22nd - 25th August 2022

21st Annual Meeting of the Oomycete Molecular Genetics Network

Program and Abstracts

OMGN2022

Oomycete biology, pathology and ecology

Faculty of Forestry and Wood Technology
Mendel University in Brno, Czech Republic

EUROPEAN UNION
European Structural and Investment Funds
Operational Programme Research,
Development and Education

MINISTRY OF EDUCATION, YOUTH AND SPORTS

PhytOphthora Research Centre

MENDELU
Faculty of Forestry
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Contents

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OMGN2022:
We walk on the footsteps of Gregor Johann Mendel…
We pay him a tribute on his 200th birthday.

Welcome to Brno, welcome to MendelU!

After a 2-years gap due the Covid-19 pandemic, we are happy that the 21st Annual Meeting of the Oomycete Molecular Genetics Network in Brno has attracted so much interest: 89 participants from 16 countries. With precautions, we try to learn how to deal with a new reality as we return to in-person meetings. We wish OMGN2022 will continue to be a place of communication and collaboration among the Oomycete scientific community, the chance to get in contact with fellow scientists, strengthening the exchange of ideas and paving the way for multinational cooperations. We hope that you will find in Brno the perfect atmosphere for this to happen.

You are welcome and we wish that your stay will be both very interesting and comfortable.

Thomas Jung

(on behalf of the Phytophthora Research Centre Team)

Phytophthora Research Centre

Follow us!

#OMGN22 #OMGN2022

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Acknowledgements

This meeting is supported by Project Phytophthora Research Centre Reg. No. CZ.02.1.01/0.0/0.0/15_003/0000453 (co-financed by the European Regional Development Fund) and the Faculty of Forestry and Wood Technology at Mendel University in Brno.

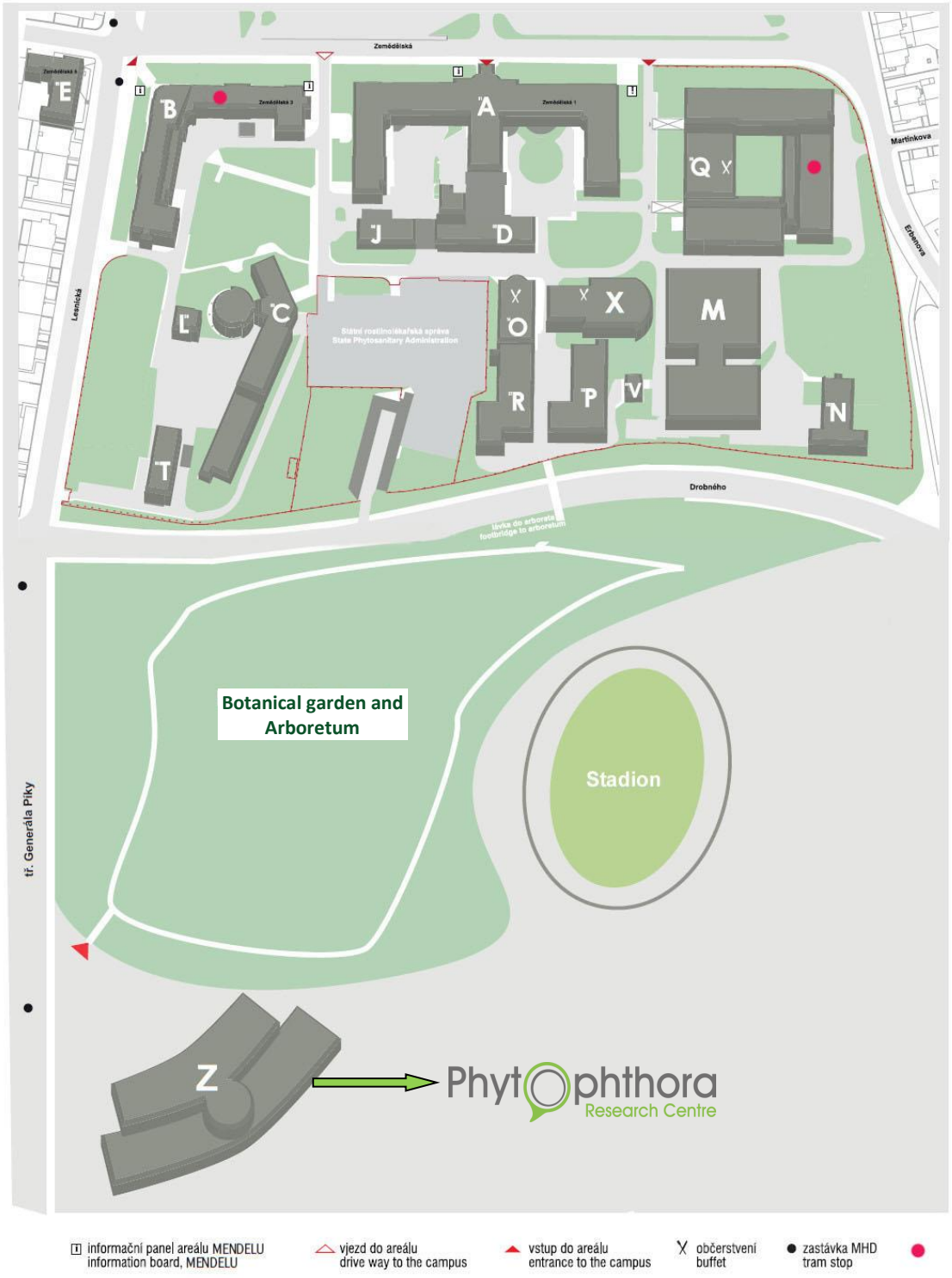
A special thanks to Joel Shuman (Virginia Tech) for his assistance with the updating of information on the webpage omgn.org, and to Francine Govers (Wageningen University) for advising on the meeting organization.

The Organizing Committee also acknowledges the support of external Partners of the OMGN2022 meeting (listed in page 83).

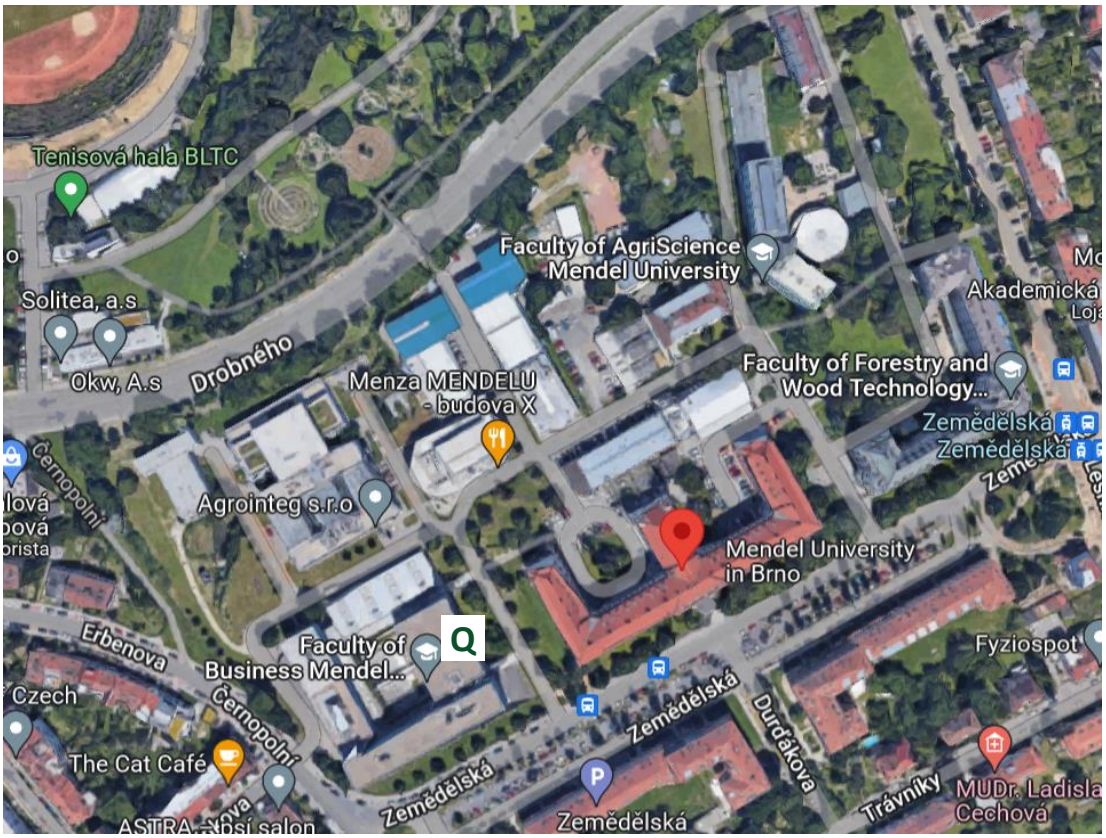
This conference is organized under the patronage of the Mayor of the City of Brno, Markéta Vaňková.

