frequent in fluxapyroxad-treated plots.

These findings provide critical insights for improving fungicide resistance management in vineyards.

## **P99 – Addressing biohazards in agriculture with a focus on crop production**

Precision pathology, digitalization and AI tools

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Pathogens constitute biohazards for the primary production of crops and livestock, causing major economic damage to farmers, the agricultural industry, society, and international trade. Most introductions of pathogens have natural causes, but they may also be accidental or intentional. Agroterrorism is the intentional introduction of plant or animal pathogens into agricultural production systems with the intention to cause socioeconomic harm and generate public fear. The risk of acts of agroterrorism is not negligible. The rapid advancements in emerging technologies arise new concerns about threats, and food supply chains are challenged with climate change and the war in Ukraine. When the food supply chains become more fragile there is an increased need for risk and vulnerability analyses. The European countries have strong agricultural protection systems in place, and they are coupled with efficient diagnostic networks. There are still several areas where improvements could be made to enhance the level of preparedness for both natural and intentional spread of pathogens.

In this study, we applied FORSA, an analytical framework for risk and vulnerability analysis, to review how to prepare for and mitigate the possible effects of natural, accidental or intentional biohazards in agricultural production. The overall goal of a vulnerability analysis is to understand the nature of the risks, strengthen crisis preparedness, reduce vulnerabilities and increase the ability to manage crises. Analysing the effects of a biohazard event involves multiple scientific disciplines. A comprehensive analysis of biohazards therefore requires a systems approach to cover all complexities. From the analysis, it can be concluded that the preparedness and ability to manage events would be enhanced by strengthened biosecurity on farms, increased surveillance and laboratory capacity, improved inter-agency communication and resource allocation. While the focus of this review is on Europe, the insights gained have worldwide applications.

## **P100 - AlmondProtect: from diagnosis to prevention**

Precision pathology, digitalization and AI tools

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Almond tree (*Prunus dulcis*) is a common crop found in the Mediterranean basin. In Portugal, particularly in Alentejo Region, almond production plays a major economic, cultural and social role. These are affected by several pathogenic agents resulting in devastating economic losses. Also, other pathogenic microorganisms are causing new emerging diseases which might culminate on serious yield reduction and an increase in the use of phytopharmaceutical products.

The AlmondProtect project is an integrated sustainable approach that aims to reduce the disease incidence level of almond orchards in several production stages, from nurseries to commercial post-harvesting, by early detection and disease prediction.

For that purpose, four main tasks were settled. The first step is the identification and characterization of microbial pathogenic agents in symptomatic almond trees and fruits, through metagenomic studies, Near-infrared spectroscopy (NIR) and sensors.

Furthermore, the development of diagnostic and monitoring methodologies of disease geographical distribution and progression, using innovative technologies for all production stages and prior to symptomatology is being optimized. Hence, procedures relying in techniques using Doctor Vida, a biomarker-based portable device that uses Loop mediated isothermal amplification (LAMP) technology and complementary molecular tests based on quantitative PCR (qPCR) are being performed.

Ultimately, the creation of disease prediction models through spores' collection and analysis, evaluation of inoculum prevalence and monitoring, is also being operated, recurring to sequencing using MinION (Oxford Nanopore).

This collaborative project will be a major contribution for almond producers, due to its sustainable character which uses early preventive diagnosis, resulting in a more competitive and adaptative agriculture.

**P101 - A model pathosystem to study the incidence and severity of black dot and silver scurf diseases in potatoes.**

Precision pathology, digitalization and AI tools

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Silver scurf (*Helminthosporium solani*) and black dot (*Colletotrichum coccodes*) are economically significant tuber blemish diseases affecting potato quality and marketability. Existing methods for studying these diseases rely on minituber assays, root dipping, tuber coating with conidial suspensions, or direct soil inoculation. In this study, we developed an improved pathosystem that enables the *in-vitro* and field assessment of silver scurf and black dot disease incidence and severity. Tuber disc inoculation in 12-well plates provided a controlled *in-vitro* environment for evaluating the disease severity. In field, an efficient pathosystem was developed using tuber coated conidial suspensions as tuber inoculum and pathogen grown on vermiculite as a soil inoculum. Both methods were successful in initiating the infection and provided a platform to study disease progression. This pathosystem offers a potential model for studying disease management strategies, including biocontrol efficacy, tolerance screening, and fungicide resistance assessments against black dot and silver scurf diseases in both lab and field conditions. Furthermore, it can also be adapted to study other tuber blemish diseases in potatoes.

**Keywords:** *Helminthosporium*, *Colletotrichum*, tuber, inoculum.

## **P102 - CResWheat: Pre-breeding climate-resilient spring wheat for the Nordic region**

Precision pathology, digitalization and AI tools

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The CResWheat project advances climate-resilient spring wheat breeding by identifying genetic resources for disease resistance, drought tolerance, and other adaptive traits suited to the Nordic region. This research is essential for sustainable wheat production, addressing challenges such as short growing seasons, increasing pest and pathogen pressure, and more frequent extreme weather events.

In Phase I (2021-2023), 24 field trials across seven locations in four Nordic countries assessed over 20 traits in European and exotic elite spring wheat. Donors were identified for resistance to major wheat diseases, including yellow rust, leaf rust, stem rust, *Septoria tritici* blotch, powdery mildew, *Stagonospora nodorum* blotch, and tan spot, as well as key agronomic traits such as resistance to pre-harvest sprouting. Nearly 70 genetic markers associated with these traits were identified, supporting marker-assisted selection.

In Phase II (2024-2026), strategic crosses are being developed to introduce resistance to targeted diseases and pre-harvest sprouting into elite germplasm. These populations will be further evaluated in multi-environment trials across the Nordic region and Estonia. Additionally, a new collection of old landraces and cultivars is being assessed for resistance to gout fly (*Chlorops pumilionis*), a pest of increasing significance in Nordic spring wheat.

CResWheat fosters collaboration by sharing knowledge, materials, and field locations across diverse environments. The project also strengthens capacity building by training PhD students and postdocs, ensuring a new generation of experts to address future challenges in wheat breeding.

## **P103 - Detection of soilborne fungal and oomycete plant pathogens through microfluidic SoilChips and deep learning**

Precision pathology, digitalization and AI tools

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### **Background and aims**

Around 40% of our global food production is lost due to pests and pathogens. Current pathogen detection methods mainly rely on visual inspection of the crop, leading to late detection and ineffective pathogen management practices. More sensitive methods, such as biomolecular methods, are seldom financially viable for average farms. This proof-of-concept study acts as an investigation of the possibility of using microfluidic technology that mimics soil structure while maintaining visual access (“SoilChips”) to locate and quantify soil-borne pathogens using deep learning-based image analysis of hyphal morphology.

### **Methods**

A selection of eight suitable soil-borne pathogens causing infection in sugar beet, rape seed and wheat (*Verticillium*, *Fusarium*, *Aphanomyces*, *Phytophthora*, *Pythium*, *Rhizoctonia*) were grown into the soilchips under different nutrient conditions. The chips were examined daily and documented under the microscope, capturing hyphal morphology and architecture in a confined environment closely mimicking soil. An image database was created and used for training a convolutional neural network model for image classification, as well as a separate instance segmentation model. Both models were trained using YOLOv11. The classification model includes all images whereas the segmentation model includes around 10% of the database, with hyphal structures manually labelled.

### **Results**

While models are still being developed further, the two models have been trained with of 90%+ accuracy. The two models can both distinguish the eight different species from each other.

### **Conclusions**

We have successfully shown that these eight pathogens can be distinguished from each other based on morphology using the SoilChips platform, microscopy and neural networks. The classification model has been able to distinguish between two related species (*V. dahliae* and *V. longisporum*) which are difficult to distinguish for experts.

Further work lies in improving selectivity of chips, sample taking in field, expanding the database and finetuning of models.

## **P104 - Evaluating UAV spray applications for grapevine downy mildew control as sustainable solution in steep slope vineyards**

Precision pathology, digitalization and AI tools

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The application of plant protection products (PPPs) in vineyards is often limited by landscape features. In Italy, many vineyards are in hilly and mountainous areas, and growers traditionally apply PPPs manually, increasing risks for both environmental and human health. To facilitate disease control in critical areas, such as steep slope vineyards, the European council proposed (2022/0196/COD) allowing spray application through unmanned aerial vehicle (UAV). This study compared a UAV spray system, fixed to the ground, with ground spraying to control grapevine downy mildew, caused by *Plasmopara viticola*. Two fungicides, a contact copper-based and a systemic metalaxyl-m-based, were applied with both spray approaches in post-flowering and bunch-closure. At 1, 5, and 7 days after treatment (DAT), treated leaves were collected from vineyard in the higher (H) and lower (L) canopy part to i) evaluate disease control efficacy using artificial inoculation with *P. viticola* sporangia and ii) determine PPPs residues by QuEChERS extraction and liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS) analysis.