Venue

The conference will be held in meeting room Q01, building **Q**, of the Mendel University in Brno. Lunches will be served in MendelU staff's canteen at building **O** (exclusive use for OMGN2022 attendees).



Faculty of AgriSciences - building **C**, **M**; Faculty of Business and Economics - building **Q**; Faculty of Forestry and Wood Technology - building **B**; Faculty of Horticulture - building **A**; Faculty of Regional Development and International Studies – building **Z**; Student Canteen - building **X**; Staff canteen - building **O**.



Transportation from the city centre to the MendelU campus:

- Tram no. 9 (direction Čertova rokle) - runs from the stop Main Railway Station to the stop Zemědělská;
- Bus no. 67 (direction Jundrov) – runs from the stop Main Railway Station to the stop Schodova.

Programme

Day 1: Monday, 22nd August 2022

14:00 - 15:00 Registration (building Q).

15:00 - 17:00 Welcome reception (Botanical Garden and Arboretum).

Day 2: Tuesday, 23rd August 2022

08:00 - 08:20 Poster hanging.

08:25 - 08:35 Opening: Martin Klimánek (Vice-rector of MendelU) and Thomas Jung.

08:35 - 09:50 Session 1 - Effectors, Virulence and Pathogenicity

Chair: Michael Seidl.

08:35 - 08:55 **1.1.** Wenbo Ma: "LWY tandem repeat in *Phytophthora* effectors enables elaborate mimicry of a host phosphatase".

08:55 - 09:10 **1.2.** Enoch Yuen: "Traffic control: A conserved *Phytophthora* effector hijacks a host RabGAP protein to inactivate a Rab GTPase that mediates defense-related secretion".

09:10 - 09:25 **1.3.** Melanie Mendel: "Stayin' alive: *In vivo* functional analysis of effectors in spinach".

09:25 - 09:40 **1.4.** Edouard Evangelisti: "A *Phytophthora* effector interferes with 14-3-3 phosphosensors".

09:50 - 10:20 Break.

10:20 - 11:00 Keynote speaker I - Andrea Sánchez Vallet:

"Natural variation in an *Avr* gene generates a quantitative gene-for-gene phenotype."

11:00 - 12:00 Session 2 - Cell Biology, Signalling and Metabolism I

Chair: Susan Breen.

11:00 - 11:20 **2.1.** Jochem Bronkhorst: "An actin mechanostat ensures hyphal tip sharpness in *Phytophthora infestans* to achieve host penetration".

11:20 - 11:35 **2.2.** Ayelen Tayagui: "Turgor regulation in encysted oomycete zoospores".

11:35 - 11:50 **2.3.** Andrei Kiselev: "Are proteases from the pathogen *Aphanomyces euteiches* important for legumes infection? Answers from multiomics studies".

12:00 - 13:00 Lunch (staff canteen, building O).

13:00 - 15:30 Tour VILLA TUGENDHAT (includes coffee break).

15:30 - 17:30 Session 3 - Host-pathogen Interaction and Resistance Mechanisms I

Chair: Edouard Evangelisti.

15:30 - 15:50 **3.1.** Sophien Kamoun: "Sensor NLR immune proteins activate oligomerization of their NRC helper".

15:50 - 16:05 **3.2.** Amena Khatun: "The plant defensin NaD1 inhibits growth of *Phytophthora* species by interfering with cell wall structure and calcium transport".

16:05 - 16:20 **3.3.** Alexander Guyon: "A broadly colonisation-responsive synaptotagmin interferes with infection by *Phytophthora palmivora*".

16:20 - 16:35 **3.4.** Yacine Badis: "Novel methods and oomycete models for a molecular understanding of Phycopathology".

16:35 - 16:50 **3.5.** Bradley Dotson: "Breeding for better biocontrol symbiosis of *Trichoderma* against *Aphanomyces*".

17:30 - 18:30 Dinner.

18:30 - 20:30 Poster session (with refreshment drinks).

Day 3: Wednesday, 24th August 2022

08:30 - 09:45 Session 4 - Cell Biology, Signalling and Metabolism II

Chair: Laurent Camborde.

08:30 - 08:50 **4.1.** Maja Brus-Szkalej: "*Phytophthora infestans* transglutaminases are necessary for the formation of a healthy cell wall and for successful infection".

08:50 - 09:05 **4.2.** Graham Peers: "Disruption of mitochondrial fatty acid oxidation reduces the infection efficacy of *Phytophthora sojae*".

09:05 - 09:20 **4.3.** Carlotta Lupatelli: "*Phytophthora* zoospores sensing and motion behaviour".

09:20 - 09:35 **4.4.** Michiel Kasteel: "*Phytophthora* zoospores display klinokinetic behaviour in response to a chemoattractant".

09:45 - 10:15 Break.

10:15 - 11:05 Session 5 - Host-pathogen Interaction and Resistance Mechanisms II

Chair: Martin Černý.

10:15 - 10:35 **5.1.** Aurélien Boisson-Dernier and Celso Litholdo: "Plant cell wall integrity mechanisms and oomycete susceptibility, an ancient story?"

10:35 - 10:50 **5.2.** Xiao Lin: "*Solanum americanum* genomes and effectoromics uncover new resistance genes against potato late blight".

10:50 - 11:05 **5.3.** Philip Carella: "Leveraging plant evolution to understand *Phytophthora* infection processes".

11:05 - 11:45 Keynote speaker II - Clive Brasier:

"Progress in understanding breeding systems in the oomycetes".

12:00 - 13:00 Lunch (staff canteen, building O).

13:00 - 14:20 Session 6 - Diversity, Taxonomy and Population Studies I

Chair: Bruno Scanu.

13:00 - 13:20 **6.1.** Thomas Jung: "*Phytophthora*: an ancient, historic, biologically and structurally cohesive and evolutionarily successful generic concept in need of preservation".

13:20 - 13:35 **6.2.** Eleanor Gilroy: "PenSeq of root rot *Phytophthora P. rubi* reveal intraspecies diversity".

13:35 - 13:50 **6.3.** Cristiana Maia: "Diversity and ecological roles of *Halophytophthora*/ *Phytophthora* species in marine and estuarine ecosystems at the Algarve coast of Portugal".

13:50 - 14:05 **6.4.** David Cooke: "Insights from probing Oomycete diversity at different taxonomic scales".

14:05 - 14:20 **6.5.** Jenifer Sundar: "Population genetic analysis of AVR2 in Chinese populations of *Phytophthora infestans*".

14:30 - 14:55 Break.

14:55 - 15:00 Group photo.

15:00 - 18:00 Visit to Mendel Museum (departure at 15.00 from MendelU).

18:00 - 22:00 Conference dinner in the Augustinian Abbey.

Day 4: Thursday, 25th August 2022

08:30 - 09:05 Session 7 - Diversity, Taxonomy and Population Studies II

Chair: Tamara Corcobado.

08:30 - 08:50 **7.1.** Gautam Shirsekar: "Entangled co-evolutionary history of *Hyaloperonospora arabidopsidis* and its host *Arabidopsis thaliana*".

08:50 - 09:05 **7.2.** Vanessa Tremblay: "The evolution of *Phytophthora sojae* pathotypes in Quebec indicates a rapid decline of *Rps* efficiency in soybean".

09:05 - 09:55 Session 8 - Oomycete Genetics and Genomics

Chair: Maja Brus-Szkalej.

09:05 - 09:25 **8.1.** Mahmut Tör: "Fundamental and translational research on downy mildews: reverse genetics, pathogenomics and biologics".

09:25 - 09:40 **8.2.** Petros Skiadas: "Gapless genome assemblies reveal the effector repertoire of the sexually evolving spinach downy mildew".

09:40 - 09:55 **8.3.** Kyle Fletcher: "Using near-complete genome assemblies to uncover new insights of oomycete biology".

10:05 - 10:35 Break.

10:35 - 11:55 Session 9 - Ecology, Metagenomics and Microbial Interactions

Chair: David Hoey.

10:35 - 10:55 **9.1.** Claire Gachon: "Weathering a wave of novel marine oomycetes of ecological and economic relevance".

10:55 - 11:10 **9.2.** Dora Pavić: "Physico-chemical properties of natural waters that affect the sporulation of freshwater pathogenic oomycetes *Saprolegnia parasitica* and *Aphanomyces astaci*".

11:10 - 11:25 **9.3.** Leticia Botella: "*Phytophthora* and *Halophytophthora* spp. are the hosts of multiple viral infections".

11:25 - 11:40 **9.4.** Carren Burkey: "Investigating the molecular basis of pathogenicity by a *Pseudomonas fluorescens* isolate on oomycetes".