Overall, post-flowering applications showed higher efficacy regardless of products, approaches, and DAT. At post-flowering, metalaxyl-m efficacy was not influenced by the considered factors, while copper efficacy was significantly affected by the spray approach ( $p < 0.001$ ) and DAT ( $p = 0.011$ ); with the highest efficacy at DAT 1 (90.2%) for ground application. At bunch-closure, metalaxyl-m efficacy differed significantly between spray systems and DAT ( $p = 0.008$  and  $p < 0.001$ ), with ground application reporting the highest efficacy at DAT 1 (94,8%). Differences in copper efficacy were only found in H height at DAT 7 (62.6% and 25.9% from UAV and ground, respectively). Residues analysis confirmed the efficacy trends for both compounds.

To better understand the efficacy-residue correlation, further studies should consider the minimum biological effective dose and optimize of UAV sprays through customized nozzles to improve both disease control and canopy coverage.

## **P105 - Green nudges for sustainable forestry and agricultural practices post 2027 - ForestAgriGreenNudge**

Precision pathology, digitalization and AI tools

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Current forestry and agri-food system trajectories are leading to biodiversity loss and land ecosystem degradation failing to provide food security, nutrition, and sustainable healthy livelihoods to many. Solving these environmental challenges relies on a green transition requiring profound changes in bioeconomy policies and initiatives towards more sustainable practices in agriculture and forestry. Most farmers and foresters seem to be aware of the urgency of increasing efforts in this area.

The European Green Deal with its package of policy initiatives (e.g. new EU Forest Strategy, CAP 2023-27), offering a key tool to achieve a sustainable transformation, placing farmers and foresters at the core of its actions. But do people act according to what they say? Do they behave in accordance with their good intentions?

At ForestAgriGreenNudge, we believe that small changes can lead to big impacts. Using insights from behavioral science, our project aims to guide decision-makers towards more sustainable practices in farming and forestry through the use of “Green Nudge”—subtle, positive interventions that reshape decision-making environments.

Our approach begins with a comprehensive review of existing initiatives using Green Nudges, followed by an in-depth assessment of their effectiveness and ethical integrity. We then field-test these nudges to identify the most successful ones, which are further refined and tailored based on feedback from stakeholders along the entire value chain.

The culmination of our efforts is the iNUDGE Academy, an online tool designed to support policymakers, farmers, foresters, retailers, and wholesalers. This resource provides practical guidelines, implementation strategies, and risk assessments for integrating nudges into everyday practices, helping drive the shift towards sustainability.

## **P106 - Harnessing unused genetic variation and fast screening of loose smut (*Ustilago nuda*) in winter barley**

Precision pathology, digitalization and AI tools

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Loose smut of barley caused by the fungal pathogen *Ustilago nuda* (Rostr.) concerns worldwide organic and low-input barley cultivation. Breeding resistant varieties is the most effective and the most environmentally friendly approach of plant protection. Current commercial barley varieties lack resistance to loose smut, resulting in increasing occurrences in practical farming in Europe. The majority of the resistance donors have been identified in the spring barley genotypes. Among the 15 resistance donors (Un1-15) described, only a few genotypes carrying Un8, Un6, and Un3 are utilized in breeding programs.

In the Flugbrand-Resist project, 470 winter barley genotypes from diverse origins, alongside several elite lines, are artificially inoculated in greenhouse trials. All phenotypically evaluated barley lines are genotyped using a recent 20 k SNP array to identify QTLs and closely linked marker-trait association in genome-wide association studies (GWAS) to discover new sources of loose smut resistance.

Conventional methods for screening loose smut are time-consuming and labour-intensive, requiring approximately 600 days over two growth cycles. To address this, we developed a protocol implementing speed breeding techniques to accelerate disease screening. Our findings showed a significant differentiation between the two approaches, reducing the time needed by around 250 days and saving at least half the time for phenotyping. Moreover, preliminary results indicate that combining speed vernalization with our inoculation protocol can further shorten the screening process to around 180 days. This approach promises to fast-track the development of loose smut-resistant winter barley varieties. The results of the project will support breeding for organic and conventional farming and facilitate environment-friendly disease control to meet the criteria of the European Green Deal and the EU Farm to Fork strategy.

## **P107 - How good are farmers' weather stations for modelling the risk of potato late blight?**

Precision pathology, digitalization and AI tools

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One objective of phytopathology research is to model the development of plant pathogens to help farmers make informed decisions about crop protection. Farmers may install their own weather stations in the field to provide more local input for these models. However, it is unclear whether the quality of data from farmers' weather stations (FWS) is sufficient given the sensitivity of certain agrometeorological models.

The objective of this study is to assess the uncertainty of FWS based on a 3-year field trial and to estimate its impact on potato late blight (PLB) modelling.

A Guntz & Divoux model (GD) will be used to assess the risk of PLB using hourly weather data. The outputs of interest (e.g. the date of the third PLB generation) will be selected and an acceptable error will be assigned to them.

To investigate the relevance of FWS, a straightforward approach involves running the GD model over three years of recorded weather data and comparing the simulations of FWS against a reference weather station. However, this approach is of limited interest due to the short historical record: only three seasons are available for analysis.

In this study, we propose modelling the temporal structure of errors on FWS data to generate a larger dataset (e.g. 1000 samples) that simulates FWS behaviour. Therefore, a more statistically robust approach based on uncertainty analysis will be conducted. The generated FWS data will be used to feed the GD model. The distributions of the outputs of interest will be analysed in relation to the acceptable errors defined earlier.

This analysis will determine whether the quality of FWS data is sufficient for use in PLB monitoring. The detailed methods and results will be presented at the congress.

## **P108 - Identification of autoecious and heteroecious *Cronartium pini* based on digital PCR and spore germination**

Precision pathology, digitalization and AI tools

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*Cronartium pini*, the causal agent of Scots pine blister rust, has two life cycle forms. The autoecious form (known as *Peridermium pini*) infects only pine. The heteroecious form (known as *C. flaccidum*) is macrocyclic and infects pine as the aecial host and many herbaceous plants as telial hosts. Both forms induce the same symptoms in pine.

Distinguishing these two forms has been a challenge. The two forms have aeciospores that are morphologically similar, and they cannot be distinguished by genes that are commonly used for phylogeny such as ITS. While inoculation tests and homozygosity/heterozygosity determined through microsatellite genotyping can distinguish them, these methods are time-consuming and labor-intensive.

Ongoing studies revealed genome-wide homozygosity in autoecious and heterozygosity in heteroecious strains. We developed a rapid and reliable identification protocol using nanoplate and digital PCR (dPCR) with three molecular markers: the housekeeping gene *TEF1*, consistently present in two copies in all dikaryotic *C. pini* genomes, and the mating locus *MAT A1*—which includes two specific alleles of *HD1* and *HD2* present as two identical copies in autoecious *C. pini* genomes. To validate this protocol, we compared the results from peony inoculation, microsatellite genotyping, and dPCR analysis across 20 autoecious and heteroecious *C. pini* strains. Furthermore, we tested the dPCR protocol on previously identified strains, confirming its efficiency and reliability in distinguishing between the two forms.

We also compared the morphology and nuclear behavior of autoecious and heteroecious *C. pini* aeciospores during germination. After 3 to 7 days, heteroecious aeciospores produce long single germination tubes with complex branching, lacking septa, and containing two nuclei. In contrast, autoecious aeciospores developed germination tubes with septa and one to four buds, which may penetrate pine needles as basidiospores, each containing a single nucleus.

This study provides a robust and efficient tool for research and forest management efforts.

## **P109 - In field validation of real-time sporangia-assessment of *Phytophthora infestans* with SwisensPoleno air-flow cytometer**

Precision pathology, digitalization and AI tools

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Potato late blight, caused by the oomycete *Phytophthora infestans*, is the most destructive disease of potatoes with an estimated annual economic damage in the European Union of almost € 900 million. The disease is polycyclic and disperses large numbers of asexual sporangia which are carried through the air and can, under favourable weather conditions, infect both neighbouring and distant potato fields. The infection can drastically reduce potato yield and quality. Therefore, making precise and timely fungicide applications is crucial for efficient and environmentally sound crop protection. At present, decision support systems (DSS) are mainly based on weather data (i.e., temperature, relative humidity, rainfall) for infection prediction but lack of reliable information on occurrence and abundance of sporangia.

The aim of real-time measurement of plant pathogen concentrations is to improve infection risk assessment and modeling, enabling even more precise plant protection measures with the existing DSSs. The SwisensPoleno measurement system monitors bioaerosols automatically in real-time (e.g. spores, pollen) using the latest optical particle measurement methods (holography and fluorescence) and uses an artificial intelligence based classification software [1].

In order to determine the real-time *P. infestans* sporangia concentration in the air, a new classifier for the SwisensPoleno was developed and validated against manual measurements with a Hirst spores trap. The monitoring was conducted in potato fields at two different locations.

The initial validation results with the new algorithm are Spearman correlation of 0.68, Kendall's Tau of 0.51 and Pearson's R of 0.82.

The focus of the study was on *P. infestans*, however, spores of other fungal pathogens of potato, such as *Alternaria solani* and *A. alternata*, can also be detected and quantified with the SwisensPoleno. Further studies across other locations are needed to validate the reliability of the algorithm.

[1] E. Sauvageat et al., 'Real-time pollen monitoring using digital holography', doi: 10.5194/amt-13-1539-2020.

## **P110 - Multispectral detection of early stress signals in wounded scots pine seedlings**

Precision pathology, digitalization and AI tools

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The economic importance of Scots pine (*Pinus sylvestris* L.) trees in Sweden underscores the need for effective monitoring of their health in nurseries and forests. Pines, regardless of age and environment, are subjected to stress from various abiotic and biotic factors. Early detection of stress is crucial for timely interventions to mitigate damage and economic losses, especially concerning pests and pathogens. Stress from these factors triggers metabolic changes in plants that are often invisible to the human eye in the early stages. Multispectral sensors, which capture images across multiple wavelengths (RGB, NIR), offer a promising solution for the early detection of plant stress by extending imaging beyond the visible spectrum. This technology can significantly enhance the monitoring of plant health and resilience. In our study, we evaluated the use of a portable, multispectral 3D laser scanner, Phenospex PlantEye®, to detect early signs of damage in Scots pine seedlings. We utilized one-year-old potted Scots pine seedlings obtained from Våge (N-SWE) and Gotthardsberg B (S-SWE), representing two provenances. The seedlings were wounded at two intensities (high and low) using a scalpel to simulate pine weevil feeding, while control plants remained intact. A total of 48 plants were scanned, and 480 scans were taken over two weeks: hourly for the first six hours (0-1-2-3-4-5-6 hours post-wounding), then at 24 hours, 168 hours, and 336 hours following the wounding. Based on the collected data, we calculated vegetation indices such as the Normalized Difference Vegetation Index (NDVI), Normalized Pigment Chlorophyll Index (NPCl), Plant Senescence Reflectance Index (PSRI), Green Leaf Index (GLI), and the Hue Index (HUE) to assess the stress signals in the treated plants. Our findings discuss the potential of developing practical monitoring applications for nurseries using this technology.

## **P111 - Powdery mildew on cucurbitaceous crops: Pathogen identification and detection of an SDHI fungicide resistance marker**

Precision pathology, digitalization and AI tools

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Powdery mildew is one of the most frequently occurring diseases of cucurbit crops. We collected powdery mildew-infected leaves from cucurbit plants in agricultural fields in western Hungary to identify the pathogenic fungus and to assess the presence of markers associated with resistance to succinate dehydrogenase inhibitor (SDHI) fungicides.

We sampled over thirty plots, collecting samples from various species and cultivars of cucurbits, watermelon, and cucumber. To identify the fungi, we sequenced the internal transcribed spacer region of the ribosomal DNA and developed a restriction digestion-based method for the rapid differentiation of the two identified species.

Fungi on approximately two-thirds of our samples belonged to the *Golovinomyces orontii* s. lat. species complex, while in other samples, we identified *Podosphaera xanthii*.

From the *P. xanthii* samples, we sequenced the *sdhC* and *sdhD* genes, which encode two subunits of succinate dehydrogenase and are known to carry markers of resistance to SDHI fungicides. All determined *sdhD* sequences were identical to wild-type reference sequences. However, in the coding region of the *sdhC* gene, we identified a G-to-A nucleotide substitution at position 612 (G612A), which results in an amino acid substitution, denoted G151R.

G151R has been detected in *P. xanthii* samples from Japan and Spain and is associated with resistance to several SDHIs. Our study is the first to report this mutation in Hungary. Our data highlight the importance of local monitoring of fungicide resistance markers and have the potential to support disease management in cucurbit production.

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## P112 - Quantifying the sensitivity and specificity of visual inspection in plant health

Precision pathology, digitalization and AI tools

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Plant pests and diseases are occurring at increasing frequency, resulting in impacts such as crop yield losses and disruption to ecosystem services. Early detection of such pests and diseases is critical to increase the likelihood of eradication and reduce the costs of disease management. Visual inspection remains a tenet of plant health surveillance, yet unlike laboratory diagnostics, the sensitivity (probability of correctly identifying a disease positive) and specificity (probability of correctly identifying a disease negative) are not routinely quantified. Knowledge of these parameters is required for basic interpretation of survey data and calculation of surveillance metrics such as probability of pest/disease absence given no detection, and likely true prevalence or distribution of a pest/disease given a detection. In summer 2022, 23 volunteers surveyed up to 175 oak trees (*Quercus* spp.) across two sites for three different symptoms of acute oak decline (AOD). The sensitivity and specificity of volunteers at detecting symptoms was then calculated using a dataset from an expert that has monitored these trees for over a decade. Expanding on these findings, a larger survey in 2024 involving 164 individuals surveying up to 80 trees each was then conducted to examine the impact of training and experience. For applications to wider plant health, the utility of a workflow using Bayesian modelling to estimate the sensitivity and specificity of visual inspection in the absence of a rarely available ‘gold-standard’ validation dataset was examined using field-realistic simulated datasets based on the AOD surveys. Our results highlight large variation in the sensitivity and specificity between individuals and symptoms, with symptom frequency, training and experience all impacting sensitivity. The workflow demonstrated that sensitivity and specificity can be estimated in the absence of a validation dataset, which provides a powerful tool for interpretation of survey data and optimisation of risk-based surveillance in plant health.

## **P113 - Rethinking plant disease detection: comparison of soil- and seedborne *Tilletia* transmission pathways**

Precision pathology, digitalization and AI tools

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Synthetic seed coatings protect plants from diseases that can be transmitted both on seed and through soil. However, agricultural policies promote a reduction of such plant protection products, creating the need for new preventive approaches. While seed health tests on untreated seed effectively protect them from seedborne pathogens, the seeds are still potentially vulnerable to unchecked soilborne inoculum in fields. This limitation is relevant for pathogens, such as *Tilletia caries* (common bunt) and *Tilletia controversa* (dwarf bunt), whose teliospores can survive in soil for 3 and 10 years, respectively. In wheat, *T. caries* is typically considered a seedborne pathogen, while *T. controversa* is thought to be primarily soilborne. The scientific underpinnings of these conceptions, however, are unclear. To better understand the effect of seed versus soil infection on the development of bunted ears, we conducted pot experiments with wheat exposed to each transmission pathway. In parallel, for both *Tilletia* species, we developed a specific qPCR detection protocol, which we applied to soil DNA extracted with different methods from our experiments. Initial results of our *T. controversa* protocol reveal that its primers are species-specific, but the extraction method affects DNA yield and quantification. A preliminary pot trial showed that *T. caries* teliospores added to soil caused significantly more diseased plants than when added to seed. An on-going pot experiment on wheat inoculated with *T. caries* or *T. controversa* through seed or soil will provide more information about their transmission pathways and our protocol's reproducibility. Our first findings highlight the need for detection tools that address seed and soil infections and distinguish between these *Tilletia* species. Ultimately, this work will provide insights into *T. caries* and *T. controversa* transmission and knowledge about their presence in soil. Our soil testing protocol, combined with seed health tests, can enhance preventive strategies against wheat pathogens.

## **P114 - Towards the development of guidelines for farm- and crop-specific measures to reduce the impact of pesticides**

Precision pathology, digitalization and AI tools

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Integrated Pest Management (IPM) is the cornerstone of the transition to pesticide-free agriculture. However, there is a lack of guidance on how to effectively manage the necessary transition. Currently, there is no comprehensive framework to operationalize IPM for farmers, extension workers and policy makers. Here, we propose to harmonize IPM nomenclature and classify IPM practices into a coherent hierarchical taxonomy based on the European Commission's 8 IPM principles. Based on these principles, we have classified IPM practices by describing them in 4 hierarchical levels. This systematic mapping leads to harmonization of nomenclature within the taxonomy and beyond. Level 1 describes the goal of adaptive management. This serves as a general descriptor for families of practices that fall under the same adapted management objective, emphasizing the practical aspect of IPM. An example might be the selection of crops or the physical control measures used. Level 2 describes the strategy of the IPM practice controlling the pest. This level refines the management objective by describing how the practices are used instead of, or to reduce, the use of pesticides. Layer 3 describes the IPM practices themselves, with the intention to be specific but adaptable to the context in different settings and to the specific conditions in different locations. Level 4 describes the different options within the level 3 practices. This layer will add specificity to and refine the differentiation between different practices. For example, crop rotations (layer 3) can be implemented in different ways, such as crop sequences or service cropping. The taxonomy can facilitate the mapping of future practices and the monitoring of the implementation and effectiveness of practices within and across taxonomic levels and are intended to support policy development by the European Commission. The work has been conducted within the EU project Agrowise.