11:50 - 13:00 Lunch (staff canteen, building O).

13:00 - 14:30 Session 10 - Host-pathogen Interaction and Resistance Mechanisms III

Chair: Yacine Badis.

13:00 - 13:20 **10.1.** Tolga Bozkurt: "Reprogramming of defense-related trafficking in plants during oomycete infection".

13:20 - 13:35 **10.2.** Maryam Hashemi: "*Pythium oligandrum*: A biocontrol agent with growth promotion and disease resistance properties which does not inhibit mutualistic interactions in legumes".

13:35 - 13:50 **10.3.** Robert Heal: "Dissecting components of *Solanum americanum* non-host resistance to *Phytophthora infestans*".

13:50 - 14:05 **10.4.** Daniel Monino-Lopez: "*Rpi-agf1*, a novel broad-spectrum *R* gene against *P. infestans*, reveals the importance of multivesicular bodies during infection".

14:05 - 14:20 **10.5.** Martin Pettersson: "*Phytophthora* detected in plants imported to Norway".

14:30 - 14:45 Closing remarks and farewell!!!

14:45 - 16:00 Open space for informal conversations and group meetings.

Discussion on open science, future research and funding opportunities.

Poster removal.

ABSTRACTS

Oral presentations

(In order of appearance)

Keynote Speakers

Oral sessions

Session 1 - Cell Biology, Signalling and Metabolism I

Session 2 - Effectors, Virulence and Pathogenicity

Session 3 - Host-pathogen Interaction and Resistance Mechanisms I

Session 4 - Cell Biology, Signalling and Metabolism II

Session 5 - Host-pathogen Interaction and Resistance Mechanisms II

Session 6 - Diversity, Taxonomy and Population Studies I

Session 7 - Diversity, Taxonomy and Population Studies II

Session 8 - Oomycete Genetics and Genomics

Session 9 - Ecology, Metagenomics and Microbial Interactions

Session 10 - Host-pathogen Interaction and Resistance Mechanisms III

Keynote Speaker I

Natural variation in an *Avr* gene generates a quantitative gene-for-gene phenotype

Andrea Sánchez-Vallet

Centro de Biotecnologia y Genomica de Plantas, Universidad Politecnica de Madrid (UPM) – Instituto Nacional de Investigacion y Tecnologıa Agraria y Alimentaria (INIA), Campus de Montegancedo-UPM, Madrid, Spain.

Successful host colonization by plant pathogens requires the circumvention of host defence responses, frequently through sequence modifications in secreted pathogen proteins known as avirulence factors (Avrs). Although Avr sequences are often polymorphic, the contribution of these polymorphisms to virulence diversity in natural pathogen populations remains largely unexplored. We determined how natural sequence polymorphisms of the avirulence factor Avr3D1 in the wheat pathogen *Zymoseptoria tritici* contributed to adaptive changes in virulence and showed that there is a continuous distribution in the magnitude of resistance triggered by different Avr3D1 isoforms. These results demonstrate that natural variation in an *Avr* gene can lead to a quantitative resistance phenotype. We further showed that even homologs of Avr3D1 in two non-pathogenic sister species of *Z. tritici* are recognized by some wheat cultivars, suggesting that Avr-R gene-for-gene interactions may contribute to nonhost resistance. We suggest that the mechanisms underlying host range, qualitative resistance and quantitative resistance are not exclusive.

Keynote Speaker II

Progress in understanding breeding systems in the oomycetes

Clive Brasier

Forest Research, Alice Holt Lodge, Farnham, Surrey GU10 4LH, United Kingdom.

Evolutionarily, oomycetes have inherited distinctive alga-like gametangia (antheridia and oogonia) and gametes. Beyond that, their breeding systems i.e. their tendency to be inbreeding or outcrossing, are a product of their genetic system, mating mechanisms and their ecology or 'life style'. As Sansome demonstrated (1960s) the oomycetes have also inherited a diploid genetic system with gametangial meiosis, which has had a significant role in the development of their mating mechanisms. Mating mechanisms In the *Saprolegniales* (*Achlya*), elucidated by Raper and Barksdale (1950s-70s), are based on hormonal control of antheridial and oogonial development, including both bisexuality and unisexuality, which facilitate outbreeding and inbreeding strategies. Individuals in the *Peronosporales* (Phytophthoras, Downy mildews) are typically bisexual but there are two main breeding strategies among taxa, each with potentially high evolutionarily flexibility. One is self-fertility ('homothallism'), inbreeding but apparently without a barrier to outbreeding between individuals. The other is facultative outcrossing, involving chemical interaction between two compatibility types A1 and A2 ('heterothallism'*), which can result in (i) both cross and self-fertilisation in A1 x A2 pairings; (ii) sporadic self-fertility through combining both compatibility types in the same nucleus; and (iii) in some species selfing of A1s and A2s in response to environmental influences. Sansome (1980s) proposed the A1 and A2 types were controlled by homozygosity and heterozygosity at single locus. She also provided strong evidence that mitotic segregation of the A1 from the A2 heterozygote was restricted by chromosomal reciprocal translocations and also, by inference, balanced lethals. Recently, homozygous and heterozygous alleles have been demonstrated for the probable compatibility locus in *Plasmopora*. Our new challenge is to better understand how an oomycete's 'life style' in its natural ecosystem informs the breeding strategy and therefore its variation and population structure. Likely influences include environmental heterogeneity, parasite pressure, the relative advantage of oospores in survival, and the selective advantage of sexual versus asexual reproduction for infection and dispersal.

* It is exactly 100 years since Ashby (1922) first described compatibility groups in *Phytophthora.*

Session 1 - Effectors, Virulence and Pathogenicity

Oral Presentation 1.1.

LWY tandem repeat in *Phytophthora* effectors enables elaborate mimicry of a host phosphatase

Hui Li[1], Jinlong Wang[2], Jan Sklenar[1], Yufei Li[1], Frank Menke[1], Yanli Wang[2], Wenbo Ma[1]

[1] The Sainsbury Laboratory, University of East Anglia, Norwich Research Park, Norwich, United Kingdom

[2] Institute of Biophysics, Chinese Academy of Sciences, Beijing, China

Phytophthora produces effectors that consist of the "(L)WY" motif in the form of highly organized tandem repeats. While structurally conserved, the (L)WY units are variable in sequences, indicating that they may mediate interactions with specific host molecules. Here, we discovered a specific (L)WY-LWY combination that is present at the amino terminus of multiple *Phytophthora* effectors and enabling direct interactions of these effectors with the protein phosphatase 2A (PP2A) in plant hosts. PP2A is the major Serine/Threonine phosphatase in eukaryotes. A PP2A holoenzyme is composed with three subunits, including the scaffold A and catalytic C subunits that form the core enzyme, and the regulatory B subunit that recruits the core enzyme to dephosphorylate various substrates. We solved the crystal structure of an effector-PP2A complex, which demonstrated that the effector binds to a region in the PP2A A subunit that overlaps with the binding site for endogenous B subunits but with a higher affinity. As such, the effectors efficiently hijack the core enzyme to form functional PP2A holoenzymes. Importantly, although these effectors all harbour the same PP2A-interacting module at their N terminus, they have divergent C-terminal LWY units, which allow them to interact with distinct sets of host proteins. These findings indicate that these effector-PP2A phosphatases have differentiating capacity in substrate-binding. Our results discovered a virulence strategy widely employed by *Phytophthora* in which an essential host phosphatase complex is appropriated to promote disease and highlight protein modularity-based functional diversification in an effector repertoire.

Session 1 - Effectors, Virulence and Pathogenicity

Oral Presentation 1.2.

Traffic control: A conserved *Phytophthora* effector hijacks a host RabGAP protein to inactivate a Rab GTPase that mediates defense-related secretion

Enoch Yuen[1], Edouard Evangelisti[2,3], Frej Tulin[2,4], Sebastian Schornack[2], Tolga Bozkurt[1]

[1] *Department of Life Sciences, Imperial College, London, United Kingdom.*

[2] *Sainsbury Laboratory (SLCU), University of Cambridge, Cambridge, United Kingdom.*

[3] *Laboratory of Phytopathology, Wageningen University & Research, Wageningen, Netherlands.*

[4] *Department of Plant Biology, Carnegie Institution for Science, Stanford, CA 94305, USA.*

Proper positioning of immune components, such as signaling molecules, antimicrobial compounds, and defense-related enzymes, at the right time and right place is a hallmark of plant innate immunity. This relies on coordinated actions by a series of transport regulators that mediate trafficking of defense cargoes in vesicles towards the infection sites. The emerging paradigm is that *Phytophthora* species employ a variety of effectors to reprogram host vesicle transport systems as a key virulence strategy. Despite recent advances, the regulations and functions of vesicle trafficking in plant immunity remain largely unknown. Here we discovered an unprecedented strategy employed by oomycete pathogens to subvert host vesicle trafficking. A conserved RXLR type of effector secreted by *Phytophthora infestans* and *P. palmivora* hijacks a host Rab GTPase activating protein (RabGAP) as a susceptibility factor to manipulate defense-related trafficking. Our proteomics screen, followed by biochemical and cell biology assays, reveal a model in which a RabGAP negatively regulates immunity by inactivating a host Rab GTPase protein that coats defense-related vesicles to mediate their transport towards the pathogen interface. Intriguingly, the RabGAP protein is guarded by plant NLR type of immune receptors against effector manipulation, whereas *P. infestans* has evolved an effector variant that suppresses this immune recognition. Our work sheds light on defense-related secretion in plants by identifying a negative regulatory mechanism governed by the RabGAP-Rab interactions, that is safeguarded by host immune receptors against pathogen manipulation.