## **P115 - Using the Taguchi method in field experiments to test integrated disease management strategies**

Precision pathology, digitalization and AI tools

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The multi-factorial nature of integrated disease management (IDM) makes experimental testing resource intensive because there are many possible combinations of control measures. This paper uses healthy canopy area duration (HAD) and yield data from experiments on winter wheat to test the Taguchi Design of Experiments (DOE) approach as a tool to design and scrutinise IDM strategies. The Taguchi approach is used widely in engineering to develop robust industrial processes, to counteract the limited space and high expense associated with experimentation within manufacturing and factory facilities.

At present, the integration of disease control methods is often tested using large factorial, randomised field trials, which limits the number of treatments that can be tested in a single trial. The Taguchi approach offers the opportunity to investigate a larger number of factor combinations than would be practical to include in a single field experiment. The approach tests a sub-set of factors and uses these to explore the effectiveness of untested combinations. A range of metrics associated with the analysis can be used to provide an assessment of the effect of individual factors on disease control and yield.

We will present evidence that demonstrates that the Taguchi approach can be used to design, analyse and predict the effectiveness of the combined action of disease control measures. We will show how the outputs, such as the signal to noise ratio and the contribution ratio, can be used to identify key factors affecting a disease management strategy and identify factors affecting yield. The accuracy and relevance of the Taguchi approach for developing these strategies will be presented alongside the the pros and cons of this approach compared with using classical design of experiments (DOE) approaches.

## **P116 - Molecular and pathogenic diversity of *Xanthomonas* spp. from ornamental plants and development of new diagnostic assays**

Precision pathology, digitalization and AI tools

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Bacteria belonging to the genus *Xanthomonas* have been regularly found in a range of ornamental plants that are frequently imported into the UK including lavender, hydrangea, peony and lobelia. The identification of these pathogens generally involves isolation, sequencing of 16S and *gyrB* genes and pathogenicity tests, which is time consuming. Previously bacterial leaf spot and shot hole of lavender and bacterial spot and blight of peony have been attributed to *X. hortorum* whilst leaf spot of hydrangea has been attributed to a new species, *X. hydrangeae*. A bacterial disease from lobelia has been attributed to *X. hortorum* or *X. dyei*.

In order to clarify the identification of *Xanthomonas* isolates associated with ornamental plants, we have whole-genome sequenced isolates from diagnostic samples received at Fera and from the National Collection of Plant Pathogenic Bacteria. The results revealed at least two clusters of isolates from lavender and distinct clusters from rosemary and peony that do not fit in the described pathovars. Recent hydrangea isolates diagnosed at Fera were attributed to *X. hydrangeae*. Zinnia and lobelia isolates form different clusters possibly outside of known species. Pathogenicity tests of selected isolates from lavender and rosemary showed differences between the clusters and pathovars of *X. hortorum*. Pathogenicity tests of isolates from zinnia and lobelia showed that zinnia isolates were strongly pathogenic mainly on zinnia and lobelia isolates on lobelia. We observed progression of infection on inoculated zinnias that led to detection of the pathogen on seeds.

Based on sequence analyses, new qPCR assays were designed to target four clusters of *X. hortorum* isolates (two clusters from lavender, and clusters from rosemary and peony) and clusters of *Xanthomonas* spp. from zinnia and lobelia. These assays will contribute to rapid diagnosis of these pathogens.

## **P118 - Assessment of bud flush and damage in young Norway Spruce trees through airborne high-resolution multispectral images**

Precision pathology, digitalization and AI tools

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Scandinavia is facing climate changes with a predicted increase in mean temperature of 2-4°C. For Swedish forests to be adapted to this challenge, the Swedish tree breeding program aims to select trees that are adapted to different biotic and abiotic conditions. Information on spring phenology, damage and vitality are important variables in the Norway spruce selection process. Traditionally, the data is gathered through manual assessment of each tree, which requires significant resources and limits the number and frequency of variables that can be measured. As an alternative, Remote Sensing is a promising technology to evaluate bud flush and vitality in conifers, offering the advantage of scoring more trees in a shorter time with fewer resources while obtaining data for several time points during the vegetation season, and its use of algorithms to measure variables reduces the risk of human error.

This project aims to develop methods that can be used within the breeding program by collecting information on spring phenology, damage and vitality using high-resolution multispectral drone images of young Norway spruce trees. Data were collected during spring 2023 and 2024. Bud flush is estimated from the spectral values of the tree crowns using manual assessment of the flush in a subset of the trees as training data. The high-resolution multispectral images will also be used to assess the damage and vitality of the new shoots. To ensure capturing the bud flush at a high temporal resolution, images were taken before the vegetation season and up to twice weekly during the period with the most rapid flush. In the final step, the spatial pattern within the study sites will be analyzed and connected to damage and vitality of the young Norway Spruce trees.

## **P153 - Farmers' Awareness and Management of MLN Disease in Kenya: A KAPs Approach**

Precision pathology, digitalization and AI tools

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Maize lethal necrosis (MLN) disease presents a considerable pathological viral challenge to maize production limiting food and nutrition security goals in Kenya. The effective management of MLN is contingent upon farmers' ability to identify the disease, their knowledge, and perceptions regarding it, and access to relevant information. Nonetheless, empirical data addressing these aspects has been limited. This study surveyed 398 maize farmers across five agroecological zones (AEZs): Moist mid-Altitude (MMA), Dry Mid Altitudes (DTA) Moist Transitional (MT), Highland Tropics (HTs), and Dry Transitional (DT). The research employed a stratified two-stage cluster sampling design. Data were collected through structured questionnaires via computer-assisted telephone interviews (CATIs) using the SurveyCTO tool and analyzed with multivariable logistic regression in STATA software. The findings indicated that 44% had heard of MLN, 41% of the farmers were aware of MLN, exhibiting significant variations across AEZs, and 20% demonstrated awareness of MLN-tolerant varieties. The knowledge levels were highest in the Moist Transitional Zone (47%). Among the identified symptoms, chlorotic mottle and leaf necrosis were the most recognized (90%), in contrast to shortened internodes, which were the least recognized (14%). Among the surveyed farmers, 60% had monitored MLN and engaged in various management practices; the most prevalent disease management strategy involved the uprooting of infected maize plants, followed by burying them (27%) or utilizing them as livestock feed (21%). The primary factors influencing MLN awareness included years of education, group membership, access to MLN information, and extension services. The results were later subsequently validated through the machine learning technique using Lasso logit regression. This study underscores the critical importance of disseminating MLN information to bolster disease monitoring and enhance awareness, thereby facilitating effective MLN management. It is recommended that existing platforms be utilized for the dissemination of MLN-related information to farmers.

## **P154 - Management of crop residues in vineyards for the preventive control of downy mildew epidemics**

Precision pathology, digitalization and AI tools

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Sanitation measures aiming at suppressing crop disease inoculum are a pillar of integrated disease management, since they improve the efficiency of other disease management practices. They are often used in annual crops, involving mostly cultural practices such as mechanical destruction or removal of crop residues containing disease inoculum, but rarely in perennial crops although they could be particularly relevant in such cropping systems where rotation is impossible.

Grapevine downy mildew, caused by the oomycete *Plasmopara viticola*, is one of the most devastating diseases of grapevine worldwide. A growing body of evidence indicates that the primary inoculum (present in leaf litter and soil as oospores) plays a major role in downy mildew epidemics. This suggests that sanitation measures could significantly contribute to the control of the disease.

To test this, we conducted a 2-year experiment in a vineyard divided in two plots: a control plot and a sanitation plot where leaf residues were removed each autumn using a hand-held blower. Disease and leaf fall dynamics were monitored every two weeks from August to November. Each month from harvest to budburst, grapevine leaf litter samples were collected in the control plot, and weighted. Soil was sampled at the onset of spring in control and “sanitation” plots. *Plasmopara viticola* DNA was quantified in soil and litter samples using ddPCR to assess the quantity of oospores in these reservoirs.

Results showed that disease considerably spread during the fall after the spraying of chemical fungicides has stopped. Crop residue removal significantly reduced primary inoculum in the vineyard and the incidence of the disease on leaves and inflorescences early in the next growing season. This work represents an important step in the design of integrated disease management strategies in vineyards, towards the phase-out of pesticides.