Session 1 - Effectors, Virulence and Pathogenicity

Oral Presentation 1.3.

Stayin' alive: *In vivo* functional analysis of effectors in spinach

Melanie Mendel[1], Femke van den Berg[1], Xander Zuijdgeest[2], Petros Skiadas[1,3], Joyce Elberse[1], Michael F. Seidl[3], Guido Van den Ackerveken[1], Ronnie de Jonge[1]

[1] Plant-Microbe Interactions, Utrecht University, Padualaan 8 3584 CH, Utrecht, The Netherlands.

[2] ZMBP Plant Physiology, University of Tübingen, Auf der Morgenstelle 32, D-72076 Tübingen, Germany.

[3] Theoretical Biology and Bioinformatics, Utrecht University, Padualaan 8 3584 CH, Utrecht, The Netherlands.

Effectors play a critical role in determining the outcome of host-pathogen interactions. We focus on the infection of the spinach downy mildew *Peronospora effusa* (*Pe*) which threatens spinach production worldwide. Emerging new *Pe*-races call for effective management strategies. Therefore, we must improve our knowledge of the *Pe* effector repertoire, specifically effector functions and evolution. Previously, classic *Agrobacterium tumefaciens* based screens for cell-death induction by *Pe* in spinach failed. In combination, with the obligate biotrophic lifestyle of *Pe*, functional analysis of *Pe* effectors on its host spinach are challenging. We set-out to establish *Pseudomonas*-based effector-screens to understand the contribution of effectors to pathogen virulence in spinach. We developed spinach-assays using a *Pseudomonas syringae pv. tomato DC3000* (*Pst DC3000*) based effector library. Our virulence screening combines reactive oxygen species (ROS) assays, bacterial proliferation assays and cell-death assays. Collectively, they can provide a starting point for evaluating the role of a given effector in pathogen virulence. The *Pst DC3000* effectors AvrE1 and HopM1 had a striking influence on the assessed virulence-characteristic of *Pseudomonas* in spinach. AvrE1/HopM1 ensure stomatal closure in *Arabidopsis thaliana* during *Pst DC3000* infection by alternating abscisic acid levels. Translating our findings to *Pe* virulence will shed a first light on effector function in spinach. In a next step, we will adapt the screens for *Pe* effectors in spinach to enable a better understanding of *Pe* effectors on their host plant. Improving our understanding of how *Pe* effectors shape pathogen virulence can help us to breed new and more resistant plants.

Session 1 - Effectors, Virulence and Pathogenicity

Oral Presentation 1.4.

A *Phytophthora* effector interferes with 14-3-3 phosphosensors

Edouard Evangelisti[1], Liron Shenhav[2], Sebastian Schornack[2]

[1] *Wageningen University & Research, Wageningen, The Netherlands.*

[2] *The Sainsbury Laboratory, Cambridge University, Cambridge, United Kingdom.*

The cacao killer *Phytophthora palmivora* is a tropical plant pathogen that causes disease in more than a hundred plant species. One of the most highly expressed *P. palmivora* effectors, FIRE, comprises a canonical binding motif to 14-3-3 phosphosensors in their C-terminal domain. FIRE interacts with multiple 14-3-3 proteins in a phosphorylation-dependent manner. The effector promotes plant susceptibility to *P. palmivora*, is mainly expressed at the biotrophic stage and co-localises with its target around haustoria. Our findings demonstrate that plant phosphosensors are not exclusively targeted by bacterial effectors. Instead, 14-3-3 protein-binding is a cross-kingdom infection strategy shared by prokaryotic and eukaryotic plant pathogens to suppress host immunity.

Session 2 - Cell Biology, Signalling and Metabolism I

Oral Presentation 2.1.

An actin mechanostat ensures hyphal tip sharpness in *Phytophthora infestans* to achieve host penetration

Jochem Bronkhorst[1†], Kiki Kots[2,3†], Djanick de Jong[1], Michiel Kasteel[2,3], Thomas van Boxmeer[3], Tanweer Joemmanbaks[3], Francine Govers[2], Jasper van der Gucht[1], Tijs Ketelaar[3*], Joris Sprakel[1,4*]

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[2] *Laboratory of Phytopathology, Wageningen University & Research, Droevendaalsesteeg 1, 6708 PB Wageningen, Netherlands.*

[3] *Laboratory of Cell Biology, Wageningen University & Research, Droevendaalsesteeg 1, 6708 PB Wageningen, Netherlands.*

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† These authors contributed equally to this work as co–first authors.

Filamentous plant pathogens apply mechanical forces to pierce their hosts surface and penetrate its tissues. Devastating *Phytophthora* pathogens harness a specialized form of invasive tip growth to slice through the plant surface, wielding their hypha as a microscopic knife. Slicing requires a sharp hyphal tip that is not blunted at the site of the mechanical interaction. How tip shape is controlled, however, is unknown. We uncover an actin-based mechanostat in *Phytophthora infestans* that controls tip sharpness during penetration. Mechanical stimulation of the hypha leads to the emergence of an aster-like actin configuration, which shows fast, local, and quantitative feedback to the local stress. We evidence that this functions as an adaptive mechanical scaffold that sharpens the invasive weapon and prevents it from blunting. The hyphal tip mechanostat enables the efficient conversion of turgor into localized invasive pressures that are required to achieve host penetration.

Session 2 - Cell Biology, Signalling and Metabolism I

Oral Presentation 2.2.

Turgor regulation in encysted oomycete zoospores

Ayelen Tayagui[1,2,3], Nicola Lacalendola[3,4], Matthew Ting[3,5], Jenny Malmstrom[3,5], Volker Nock[1,3], Geoff R. Willmott[3,4,6], Ashley Garrill[2]

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[2] *School of Biological Sciences, University of Canterbury, Christchurch, New Zealand.*

[3] *The McDiarmid Institute for Advanced Materials and Nanotechnology, Wellington, New Zealand.*

[4] *Department of Physics, The University of Auckland, Auckland, New Zealand.*

[5] *Department of Chemical and Materials Engineering, The University of Auckland, Auckland, New Zealand.*

[6] *School of Chemical Sciences, The University of Auckland, Auckland, New Zealand.*

Zoospores are motile, asexual reproductive propagules that enable oomycete pathogens to locate and infect new host tissue. Once they locate host tissue, they encyst, enabling the generation of turgor pressure that will make them stiffer. This, in turn, provides the driving force for germination and invasion of the host. It is not known how spores respond to osmotic stresses that might arise due to different environments on and around their hosts and how this may impact turgor. This presentation will describe microaspiration and atomic force microscopy measurements on encysted zoospores of *Achlya bisexualis* to investigate their mechanical properties and how these change after hyperosmotic stress. With a small hyperosmotic stress (media osmolality changes < 295.6 mOsmol/kg), zoospores initially became stiffer with an increase in the Young's modulus (*E*) over 30 mins from 0.16 MPa to 0.25 MPa. *E* then returned to its original value after 120 min. With a greater osmotic stress (media osmolality changes of 438.2 - 787.6 mOsmol/kg) the reverse occurred, with an initial decrease in E over 30 – 60 mins down to 0.08 MPa, before recovery to the original value after 120 min. The responses are consistent with rapid changes in cell wall thickness and a turgor regulation mechanism. Turgor regulation is further supported by the shrinkage and retraction of the protoplast from the cell wall and its subsequent re-expansion after hyperosmotic challenge. As far as we are aware this is the first demonstration of turgor regulation, not just in encysted zoospores, but in oomycetes in general.

Session 2 - Cell Biology, Signalling and Metabolism I

Oral Presentation 2.3.