## **P155 - Smart surveillance for detecting mutations in fungicide target genes of *Septoria tritici***

Precision pathology, digitalization and AI tools

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*Septoria tritici* blotch (STB), caused by the fungal pathogen *Septoria tritici* (syn. *Zymoseptoria tritici*), is a major threat to global wheat production. The disease is responsible for significant yield losses, and its control relies heavily on the application of fungicides. However, the emergence and spread of fungicide-resistant strains, driven by mutations in target genes, present a growing challenge to effective disease management. Detecting these mutations at an early stage is essential for minimising the impact of resistance and ensuring sustainable crop protection strategies.

This study explores the application of smart surveillance techniques for detecting mutations in the fungicide target genes and gene regions of *S. tritici*. Specifically, we focus on utilising spore trapping mechanisms integrated with advanced molecular diagnostics, to monitor airborne *S. tritici* spores in wheat fields. Spore traps are deployed in wheat fields to continuously capture fungal spores, which are then subjected to high-throughput amplicon sequencing targeting all six relevant fungicide resistance genes and gene regions, enabling the detection of both known and novel mutations associated with resistance.

The integration of spore trapping with molecular tools enables rapid and efficient detection of pathogen populations in the environment before symptoms manifest in crops. Continuous monitoring of the genetic composition of *S. tritici* populations provides valuable insights into the evolution of fungicide resistance, facilitating timely intervention to mitigate its spread.

This smart surveillance approach offers a proactive solution to managing fungicide resistance in *S. tritici*, supporting sustainable disease management strategies. It demonstrates the potential of combining innovative monitoring technologies with molecular diagnostics for improved agricultural disease surveillance.

# **Invasive and emerging plant diseases**

## Transgenerational response of alders to stress: effects of maternal biotic stress on seedling drought tolerance

Invasive and emerging plant diseases

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Alders are key species in riparian forests, contributing to the maintenance of ecosystem services such as water quality, flood mitigation, and biodiversity. Its regeneration is essential for ecosystem functioning but is threatened by both abiotic and biotic stressors. Among them, the *Phytophthora alni* species complex is the main driver of alder decline in Europe, causing trunk necrosis, defoliation, and mortality, especially in seedlings.

Seedling drought tolerance is influenced by environmental conditions and genotype, but also by the maternal environment through seed provisioning or epigenetic mechanisms. In this study, we evaluated the transgenerational effect of maternal biotic stress (i.e., symptoms of *P. alni* infection) on the drought response of the progeny.

Seeds were collected from symptomatic and asymptomatic trees in two localities in Northern Portugal where *P. alni* was confirmed. Seedlings were grown for seven months and then subjected to a drought experiment in a greenhouse. Half of the seedlings were exposed to reduced soil water content (gradually lowered to 25% of the field capacity), while the other half remained at 100%. We assessed drought response through physiological (photosynthesis, gas exchange), morphological (height, leaf number, biomass allocation), and biochemical and chemical traits (phenolic compounds, N content).

Exposure of mother trees to biotic stress altered seed composition, increasing phenolic compounds, but did not affect early seedling height. Under moderate drought (50% field capacity), seedlings from symptomatic mothers maintained higher photosynthesis compared to those from asymptomatic mothers, possibly linked to a higher leaf nitrogen content. They also preserved root mass fraction under drought, unlike seedlings from asymptomatic trees.

These findings suggest that maternal exposure to *P. alni* may confer improved drought resilience to progeny, highlighting the importance of transgenerational effects in alder regeneration under stress.

## **O5.1 - *Phytophthora palmivora*: A serious threat to papaya production in South Africa**

Invasive and emerging plant diseases

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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.

## **O5.2 - Situation of elms in the light of Dutch elm disease in northern Europe**

Invasive and emerging plant diseases

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As elms' (*Ulmus*) natural distribution in northern part of Europe is in their northern limit, they grow quite scarcely in this region and are not important for wood production. In the other hand elms have cultural and ecological values. Dutch Elm Disease (DED) had been registered in Baltic and Nordic countries in different times. The updated overview of the spread of the disease in that region is given. The pathogen causing that disease can vary based on its genome and name. By the behaviour of this pathogen some strains can be more aggressive.

Some background of my Postdoc topic New approaches for conservation of threatened elm trees through holobiont concept is given. The overall goal of this project is to expand the current knowledge base for development of sustainable conservation strategies for elms (*Ulmus* spp.) under the threat of Dutch elm disease.

### **O5.3 - Swedish risk ranking of plant pests**

Invasive and emerging plant diseases

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SLU Risk Assessment of Plant Pests is a unit at SLU tasked with performing independent analyses and pest risk analysis (PRA) of new plant pests to support the risk management conducted by the Swedish Board of Agriculture (the Swedish National Plant Protection Organisation, NPPO). Depending on the current needs and objectives of the NPPO, different types of risk assessments are performed.

Upon request from the NPPO, a risk ranking project was initiated with the aim to rank all quarantine pests relevant to Sweden as well as selected non-regulated pests as needed. The risk ranking was performed using the FinnPRIO model by Heikkilä et al. (2016, *Biol. Invasions* 18) which is based on rapid assessments of components included in full PRAs, i.e. the likelihood of entry, establishment combined with spread, magnitude of impact and management ability. The model is semi-quantitative and enables a ranking of plant pests based on their risk (Entry × Establishment & Spread × Impact) or any of the assessed components separately. The model also includes an estimation of the uncertainty of the assessments. The project is ongoing and new pests are assessed and ranked continuously.

Risk ranking of plant pests can provide valuable information to support plant health risk management decisions and resource allocation. The assessments and rankings at the national level can be used to allocate and prioritize resources to pests with the highest risk, e.g. for inventories, as well as be used to evaluate the relative risk of currently non-regulated pests.

## O5.4 - Plant pathogens detected in horticultural plants and seeds imported to Norway

Invasive and emerging plant diseases

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Global plant trade has increased dramatically over the past 50 years and so has the establishment of invasive disease-causing pathogens. In the project STOPPest (Risk management of imported plants and seeds: possibilities for improved pest detection to prevent the introduction and spread of new pest; 2021-2025) we have studied different plant pathogens that can accompany imported plant material. In 2021, investigations were primarily focused on plant pathogenic *Phytophthora* species, which are mainly introduced as stowaways in the root balls of imported landscaping and garden plants. In 2022, the focus was both on the quarantine pathogen *Phytophthora ramorum*, which mainly arrives with imported *Rhododendron* to Norway, and on pathogens of strawberry plants. In 2023, the focus was on fungal and viral plant pathogens in imported seeds of selected species of vegetables and flowers for the hobby market. The study included laboratory analyses for *Phytophthora* and fungal pathogens on horticultural- and strawberry plants, viruses and viroids in tomato seeds and fungal pathogens in seeds of eight crops popular in garden cultivation. Altogether many common and less common pathogens were detected in samples from the imported plants and seeds, even though they were accompanied with a Plant Health Certificate. Two of them were the quarantine pathogens *Phytophthora ramorum* in *Rhododendron* and *Pieris* plants, and potato spindle tuber viroid (PSTVd) in tomato seeds. *Phytophthora* species were particularly prevalent with 16 species found in 65 of 150 soil/rot samples (43.3%) from 43 different host plants in the 2021 case study. These findings show that current regulations and enforcement do not prevent invasive disease-causing pathogens from being introduced, leading to negative consequences for Norwegian plant production and native flora.

## **O5.5 - Bacterial leaf streak caused by *Xanthomonas vasicola* pv. *vasculorum*, a disease that might threaten European maize crops**

Invasive and emerging plant diseases

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Plant diseases caused by *Xanthomonas* species affect many different crops worldwide. The species *Xanthomonas vasicola* currently includes four pathovars: *arecae*, *musacearum*, *vasculorum* (Xvv), and *holcicola* (Xvh). Of these only Xvh has a validly published name, although the others have been proposed.

Maize is the most widely grown cereal in the world. Bacterial leaf streak (BLS) of maize caused by Xvv was first reported in South Africa in 1948 and was not reported anywhere else until 2014 when it was identified in Nebraska; BLS then spread rapidly and is now present in ten states of the USA and it has been reported in some regions of Argentina and Brazil.

The main host of Xvv is maize, but artificial inoculations showed that other monocot species like oat and rice are also hosts. The most likely pathway of entry in Europe will be on seed, although there are only a few reports of seedborne infections.