Are proteases from the pathogen *Aphanomyces euteiches* important for legumes infection? Answers from multiomics studies

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Aphanomyces euteiches is a pathogenic oomycete belonging to Saprolegniales. In the field it causes the root rot disease of legume crops as pea and alfalfa and interacts in the lab with the legume model *Medicago truncatula*. During infection, *A. euteiches* secretes myriad of proteins called effectors that modulate host physiology and promote disease. To decipher the secretome composition of *A. euteiches*, we performed comparative genomic analyses using long-read assembly of *A. euteiches* isolated from pea field and Illumina assemblies of four strains with specificity to pea or alfalfa. We focused on the identification of the core and specific effectors, which could explain the legume preference of *A. euteiches.* Secreted proteases and Carbohydrate-Active Enzymes (CAZymes) appeared to be a determinant of a core effectome whereas, small secreted proteins (SSP) appeared to play a role in strain specialisations. To check whether microbial secreted proteases act as virulence factor during roots colonisation, we aimed to identify the active forms during pea infection. By using a combination of transcriptomics and activity-based proteome profiling (ABPP) approaches, 26 serine and cysteine proteases from *A. euteiches* were detected in the apoplastic fluid of the infected pea roots.

Session 3 - Host-pathogen Interaction and Resistance Mechanisms I

Oral Presentation 3.1.

Sensor NLR immune proteins activate oligomerization of their NRC helper

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Intracellular nucleotide-binding domain and leucine-rich repeat (NLR) immune receptors are important components of plant innate immunity. They perceive pathogen virulence effectors, and subsequently mediate immune signaling and disease resistance. NLRs can function as individual units, known as singletons, mediating both effector perception and downstream signaling. Upon activation, singleton NLRs form multiprotein homo-complexes termed resistosomes. However, many NLRs operate in higher-order configurations, such as pairs or networks. In these cases, a sensor NLR mediates pathogen perception and requires a second helper NLR to initiate immune signaling. In solanaceous plants, NRCs (NLRs required for cell death) are central helper nodes in a complex NLR network that mediates resistance against diverse plant pathogens. While the genetics that underpin the NRC network are known, our biochemical understanding of networked NLR activation is limited. In this work, we established a Blue Native polyacrylamide gel electrophoresis to study the precise mechanisms of NLR activation in the NRC network using the NRC-dependent sensor NLR Rx and its downstream helper NRC2 as an experimental system. We show that upon pathogen perception, Rx mediates homo-oligomerization of NRC2 but does not associate with the NRC2 resistosome. The activated NRC2 resistosome accumulates at the host plasma membrane, separate from Rx. Finally, we show that a nematode effector with NRC2 suppression capacities can block NRC2 oligomerization and plasma membrane association to suppress immune signaling. Our work establishes an activation-and-release model for sensor-helper activation of NLRs in the NRC network and has implication for resistance of asterid plants to oomycete pathogens.

Session 3 - Host-pathogen Interaction and Resistance Mechanisms I

Oral Presentation 3.2.

The plant defensin NaD1 inhibits growth of *Phytophthora* species by interfering with cell wall structure and calcium transport

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The *Nicotiana alata* Defensin 1 (NaD1) peptide has antifungal activity but its mode of action is unknown. Here we investigated whether NaD1 exerts similar activity against oomycetes and used multiple approaches to shed light on its mode of action. The defensin was tested against four different *Phytophthora* species and exposure to the peptide led to suppression of apical dominance, cytoplasmic granulation and hyper-branching. Previous studies in fungi showed that these alterations are linked to Ca^{2+} channel inhibition. Interestingly, our data show that inhibition of hyphal growth in *Phytophthora cinnamomi* by NaD1 can be rescued by addition of extracellular Ca^{2+}, while potassium was ineffective. In addition, analysis of cytosolic Ca^{2+} by fluorescence microscopy using Fluo-3 AM revealed calcium homeostasis disruption by NaD1. RNA sequencing experiments were consistent with this observation, showing decreased expression of Ca^{2+} transport proteins in treated hyphae. Moreover, scanning and transmission electron microscopy revealed changes in cell wall thickness and morphology suggesting altered composition or ultrastructure. Glycosidic linkage analysis conducted on NaD1-treated hyphae revealed a significantly decreased cellulose content compared to untreated control samples. Our data suggest that NaD1 inhibits growth of *Phytophthora* species by interfering with intracellular Ca^{2+} homeostasis and modifying cell wall biosynthesis and properties.

Session 3 - Host-pathogen Interaction and Resistance Mechanisms I

Oral Presentation 3.3.

A broadly colonisation-responsive synaptotagmin interferes with infection by *Phytophthora palmivora*

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Interactions between plants and microbes, both symbiotic and pathogenic, are hugely important for plant fitness with large implications for crop yields and food security. A crucial part of biotrophic interactions between the host plant and filamentous microbes is the formation of membrane interfaces, such as haustoria or arbuscules, within living plant cells at which both organisms deploy proteins for nutrient exchange, defence, and communication. I aim to identify the extent to which intracellular interfaces formed by mycorrhizal fungi and pathogenic oomycetes in roots and shoots share the same plant components and processes. To identify protein constituents of interfaces, I hypothesise that transcripts commonly induced in root interactions with unrelated filamentous microbes contribute to the establishment and maintenance of interfaces. Using this approach in *Nicotiana benthamiana*, a synaptotagmin (SYT) was identified. When constitutively expressed in *N. benthamiana* leaves, SYT localised to putative endoplasmic reticulumplasma membrane contact sites and to *Phytophthora palmivora* haustoria suggesting it may act at the intracellular interface. Constitutive expression of SYT reduced the necrotic lesion area caused by *P. palmivora* leaf infection. This phenotype may be caused by increasing defence or by disrupting a finely balanced system of synaptotagmin dimers and therefore impairing colonisation. Expanding our approaches into arbuscular mycorrhizal symbiosis will inform broad applicability against a range of filamentous microbes.

Session 3 - Host-pathogen Interaction and Resistance Mechanisms I

Oral Presentation 3.4.

Novel methods and oomycete models for a molecular understanding of Phycopathology

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The marine environment harbours a high number of intracellular oomycete parasites that must rely on secreting effectors to infect their diverse phytoplanktonic and seaweed hosts. Seaweeds are of considerable interest for both fundamental and applied research: their aquaculture develops exponentially worldwide and suffers severe losses due to pathogens, highlighting the need for better farming practices and resistant cultivars. Yet, there is very little molecular understanding of the immune system of seaweeds, which currently hinders modern breeding approaches. Here, we will report our latest progress in establishing molecular methods and novel pathosystems using oomycetes infecting brown and red algae. Importantly, these marine oomycetes are very distant (i.e. early diverging) from the plant pathogenic models investigated to date, and we found no evidence of classical oomycete effector families in transcriptomic data. We will present putative effector candidates of the brown algal oomycete endoparasite *Eurychasma dicksonii*, and discuss how applying effectomics to these emerging pathosystems could drive the elucidation of presently unknown macroalgal immune systems.

Session 3 - Host-pathogen Interaction and Resistance Mechanisms I

Oral Presentation 3.5.

Breeding for better biocontrol symbiosis of *Trichoderma* against *Aphanomyces*

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The oomycete genus of *Aphanomyces* is the causal agent for several plant diseases. *Aphanomyces cochlioides* preferentially attacks plants from the Amaranthaceae clade, e.g. beets, spinach, and quinoa. In *Beta vulgaris subsp. vulgaris* (sugar beet), *A. cochliodies* causes two distinct diseases at different life stages of the plant and both cause significant yield losses. Natural parasites of oomycetes, such as the fungi in the genus of *Trichoderma*, have been used previously to improve several crop diseases. However, the links between host mechanisms of disease control and the mechanisms of biocontrol and biostimulation by beneficial microbes such as *Trichoderma* are poorly understood. We investigated the effectiveness of the biocontrol agent and plant symbiont *T. afroharzianum* T22 against *A. cochliodies* using breeding lines of sugar beet. Notably, T22 was effective at reducing disease symptoms in a subset of lines. Surprisingly in other lines we observed enhanced disease symptoms after treatment with T22. Furthermore, we also observed significant variation in growth promotion (positive symbiosis) of the T22 within the sugar beet elite breeding lines, leading us to hypothesize that potentially independent host plant genetic factors may be important for the success of symbiotic microbes as plant symbionts, biocontrol agents and for biostimulation. Future work will focus on the identification of these plant genetic factors and their incorporation into crop breeding programs, with the ultimate goal to breed crop plants for better interactions with symbionts.

Session 4 - Cell Biology, Signalling and Metabolism II

Oral Presentation 4.1.

Phytophthora infestans transglutaminases are necessary for the formation of a healthy cell wall and for successful infection

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Transglutaminases (TGases) are highly conserved enzymes that play an important role in protein cross-linking. One of the putative TGases of *Phytophthora infestans* has previously been shown to be localised to the cell wall; and the gene encoding this protein to be highly expressed during development of the infectious structures and at early infection time points. Based on sequence similarity we were able to identify six more genes annotated as putative TGases and show that these seven genes group together in phylogenetic analysis. All of the seven proteins are predicted to contain transmembrane helices and both a TGase domain and a MANSC domain, the latter of which was previously shown to play a role in protein stability. Chemical inhibition of transglutaminase activity (demonstrated in an enzymatic assay) and silencing of the entire family of the putative cell wall TGases are both lethal to *P. infestans*. Extensive bursting observed in the few recovered lines indicates the importance of these proteins in cell wall formation and stability. The intermediate phenotype obtained with lower drug concentrations and less efficient silencing displays a number of deformations to germ tubes, especially at the growth tip. Both chemically treated and silenced lines show lower pathogenicity than the wild type in detached leaf infection assays. Finally, turgor pressure, estimated by cell cytorrhysis, was lower in germ tubes treated with the TGase inhibitor.

Session 4 - Cell Biology, Signalling and Metabolism II

Oral Presentation 4.2.

Disruption of mitochondrial fatty acid oxidation reduces the infection efficacy of *Phytophthora sojae*

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The zoospores of *Phytophthora* contain considerable amounts of triacylglycerol (TAG) and it was presumed that this provides energy to support motility and catabolism during plant infection. Our group previously demonstrated that the photosynthetic diatom *Phaeodactylum tricornutum* utilizes a Stramenopile-specific mitochondrial Acyl-CoA Dehydrogenase (ACAD) for beta-oxidation of TAG at night. We reasoned that the orthologous pathway in oomycetes may be important to their physiology. Deletion of the ACAD gene in *P. sojae* resulted in no changes to mycelial growth rates on V8 medium. However, 3 independent mutant lines showed ~50% reduction in cyst germination rates compared to WT. Additionally, the area of lesions on soybean leaves was severely reduced on plants infected with an ACAD knockout strain compared to a WT strain. Preliminary fluorescence imaging also suggests a hyper-accumulation of TAG in the knockout vs. WT. Together, these data suggest that catabolism of endogenous TAG is required for effective transition through the cyst-zoospore-infection steps of the oomycete life cycle.

Session 4 - Cell Biology, Signalling and Metabolism II

Oral Presentation 4.3.

Phytophthora zoospores sensing and motion behavior

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The epidemic spread of Phytophthora diseases is primarily based on dispersal of unicellular, bi-flagellated zoospores in the soil. Recent studies suggest that guidance factors included those emitted by soil particles and host plants orchestrate the early events of zoospores root colonization. However, the mechanisms underlying zoospores perception, resulting in the directed motion, remain to be elucidated. Proteins in the plasma membrane play a fundamental function during plant-pathogen interactions, e.g., the stimuli perception by pathogens. Lack of detailed information on membrane proteins in Phytophthora zoospores prevents a comprehensive understanding of its pathogenic interaction with host surface. Firstly, we aim to investigate the membrane protein content of *Phytophthora parasitica* zoospore, distinctly isolated from cell body and flagella. Detection of peptides by LC-MS/MS approach and identification of related-proteins by mapping against the *P. parasitica* reference proteome will enable to characterise and compare the membrane repertoire of both zoospores cell body and flagella fractions. Secondly, we will reveal the biochemical mechanisms and the physical forces governing the directed motion of *P. parasitica* zoospores. Pharmacological and physical approaches are developed to functionally characterise our proteomic findings and to analyse zoospores motion metrics (velocity, cell rotation and flagellar beating) under different experimental conditions. Finally, we have developed a live-cell analysis method that allows, at the microenvironmental scale, to specifically discriminate the effect of distinct external stimuli on zoospores sensing and motility capability in comparison with other rhizospheric species. Together, these results will improve our understanding on the sensing events and motion responses governing plant-infecting zoospores.

Session 4 - Cell Biology, Signalling and Metabolism II

Oral Presentation 4.4.

Phytophthora zoospores display klinokinetic behaviour in response to a chemoattractant

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Phytophthora infestans, the causal agent of potato late blight, makes use of dispersal agents called zoospores to rapidly spread and infect. Being motile, these zoospores have been thought to actively track down their hosts using chemical cues such as sugars, amino acids and isoflavonoids. In this study, we used high speed cameras to track zoospores over time and have quantified key trajectory parameters to describe their response to glutamic acid (Glu). We find zoospores to adapt their native run-and-tumble state in response to Glu by greatly increasing the frequency at which they turn. When simulated, we find tuneable tumble frequencies to be sufficient to explain aggregation, implying zoospores to have access to a klinokinetic accumulation strategy to aggregate. We used the same experimental set-up to monitor zoospores of a mutant compromised in heterotrimeric G-protein signalling, and show that their aberrant swimming behaviour is not due to a defect in Glu-chemotaxis, but to aberrantly high and consistent tumbling frequencies.

Session 5 - Host-pathogen Interaction and Resistance Mechanisms II

Oral Presentation 5.1.

Plant cell wall integrity mechanisms and oomycete susceptibility, an ancient story?

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Growing plant cells tightly coordinate the loosening and turgor pressure-driven deformation of their pre-existing cell wall (CW) with the precise delivery of new membrane and CW material. To achieve such remarkable coordination in time and space, plant cells have developed CW integrity mechanisms to relay information about CW performance to their intracellular growth machinery. In the higher plant model Arabidopsis, we previously revealed CW integrity mechanisms that control tip-growth of pollen tubes and root hairs and are governed by the Malectin-like receptor kinase (MLR) subfamily members AtANXUR1/2 (AtANX1/2) and AtFERONIA (AtFER), respectively, and the receptor-like cytoplasmic kinase AtMARIS (MRI). Loss-of-function mutants for these signalling components lead to growing plant cells that burst spontaneously. Moreover, we showed that these pathways are conserved in the tip-growing rhizoids of the basal land plant *Marchantia polymorpha*. Mutants in the Marchantia single-copy orthologues of AtFER and AtMRI, MpFER and MpMRI, display rhizoids that also burst during growth.

Interestingly, the absence of the CW integrity receptor AtFER was shown to render Arabidopsis mutant plants resistant to infection by some parasitic pathogens indicating that CW integrity mechanisms are subverted by parasitic pathogens to establish disease. In good agreement, we will present our recent transcriptomics and proteomics analyses of the Mp*fer* and Mp*mri* mutants showing a remarkable overlap of the differentially expressed genes and enriched proteins between the CW integrity mutants and the wild-type Marchantia infected with *Phytophthora palmivora*. We will discuss the phenotypic responses of the Marchantia CW integrity mutants in response to *P. palmivora*.

Session 5 - Host-pathogen Interaction and Resistance Mechanisms II

Oral Presentation 5.2.

Solanum americanum genomes and effectoromics uncover new resistance genes against potato late blight

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Potato late blight triggered the Irish famine and remains a problem today. The causal agent is the notorious oomycete pathogen *Phytophthora infestans* – the plant and *R* gene destroyer. *Solanum americanum* (Sam) is a globally distributed, wild Solanaceae plant. Most *S. americanum* accessions are highly resistant to late blight, making it an ideal material for cloning of *R* genes. Here we generated high-quality reference genomes of four Sam accessions, re-sequenced 52 accessions, and constructed its NLRome. We further screened for an interactome of ~300 *P. infestans* RXLR effectors with 52 Sam accessions. Using these genotypic and phenotypic data, we cloned three novel *Rpi* genes that can recognize RXLR effectors from *P. infestans*. Transgenic and knockout analysis in Sam, *N. benthamiana* or potato confirmed their function in response to *P. infestans* infection. This study allows us to understand the effector triggered immunity (ETI) landscape of *S. americanum* and *P. infestans*, and we developed a pipeline that enables fast *R* gene cloning from wild crop relatives, that will ultimately assist breeders to turn potato into a "nonhost" plant of late blight.

Session 5 - Host-pathogen Interaction and Resistance Mechanisms II

Oral Presentation 5.3.

Leveraging plant evolution to understand *Phytophthora* infection processes

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The colonization of plant tissues by filamentous pathogens requires a complex compendium of virulence factors capable of modulating key host immune and cellular processes. This paradigm is extensively explored during host-pathogen interactions within evolutionarily young land plant lineages (angiosperm flowering plants). By comparison, our knowledge of plant-microbe interactions across a greater diversity of green plants has remained extremely limited. To better understand the core processes underpinning plant colonization by filamentous pathogens, we leverage the broad host range of the hemi-biotrophic oomycete *Phytophthora palmivora* to challenge an extended range of green plants, from the early divergent bryophytes (non-vascular, non-seed) to angiosperms. We reveal a comparable *P. palmivora* infection program centred on secreted virulence factors as well as conserved host responses during disease. Collectively, this work sheds new light on the keystone weapons employed by a broad host pathogen in addition to conserved responses to disease across distantly-related hosts.

Session 6 - Diversity, Taxonomy and Population Studies I

Oral Presentation 6.1.