We have confirmed pathogenicity of isolates from the NCPPB collection and isolates received from USA and Brazilian collections. A range of maize varieties grown in the UK were shown to be susceptible to this pathogen. Two varieties were tested at three different temperatures, 15°C, 20°C and 25°C, and while symptoms were recorded at all temperatures, the severity of symptoms notably increased at higher temperatures. There is uncertainty whether UK summer temperatures are sufficiently hot for extensive establishment of the disease, but hotter summers might be suitable, and the risk will be higher in the south of Europe.

We have whole genome sequenced isolates from the USA, Argentina and Brazil and compared the sequences with publicly available data. The results indicated that there have been at least two introductions in Brazil.

Although not currently present in Europe, BLS should be considered as a potential threat to maize crops.

## 05.6 - Ash dieback: impact of two invasive species and resistance potential

Invasive and emerging plant diseases

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Two invasive pests threaten ash trees (*Fraxinus* spp.) in Europe: ash dieback (ADB), caused by *Hymenoscyphus fraxineus*, and the Emerald Ash Borer (*Agrilus planipennis*, EAB). ADB has been present since the 1990s, severely affecting European ash (*Fraxinus excelsior*), while EAB was first detected in eastern Ukraine in 2019, initially infesting *F. pennsylvanica* before spreading to *F. excelsior* in urban and forest stands.

This study aims to assess the impact of ADB and EAB on *F. excelsior* in Ukraine, monitor disease progression, and identify potentially resistant trees for conservation and breeding. *H. fraxineus* is now widespread, causing crown dieback, basal lesions, secondary fungal infections, and insect infestations, leading to tree mortality. EAB infestation further accelerates ash decline.

Long-term field trials in Eastern and Central Ukraine (14 monitoring plots, 234 trees) revealed significant variation in susceptibility, with disease severity ranging from minimal symptoms to over 90% dieback. Between 2016 and 2024, ash mortality increased, particularly from 2020 to 2021. Combined with intensive logging, this has led to significant decline of *F. excelsior*. Since 2021, EAB has further intensified mortality. By 2024, most trees were classified as intermediate or severely damaged (categories 2–4), while only 17% remained in the healthy category (0–1). Despite these threats, a small fraction of *F. excelsior* trees remain vigorous, exhibiting little to no symptoms. Through long-term monitoring, we phenotype these potentially resistant trees, offering hope for natural selection and breeding efforts. Recent surveys indicate EAB is expanding westward and attacks ash trees indiscriminately, regardless of ADB symptoms. Our study reviews disease and pest progression, mortality patterns, and resistance potential, highlighting the need to select resistant genotypes for conservation and restoration across Europe.

## **P67 - Efflux pumps mediate reduced sensitivity to fluazinam in *Phytophthora infestans***

Invasive and emerging plant diseases

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Potato late blight, caused by *Phytophthora infestans*, is a critical agricultural issue globally. Fluazinam, a fungicide that uncouples oxidative phosphorylation in mitochondria (FRAC code: 29), is classified as having a low risk of resistance development. It has been hypothesized that the fluazinam molecule itself acts as a proton carrier in the mitochondrial inner membrane independently of any specific protein target. This suggests that fluazinam does not have a protein target. However, a strain exhibiting reduced sensitivity was identified in 2018. This study examines the role of efflux pumps in this reduced sensitivity and explores strategies to preserve fluazinam's efficacy. Using clorgyline, an efflux pump inhibitor, we demonstrated that fluorescent dye Rhodamine 6G, typically not retained in cystospores of the resistant strain, was accumulated upon inhibitor treatment. Additionally, fluazinam's inhibitory effects on cystospore germination were restored in a dose-dependent manner with clorgyline. These findings strongly suggest that efflux pumps, rather than mutations in a target protein, are responsible for the observed sensitivity reduction. Further experiments showed that combining fluazinam with fungicides that potentially affect the activity of the ATP-driven efflux pump located at the cell membrane, cyazofamid (an ATP synthesis inhibitor) or valifenalate (a cell wall synthesis inhibitor) enhanced its fungicidal activity. These results underscore the importance of efflux pump modulation as a complementary strategy for disease management. Our findings confirm fluazinam's status as a low-risk fungicide and highlight its continued potential in managing *P. infestans* outbreaks.

## **P119 - *Corinectria* pathogens – a new threat to *Abies* spp.**

Invasive and emerging plant diseases

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The genus *Corinectria* was recently introduced to accommodate *C. constricta*, *C. fuckeliana* (formerly *Neonectria fuckeliana*), and *C. tsugae* (formerly *N. tsugae*). Fungi in this genus include economically significant pathogens of forest trees. *Corinectria* species have been reported from both the Southern and Northern Hemispheres and are known to cause canker diseases in coniferous tree genera such as *Abies*, *Larix*, *Picea*, and *Pinus*. Plantations used for timber production are particularly vulnerable to infections by these fungi, but trees in other settings can also be affected.

Recent observations of declining *Abies* trees revealed disease symptoms, including stem or branch deformation, branch and twig dieback, and cambium necrosis. Fruiting structures on diseased tissues, along with pure culture isolates, resembled fungi within *Nectriaceae*. To determine their identity and pathogenicity, molecular identification, phylogenetic analyses, and pathogenicity tests were conducted. ITS rRNA sequencing confirmed that fungi isolated from declining *Abies* trees belonged to the genus *Corinectria*. A multi-locus phylogenetic analysis revealed that these isolates represented two distinct, previously undescribed species, which were well separated from other known *Corinectria* species. Pathogenicity tests demonstrated that these novel *Corinectria* species were pathogenic to *Abies* seedlings, although infection rates and incidences of seedling dieback varied among species.

## **P120 - *Aphanomyces macrosporus* causing root rot in barley and some other plants**

Invasive and emerging plant diseases

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A new root rot disease in barley, caused by an *Aphanomyces* species, was found in field surveys in Southern Sweden and Denmark. The symptoms included yellowing of leaves, brown coleoptiles, and discoloration of roots. Soil wetness after high rainfall favored the disease, resulting in yellow patches, which could extend to entire fields, with significant yield losses. Oospores were found in the fine roots, and *Aphanomyces* isolates were obtained from these roots. Based on morphological analysis, the isolates were similar to those already obtained from barley and spinach roots in the 1990s in the same growing area, but at that time identified as an unknown *Aphanomyces* sp. Morphological and molecular analyses clearly separated and distinguished these isolates from other known *Aphanomyces* species, and *Aphanomyces macrosporus* has been proposed as a new plant pathogenic species. It can at least infect barley, spinach, and sugar beet, indicating a broad plant host range. The widespread presence and presumably broad host range of this new pathogenic *Aphanomyces* species must be considered in crop rotation planning.

## **P121 - First report of Raspberry leaf blotch virus on Rubus in Latvia**

Invasive and emerging plant diseases

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Red raspberry is a valuable soft fruit crop due to its flavour and high nutritional value. Its market demand increases each year, but the high request for local products cannot be met due to low berry yield and quality. In recent years, the raspberry leaf and bud mite *Phyllocoptes gracilis* has become more prevalent in raspberry fields, most likely due to favourable weather conditions. Previously, damage from raspberry leaf mites was observed only in protective plastic tunnels, not in open-field conditions. Additionally, replacing the assortment of raspberry cultivars which produce firm berries with large drupelets, but are more susceptible to mite damage, may be contributing to the spread of raspberry leaf mites. Raspberry leaf mite is also a vector of *Emaravirus idaeobati* (Raspberry leaf blotch virus; RLBV) which is attributed to leaf chlorosis, distortion and patchy necrosis and death of lateral branches and subsequent loss of fruit yield. In the *Rubus* genetic resource collection, several plants exhibited diffuse yellow areas and chlorotic blotches on their leaves. A total of 57 samples were collected from plants with indicative symptoms of viral infections, including chlorosis, yellowing, chlorotic spots, flecks, and the formation of crumbly fruit. RLBV detection was performed with One Step RT-PCR assays using previously published virus-specific primers. RLBV was detected in seven plants exhibiting yellow leaf blotch symptoms typical for the virus. The amplified PCR products were sequenced, confirming the presence of RLBV in Latvia for the first time.

**P122 - Identification and characterization of *Alternaria alternata* and *Stemphylium vesicarium* associated with almond leaf spot.**

Invasive and emerging plant diseases

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Almond (*Prunus dulcis* Mill.) diseases are a major focus of research worldwide, especially in regions where almond cultivation is an integral part of agriculture. This study investigates the fungal species associated with a leaf spot outbreak observed in a commercial almond orchard in Latium region in Italy. Symptoms were observed on leaf blades during late spring and summer, appearing as small, circular, brown spots 1-3 mm in diameter, which later expanded to 5-20 mm. Representative fungal isolates were characterized by multigene phylogenetic analysis using internal transcribed spacer (ITS), translation elongation factor 1- $\alpha$  (EF-1 $\alpha$ ), glyceraldehyde-3-phosphate dehydrogenase (GAPDH), a SCAR marker (OPA10-2) for *Alternaria sp.* and calmodulin for *Stemphylium sp.* The fungi isolated from lesion margins were identified as *Alternaria alternata* and *Stemphylium vesicarium*. Pathogenicity tests on almond leaves confirmed that both pathogens caused similar symptoms, and the fungi were successfully re-isolated from symptomatic tissue, fulfilling Koch's postulates. This study highlights the need for targeted disease management strategies to mitigate the impact of foliar pathogens on almond production, particularly emerging pathogens in intensive farming systems.

## **P123 – Natural regeneration of beech (*Fagus sylvatica*) on *Phytophthora*-infested soils**

Invasive and emerging plant diseases

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Soil-borne pathogens from the genus *Phytophthora* increasingly threaten Southern Swedish beech (*Fagus sylvatica*) forests by damaging tree roots, thereby predisposing them to rot fungi, insect infestations, and windthrow. The inability to eliminate soil infections raises challenges for forest regeneration in affected areas.

While nursery plants are known vectors for *Phytophthora* spread, natural regeneration may offer a safer alternative. However, there is limited understanding of how naturally regenerated beech seedlings and seeds are impacted by existing *Phytophthora* in the soil. To address this knowledge gap, we investigated a 120-year-old beech forest in Southern Sweden, where *Phytophthora* was previously detected in mature trees and soil.

We analyzed naturally regenerated seedlings (2–3 years old) and seeds from sites with and without *Phytophthora* presence, employing microscopy, cultivation tests, and molecular methods. This study focused on determining the prevalence of *Phytophthora* in seedlings and seeds to evaluate the risks associated with natural regeneration in infected soils. Our results showed an extremely low prevalence of *Phytophthora* in young seedlings (0.31% of isolates), with no detection in seeds.

Most seedlings exhibited good health, though some showed symptoms like leaf spots (85.71%) or root lesions (8.8%). Natural regeneration and direct seeding appear to be viable strategies for establishing new beech forests facing *Phytophthora* risk.

## **P124 - Overview of EFSA activities in plant health and risk assesment.**

Invasive and emerging plant diseases

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The European Food Safety Authority (EFSA) is the Agency of the European Union responsible for risk assessment in food safety, animal health and welfare and plant health. EFSA conducts risk assessment for individual plant pests with a two-phase fit for purpose approach, a simpler pest categorisation first, followed by a quantitative pest risk assessment for more complex questions. Moreover, EFSA also assess the probability of pests being introduced with plants for planting and other plant material in commodity risk assessment. On top of other activities EFSA is also funding research projects to reduce key uncertainties and knowledge gaps by generation and collection of observational and experimental evidence.

## **P125 - Presence of *Diaporthe* species in canes impacts the production of healthy propagation material in grapevine nurseries**

Invasive and emerging plant diseases

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The genus *Diaporthe* consists of fungi with a wide host range, including grapevine. The aim of the present study was to investigate the transmission capability of *Diaporthe* species through the propagating material, utilizing phytopathological and molecular methods. Initially, Nested PCR approaches were performed to detect *Diaporthe* spp. in grapevine canes collected from rootstock mother vines. The molecular detection showed that rates ranged from 75%-100% at the basal end of the canes and from 0% to 100% at the cane tips. Subsequently, *Diaporthe* species isolated from symptomatic rooted-grafted vines grown for two months in the nursery field, were characterized by a multi-locus sequence tagging method and five distinct fungi belonging to the species *D. ampelina*, *D. eres*, *D. foeniculina*, *D. infecunda* and *D. novem* were identified. The pathogenicity trials on young grapevine plantlets derived from micropropagation showed that the species *D. eres* and *D. novem* were the most aggressive, while the species *D. infecunda* was characterized as a mild pathogen. This is the first report of *D. infecunda* and *D. novem* in Greece, while the species *D. eres* and *D. foeniculina* were identified on grapevine for the first time in Greece, as they have been previously found to infect other tree crops. The presence of *Diaporthe* spp. on rootstock mother vines and the isolation of different species from rooted-grafted vines in the nursery field, provide solid evidence of their spread with the propagating material, while the proper molecular identification coupled with the morphological traits of *Diaporthe* species is essential for the development of their management in grapevine nurseries.

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## **P126 - Powdery mildew of okra (*Abelmoschus esculentus*) is caused by *Golovinomyces bolayi* in Hungary**

Invasive and emerging plant diseases

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Okra (*Abelmoschus esculentus*) is a vegetable crop, cultivated for its fibrous pods throughout the world in the tropical and warm temperate regions. In Europe, its cultivation and consumption have been increasing. In Hungary, okra is primarily grown on a small scale by hobby growers. In 2023, powdery mildew symptoms were observed on okra plants grown in a garden in Western Hungary. As several species of powdery mildew fungi are known to infect okra plants, our aim was to identify the causal agent.

For this, we conducted morphological observations and sequenced the internal transcribed spacer, stretches of the intergenic spacer region, and the 28S rRNA encoding region of the ribosomal DNA.

Observations revealed catenate conidial development, a lack of fibrosin bodies in the conidia, and simple, solitary hyphal appressoria, suggesting that the powdery mildew was caused by a *Golovinomyces* species. Based on the sequences obtained, the causal agent was identified as *G. bolayi*, a polyphagous species pertaining to the *G. orontii* s. lat. species complex. This species has only been known to infect okra in Iran. Thus, this is the first report of the fungus infecting okra in Europe.

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## **P127 - Studies on fungal pathogens associated with emerging blackcurrant decline in Latvia**

Invasive and emerging plant diseases

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Blackcurrants (*Ribes*) is the second most widely commercially grown fruit species in Latvia. It is grown with rainfed cultivation, and about 70% is cultivated with organic farming practices. Recently, grower concerns have been raised about severe blackcurrant decline potentially induced by a combination of drought and heat periods during the last vegetation seasons and increased damages by fungal pathogens. Fungal diseases, such as cankers, diebacks, blights and decline, caused by various pathogens on woody fruit crops, are in hot spots worldwide due to the increasing damages predisposed by fungicide availability reduction, intensified cultivation, global trade and climate changes. The research was initiated in 2024 within the State Research Program project “GreenAgroRes” (No.VPP-ZM-VRIILA-2024/1-0002) to identify fungal pathogens involved in the blackcurrant decline. Fields in organic farms and genetic resource collections were surveyed in the spring of 2024; whole shoots were sampled from declined plants; fungal isolations were made in pure cultures and identification was performed by combining morphological traits and ITS1/5.8S/ITS2 region sequencing. The great diversity of fungal taxa was identified on declined blackcurrants, including fungi from genera comprising well-known woody host pathogens or previously reported on *Ribes* and known as weak pathogens. These included *Eutypa*, *Sporocadus*, *Tyronectria*, *Dothiora*, and *Fusarium* species. Blackcurrants are very susceptible to drought and must undergo dormancy with satisfactory chill hours. In recent years, warm winters, regular draught and heat stress during the vegetation season may have influenced blackcurrant susceptibility to weak pathogens or have favoured some pathogen development that has led to increased disease spread and damages caused. The bioassay series has been started on potted plants in the greenhouse to assess the pathogenicity of identified fungi on various *Ribes* species and cultivars.

## P128 - Studies on fungi causing stem canker and decline of sea buckthorn in Latvia

Invasive and emerging plant diseases

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Sea buckthorn (*Hippophae rhamnoides* L.) is the third most widely grown fruit species in Latvia, often grown in organic farming. The plantation areas increase every year, reaching 1535 ha in 2023. It is a highly adaptive species to various extreme environments but is vulnerable to diseases and pests. With the increase in cultivation, the disease damages have increased significantly, indicating that plant health concerns must be addressed more intensively. In initial studies, diverse fungal genera have been identified on diseased sea buckthorn in Latvia. These also included fungi from genera comprising well-known woody host pathogens (e.g. *Diaporthe*, *Eutypa*, *Eutypella*, *Diatrypella*, *Fusarium*) reported as causes of cankers, dieback and wilt on other plant species. The aims of the study were: 1) identify the species of fungi associated with the stem canker and decline of sea buckthorn in Latvia; 2) assess the pathogenicity of dominating fungi on sea buckthorn and other possible hosts; 3) evaluate cultivar tolerance and characterize pathogen virulence on different cultivars. The bioassay series was established on detached sea buckthorn leaves and potted plants in the greenhouse (five sea buckthorn genotypes, apple and pear seedlings). Ten strains of diatrypaceous fungi, one strain of *Hymenopleella* and two strains of *Diaporthe* were tested. All tested fungal species and strains caused cankers on sea buckthorn. The sea buckthorn has been introduced from different parts of Eurasia, and some of the identified species indicated that pathogens could be introduced together with the host from other regions. First observations suggest that the host range of pathogens should be considered, and the cultivar resistance may be considered to control sea buckthorn stem canker and decline.