Phytophthora: an ancient, historic, biologically and structurally cohesive and evolutionarily successful generic concept in need of preservation

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The 20 genera of obligately biotrophic, angiosperm-foliage specialised DMs evolved from *Phytophthora* at least twice via convergent evolution, making the DMs as a group polyphyletic and *Phytophthora* paraphyletic in cladistic terms. Although such paraphyly is common in successful organisms a proposal has been made to split the genus into multiple new genera. We have reviewed the status of *Phytophthora* and its relationship to the DMs. In our assessment of 196 *Phytophthora* species, distributed across twelve major clades in a relatively tight monophyletic cluster, for twenty morphological and behavioural criteria the clades show good biological cohesion. Saprotrophy, necrotrophy and hemi-biotrophy of woody and non-woody roots, stems and foliage occurs across the clades. Phylogenetically less related clades often show strong phenotypic and behavioural similarities and no one clade or group of clades shows synapomorphies that might justify a unique generic status. The proposal to divide *Phytophthora* appears more a device to address the issue of the convergent evolution of the DMs than the structure of *Phytophthora per se*. We consider it non-Darwinian, putting the emphasis on the emergent groups (the DMs) rather than the progenitor (*Phytophthora*) and ignoring the evolutionary processes that gave rise to the divergence. Further, the generic concept currently applied to the DMs is narrower than that between some closely related *Phytophthora* species. Considering the biological and structural cohesion of *Phytophthora*, its historic and social impacts and its importance in scientific communication and biosecurity protocol, we recommend that the current broad generic concept is retained by the scientific community.

Session 6 - Diversity, Taxonomy and Population Studies I

Oral Presentation 6.2.

PenSeq of root rot phytophthora *P. rubi* reveal intraspecies diversity

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Phytophthora fragariae and *P. rubi* are two very closely related oomycete crop pathogens that are limited to distinct Rosaceae host species. Both pathogens have driven soft growers out of the soil and into pot and substrate-based growing systems. PCR-based amplification of particular house keepers often struggle to distinguish *P. fragariae* and *rubi* and failed to detect intraspecies diversity.

Here we deployed enrichment sequencing of pathogen derived housekeepers and virulence factors (PathSeq) using bespoke RNA baits designed from available *P. fragariae* and *P. rubi* genomes. Here we successfully performed Pathseq on gDNA from 20 *P. rubi* and 4 *P. fragariae* isolates. Significant presence/absence and sequence polymorphisms across multiple individuals was found across *P. rubi*. PenSeq facilitated a cost-effective and statistically powerful population genomic study on the effector genes underlying pathogenicity and host adaptation. K-mer analysis of the Penseq dataset allowed significant diversity between the isolates to be visualised, including clusters from cross continental demographics suggesting there could have been several introductions between UK and Europe. Consequently we can inform growers of the potential diversity within the *P. rubi* population and highlights the future challenge to the durability of deployed and future host resistances.

Session 6 - Diversity, Taxonomy and Population Studies I

Oral Presentation 6.3.

Diversity and ecological roles of *Halophytophthora/Phytophthora* species in marine and estuarine ecosystems at the Algarve coast of Portugal

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During a small-scale survey at the Algarve coast in Portugal, a high diversity of *Phytophthora* and *Halophytophthora* spp., including eight new *Halophytophthora* species, was discovered. The latter have been recently described as *H. thermoambigua*, *H. lusitanica*, *H. lateralis*, *H. frigida*, *H. sinuata, H. macrosporangia, H. brevisporangia* and *H. celeris*, based on multigene phylogenetic analyses, growth-temperature relationship tests and morphological and morphometric studies. *Halophytophthora* species are oomycetes inhabiting marine and estuarine ecosystems from the tropics to temperate regions. Despite being described as saprophytes playing an important role on litter decomposition due to their ability to rapidly colonize fallen leaves, they might also act as pathogens under certain conditions. The genus *Halophytophthora* is closely related to *Phytophthora*, which contains notorious pathogens of numerous terrestrial plant species. Some *Phytophthora* species like *Phytophthora gemini* and *P. inundata* have also been found in marine environments and may be involved in the widespread decline of the seagrass *Zostera marina.* In order to clarify the ecological role of the eight new *Halophytophthora* species, *H. avicennae* and *Phytophthora inundata*, their saprophytic ability was assessed in a decomposition experiment with fresh and air-dried leaves of *Z. marina* and *Cymodocea nodosa*. The decomposition of the leaves was evaluated by recording the mass loss every four weeks and the trial was concluded after three months. The results of this experiment and the seagrass decomposition capacity of the species tested will be discussed.

Session 6 - Diversity, Taxonomy and Population Studies I

Oral Presentation 6.4.

Insights from probing Oomycete diversity at different taxonomic scales

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The genus *Phytophthora* comprises more than 200 species of oomycete plant pathogen that cause significant crop damage and losses in agriculture, horticulture, and forestry in addition to their impact on plants in natural ecosystems. At the James Hutton Institute we have been using DNA-based methods to study *Phytophthora* diversity at two extremes of the taxonomic scale. Firstly, populations of the potato late blight pathogen, *Phytophthora infestans,* from European crops have been genotyped using simple sequence repeat markers. Although both mating types are frequently found in UK crops, our long-term survey has shown this heterothallic species has a predominantly clonal population with sexual recombinants sampled in relatively limited and localised populations. We will discuss the genetics, ecology and factors that drive population change in *P. infestans*. At a wider taxonomic scale, populations have been examined using an rDNA ITS-based metabarcoding method that detects and identifies the species of *Phytophthora*, *Nothophytophthora* and downy mildew present in environmental DNA (eDNA) from an ecosystem. Careful use of controls and a detailed analysis pipeline have been used to process hundreds of water eDNA samples from natural ecosystems in Scotland. Barcodes representing more than 60 known and unknown *Phytophthora* taxa have been detected, providing an insight into the diversity, ecology and biology of populations of this pathogen in natural vegetation. The contrasting inter and intra-specific genetic diversity of populations of *Phytophthora* at these scales will be examined.

Session 6 - Diversity, Taxonomy and Population Studies I

Oral Presentation 6.5.

Population genetic analysis of AVR2 in Chinese populations of *Phytophthora infestans*

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Avoiding host detection and maintaining virulence are crucial for pathogen fitness, which may lead to rapid sequence diversification in the pathogen to overcome host resistance. One such example is the fast-evolving pathogen *Phytophthora infestans*, the causal agent of potato late blight. *P. infestans* secretes an arsenal of effector molecules, designed to promote infection and overcome or subvert host resistance responses, including those triggered by *R* genes. Effector genes are thus subject to selection pressure and may rapidly evolve in *P. infestans* populations. Effector evolution can depend on geographical separation but also be linked to adaptation to disease pressure and environmental elements. Understanding the effect of these factors can help predict effector adaptation to an ever-changing climate and to develop sustainable breeding strategies against late blight. We focused on the widely studied AVR2 effector in clonal Chinese populations across 18 provinces, known to differ in climate. Through sequence alignment and haplotype reconstruction, we found selection on AVR2 in only 3 provinces. Our haplotype network confirms previously published results using a very small data set. Almost all mutations in these haplotypes are single nucleotide polymorphisms (SNPs). We aim to understand if climate or provincial borders affect evolution in these provinces and whether the SNPs result in synonymous or non-synonymous changes, i.e. whether they change the expressed protein structure and/or function. Thus, we conclude that effector genes such as AVR2 can also be a useful tool by which to study variation at the population level.

Session 7 - Diversity, Taxonomy and Population Studies II

Oral Presentation 7.1.

Entangled co-evolutionary history of *Hyaloperonospora arabidopsidis* and its host *Arabidopsis thaliana*

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Hyaloperonospora arabidopsidis (*Har*) is an obligate biotrophic pathogen that causes downy mildew on *Arabidopsis thaliana* (*Ath*). Molecular biology of Arabidopsis-downy mildew interaction has revealed several key players in plant immunity and pathogen virulence. Despite these advances, how genetic diversity in *Har* maps onto genetic diversity of *Ath* through their presumably shared co-evolutionary history remains completely unknown. Here we report the first range-wide collection of *Har* (over 300 collection sites in 16 European countries) that was whole genome-sequenced along with the host *Ath* individuals. *Har* population structure analysis in the spatial framework reveals presence of three ancestral lineages. Demographic inference using sequentially Markovian coalescence (SMC)-based and site-frequency spectrum (SFS)-based approaches on the ancestral lineages of both the *Har* and *Ath* demonstrates almost contemporaneous population splits and migration events in this highly co-evolved pathosystem. We used parameters from demographic inference to perform forward simulations to compare with the observed patterns of genetic diversity in *Har* and *Ath.* Our results show that many known and putative *Har* effector loci have undergone episodes of balancing selection through negative-frequency dependent selection (NFDS). Further, signatures of NFDS-dependent balancing selection on cognate *Ath* immune receptors of known *Har* effectors strongly suggests entangled co-evolutionary history of *Har-Ath* pathosystem.