## **P129 - Unraveling *Sarocladium* pathogenicity in rice: Toxin biosynthesis, host infection, and sheath rot disease progression**

Invasive and emerging plant diseases

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Sheath rot disease, primarily caused by *Sarocladium* spp., poses a growing threat to rice production, with *S. sparsum* identified as the most virulent species. Comparative genomics of diverse *Sarocladium* isolates has revealed several genetic elements that may play a role in pathogenicity, such as biosynthetic gene clusters (BGCs) involved in toxin production. However, the precise molecular mechanisms through which these elements contribute to infection, tissue necrosis, and disease progression remain poorly understood. To address this gap, our study focuses on experimentally validating these candidate virulence factors through a combination of CRISPR-Cas9 genome editing and GFP-based live imaging techniques.

CRISPR-Cas9-mediated gene knockouts will target specific BGCs responsible for the biosynthesis of key toxins like helvolic acid and cerulenin. By comparing the pathogenicity of wild-type and mutant strains in *in-planta* assays, we aim to determine the contribution of these factors to disease development. Furthermore, GFP-labeled *Sarocladium*'s strains will be employed to monitor fungal colonization and host-pathogen interactions using multispectral imaging. This approach will allow detailed observation of infection dynamics, including tissue invasion patterns and the timing of virulence factor expression.

Ongoing investigations will further dissect the contributions of additional virulence factors identified through genomic analyses, bridging the gap between genomic predictions and functional validation. These insights will enhance our understanding of *Sarocladium*'s infection strategies and lay the groundwork for developing targeted disease management approaches.

**P156 - *Colletotrichum acericola* as a threat to native maple species (*Acer* spp.) in Europe.**

Invasive and emerging plant diseases

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*Colletotrichum acericola* is a fungus belonging to the *Colletotrichum* agaves species complex. It has been isolated in Poland from the fruit of boxelder maple (*Acer negundo*), a tree from the *Acer* genus native to North America. The aim of this study was to assess the potential threat posed by this fungus to *Acer* species native to Europe, specifically common maple (*Acer platanoides*) and sycamore maple (*Acer pseudoplatanus*). Pathogenicity tests conducted on leaves, shoots, and fruit confirmed that multiple strains of *C. acericola* are capable of infecting *A. platanoides* and *A. pseudoplatanus*. Infected plant tissues exhibited necrosis and sparse to woolly white mycelium, as well as orange to black acervuli on fruit and leaves.

## **P157 - *Discosia* sp.nov. isolated from the rare and endangered species *Oxytropis pilosa*.**

Invasive and emerging plant diseases

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*Discosia* is a genus of saprobic or plant-pathogenic fungi, commonly referred to as 'pestalotioid fungi,' within the *Amphisphaeriaceae* family (*Xylariales*). This genus has a broad cosmopolitan distribution and is known to cause leaf spot diseases in various plant species, including *Rhododendron*, *Galium odoratum*, *Gymnocarpium dryopteris*, *Oxalis acetosella*, *Tilia cordata*, *Prunus serotina*, and *Acer rubrum*. Infected leaf tissues typically develop epiphyllous, black, and shiny pycnidia, within which conidia are produced. These conidia are multicellular and possess appendages at both the base and the terminal cell. In 2024, an unidentified strain of pestalotioid fungi was isolated from *Oxytropis pilosa*, a rare and endangered plant species growing in Wigierski National Park, Poland. Initial molecular identification based on the ITS region sequence classified the isolate as *Discosia* sp. However, subsequent phylogenetic analyses incorporating ITS, LSU, and  $\beta$ -tubulin loci confirmed that the strain represents an undescribed species. Morphological characterization and growth rate assessments were conducted on MEA and PDA media. This study highlights the discovery of a potentially novel *Discosia* species, contributing to the understanding of fungal diversity and its impact on endangered plant hosts.

## **P158 - Goldenrod rust - a new threat to Europe P30?**

Invasive and emerging plant diseases

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Rust fungi belong to the order *Coleosporium* (*Pucciniales*) and are obligate parasites of vascular plants. Most of them are specialized pathogens targeting specific plant species, including goldenrod rust. Their telial hosts are plants from the Asteraceae family, while their aecial hosts are *Pinus* spp. Their presence in Europe has been recorded in France, Portugal, Switzerland, and Germany. It has also been identified in Poland. Due to discrepancies in genetic studies, the precise phylogenetic affiliation of the species causing this disease has not been determined. This has created a need for further research to accurately identify the species and assess whether they pose a threat to, among others, *Pinus sylvestris*, which is the primary forest-forming species in Poland. The aim of this study is to provide an introduction and familiarize readers with the pathogen, as well as to preliminarily assess its potential threat to Europe.

## **P159 - Unveiling Oman's mycological wealth: New fungal species and their Potential applications**

Invasive and emerging plant diseases

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Oman's unique position at the southeastern edge of the Arabian Peninsula, bridging temperate and tropical zones, endows it with a rich tapestry of landscapes—from coastal plains and deserts to mountain ranges—where temperatures fluctuate between 0°C and 50°C. This environmental diversity fosters a wide array of plant and fungal life. Between 2016 and 2023, extensive explorations across various regions of Oman led to the collection of over 300 fungal and mushroom specimens. Through morphological assessments and nucleic acid sequencing, we have identified at least 24 novel species. Notably, nine of these are mushroom species spanning genera such as *Agaricus*, *Amyloporus*, *Fuscoporia*, *Hymenagaricus* (two species), *Micropsalliota*, *Termitomyces*, and *Xanthagaricus* (two species). The remaining new species belong to genera including *Bipolaris*, *Acrocordiella*, *Talaromyces*, *Cladosporium*, *Hydeomyces* (two species), *Alternaria*, *Bimuria*, *Phaeosphaeriopsis*, *Calophoma*, *Dictyoarthrinium*, *Omania*, *Desertiserpentina*, *Montanitestudina*, and *Basidiodesertica*. Investigations into some of these fungi have unveiled their potential as plant growth promoters and biocontrol agents. Despite these promising discoveries, the fungal diversity in Oman remains underexplored, leaving much of the nation's mycological wealth undocumented. Given the rising global interest in the ecological roles and biotechnological applications of fungi, Oman offers a promising frontier for future research. Further exploration and documentation are imperative to fully grasp the extent and ecological significance of Oman's fungal diversity, especially within its arid and semi-arid ecosystems.

## **P160 The occurrence of bean yellow mosaic virus and cucumber mosaic virus in organic cultivation of yellow lupin in Poland**

Invasive and emerging plant diseases

Diana Czarnecka, Anna Czubacka, Anna Trojak-Goluch, Jerzy Księżak

Yellow lupin (*Lupinus luteus* L.) belongs to the Fabaceae family, and its seeds provide protein-rich animal feed. Recently, there has been growing interest in organic farming. Unfortunately, under such farming conditions, plants are more vulnerable to diseases. Of the viral diseases threatening yellow lupin crops, bean yellow mosaic virus (BYMV) and cucumber mosaic virus (CMV) are of greatest economic importance.

In the present study, the occurrence of BYMV and CMV was determined in 3 locations of organic yellow lupin cultivation in the eastern Poland, i.e. Osiny (Lublin voivodeship), Szepietowo (Podlaskie voivodeship) and in Grabów (Mazowieckie voivodeship). In 2022-2024, 6 varieties (Goldeneye, Salut, Diament, Puma, Mister, Bursztyn) were evaluated in late June and early July. The presence of viruses was detected by DAS-ELISA immunoassay with specific antibodies. Leaves from 40 randomly selected plants of each variety were collected for analysis.

A small number of BYMV-infected plants were observed in Szepietowo in 2022 and 2023, respectively: 42 and 24 plants. However, in 2024, there was a significant increase in BYMV infection in Szepietowo - 165 infected plants. In Grabów, 71 plants were infected in 2022, 106 in 2023, and 222 in 2024. In Osiny, 64 plants were infected in 2022, 79 in 2023, and only 59 in 2024.

In 2022, CMV was detected in 18 plants in Grabów and in a single plant in Szepietowo. In 2023, CMV infection was found in 56 plants in Szepietowo and 51 in Grabów and in only 5 plants in Osiny. An increase in infection was observed in 2024, when 130 CMV-infected plants were found in Grabów, followed by 59 plants in Osiny and 31 plants in Szepietowo. Among the varieties tested, Goldeneye seems to be the most susceptible to BYMV but not to CMV. The highest virus pressure was observed in 2024 in Grabów.



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