Session 7 - Diversity, Taxonomy and Population Studies II

Oral Presentation 7.2.

The evolution of *Phytophthora sojae* pathotypes in Quebec indicates a rapid decline of *Rps* efficiency in soybean

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Phytophthora sojae is one of the major parasites of soybean. The deployment of *Rps* (resistance to *Phytophthora sojae*) genes in soybean has been known to provide effective resistance against *P. sojae* infection. However continuous interaction between *P. sojae* and soybean in nature leads to the mutation of avirulence genes and hence the evolution of the virulence profile (pathotype) of *P. sojae* populations. In this work, we used a recently developed molecular assay to assess the pathotypes of *P. sojae* isolates obtained throughout the province of Québec, Canada. The molecular tool showed an equivalent prediction of the pathotypes as a phenotyping assay. Following the analysis of 182 isolates obtained from 2018 to 2021, 27 different pathotypes were detected and pathotype 1a, 1c, 1d was predominant. Overall, the results showed that more than 95% and 85% of the isolates carried pathotype 1a or 1c, respectively, suggesting that *Rps1a* and *Rps1c* were no longer effective in Québec. Virulence on *Rps1k* was present only in 18% of the isolates in 2018 and rose to 38% in 2021. Additionally, there is also an increase in the use of soybean varieties with *Rps1k* from 3% to 30% over the four years in Quebec province. Field experiments have shown that yield can be increased by up to 15% when an effective *Rps* is used in a field where the pathotype is known. While highlighting an easier and more precise technology to assess pathotypes, this study stresses the importance of proper management of *Rps* genes against pathotype evolution of *P. sojae*.

Session 8 - Oomycete Genetics and Genomics

Oral Presentation 8.1.

Fundamental and translational research on downy mildews: Reverse genetics, pathogenomics and biologics

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Downy mildews are well known plant pathogens and we have been carrying out both fundamental and translational collaborative research on some of them. In one project, we have been using the model system of *Arabidopsis-Hyaloperonospora arabidopsidis (Hpa)* to carry out fundamental research using an sRNA-based pipeline for a high-throughput genetic screen for identification and analysis of genes specifically involved in germination, infection, mycelial development and sporulation, nutrient uptake, and host immune suppression. We have already selected some target genes and our preliminary investigations show that the sRNA approach will help us reveal the role of genes in *Hpa* infection and development. In another project, we are carrying out translational research on the pulse-downy mildew pathosystem, caused by the *Peronospora viciae* f.sp. *pisi* (*Pvp*), on pea, and *P.viciae* f.sp. *fabae* (*Pvf*), on broad beans, for translational science. Here, we aim to identify new *R*-genes, develop tools for the accurate detection and diagnostics of *Pvp/Pvf* isolates using genomics, and use biological control agents to suppress downy mildew pathogens. New sources of resistance in pea and faba bean have been identified and the use of genome-wide association studies is underpinning the development of molecular markers linked to the loci involved. Draft reference genomes of *Pvp* and *Pvf* have been assembled and more than 40 isolates of *Pvp* have been collected and are being sequenced to identify genetic variation between isolates. Strains of *Bacillus* and *Pseudomonas* have been assessed for activity against the pathogen using *in vitro* and *in planta* antagonism assays.

Session 8 - Oomycete Genetics and Genomics

Oral Presentation 8.2.

Gapless genome assemblies reveal the effector repertoire of the sexually evolving spinach downy mildew

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Peronospora effusa causes downy mildew, the economically most important disease of cultivated spinach worldwide. The extensive deployment of genetic disease resistances exerts strong selective pressure on the pathogen, and *P. effusa* rapidly breaks resistances of newly introduced varieties. To date, 19 *P. effusa* races have been denominated based on their capacity to break spinach resistances, but their genetic diversity and the evolutionary processes that contribute to race emergence are unknown. We have recently shown that *P. effusa* races emerge by both asexual and sexual reproduction. We now generated complete and fully phased genome assemblies of six *P. effusa* races by using a combination of long-read and Hi-C sequencing. The comparison of these assemblies revealed a repeat-rich (55% repeats) genome of 58 Mb that is distributed over 17 chromosomes, which display conserved structure and 3D organization. We uncovered a single accessory chromosome that is present in a subset of the races as well as extensive variation in heterozygosity between races but also between the chromosomes of the same race. Each *P. effusa* race encodes more than 380 host translocated effectors, most belonging to the RXLR family. Effectors that share sequence and-or structural similarity are often clustered in the genome. This rich effector repertoire will now enable further research into the molecular mechanisms underlying the interactions between *P. effusa* and spinach.

Session 8 - Oomycete Genetics and Genomics

Oral Presentation 8.3.

Using near-complete genome assemblies to uncover new insights of oomycete biology

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Technological advances in genome sequencing have enabled the generation of near complete genome assemblies of oomycetes. We have generated chromosome-scale genome assemblies for *Bremia lactucae*, *Peronospora effusa*, and *Peronosclerospora sorghi*, which cause downy mildew of lettuce, spinach, and sorghum/maize, respectively. Comparative genomics determined that all downy mildew causing oomycetes share a 17-chromosome ancestral state. Phylogenetics supported that this ancestor is also common with at least *Phytophthora* clades 1 to 5. Annotation of these assemblies revealed that some effector genes are organized as high-identity gene clusters. Comparative genomics between closely related species also identified gene clusters that have diverged more rapidly than the whole genome average. Short-read sequencing of multiple isolates revealed hallmarks of somatic adaptation and demonstrated that isolates containing distinct nuclei may have arisen from single founding genotypes. This has consequences for determining pathotypes and genotyping individuals.

Session 9 - Ecology, Metagenomics and Microbial Interactions

Oral Presentation 9.1.

Weathering a wave of novel marine oomycetes of ecological and economic relevance

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Research in the past 5 years has uncovered an immensely diverse and apparently monophyletic clade of obligately intracellular, marine oomycete pathogens that infect algal hosts such as diatoms, green, red and brown seaweeds. Metabarcoding studies suggest that they are widespread globally and sometimes abundant; thus, they might play an important role in regulating phytoplankton dynamics. Also, the exponential development of seaweed cultivation incurs a global trade of seed stock, which correlates with an apparent worsening of disease outbreaks; however, and contrary to plants, there is hardly any biosecurity framework to monitor and limit the unintentional translocation of pathogens. In this talk, I will present the state of the art concerning the diversity of these intracellular oomycetes and present several projects aiming at accelerating their taxonomic description, their physiological and ecological characterisation, and the development of biosecurity in seaweed aquaculture.

Session 9 - Ecology, Metagenomics and Microbial Interactions

Oral Presentation 9.2.

Physico-chemical properties of natural waters that affect the sporulation of freshwater pathogenic oomycetes *Saprolegnia parasitica* and *Aphanomyces astaci*

Dora Pavić[1], Dorotea Grbin[2], Marija Gregov[1], Josip Ćurko[1], Tomislav Vladušić[1], Lidija Šver[1], Anđela Miljanović[1], Ana Bielen[1].

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Animal pathogenic oomycetes, such as *Saprolegnia parasitica* and *Aphanomyces astaci*, are causing serious disease outbreaks both in natural freshwater environments and in aquaculture. Sporulation, i.e. formation of infectious zoospores, can be triggered in the laboratory by washing the mycelium with natural water. However, the physico-chemical properties of the water that could inhibit or induce sporulation are still unexplored. Therefore, we have washed the mycelia of *S. parasitica* and *A. astaci* with a range of natural water samples collected from different parts of Croatia and observed differences in sporulation efficiency. The results of PLS-R modeling showed that environmentally relevant SAC (spectral absorption coefficient measured at 254 nm), DOC (dissolved organic carbon), ammonium-N and fluoride values had the strongest positive effect on sporulation of *S. parasitica*, while the sporulation of *A. astaci* was not significantly correlated with any of the analyzed physico-chemical parameters of water. In line with this, experimental addition of humic acid, which is an important contributor to SAC and DOC, had a positive effect on the sporulation intensity of *S. parasitica* but not on *A. astaci*. Our results could help to predict the water conditions that promote the pathogen spreading in natural environments and aquaculture facilities, and therefore present a first step in the development of preventive measures. Moreover, this study is a starting point for the design and optimization of laboratory protocols that would result in reproducible oomycete sporulation.

Session 9 - Ecology, Metagenomics and Microbial Interactions

Oral Presentation 9.3.

Phytophthora and *Halophytophthora* spp. are the hosts of multiple viral infections

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Over the last years the diversity of fungal and oomycete viruses (mycoviruses) is greatly expanding thanks to new technologies. Nevertheless, there is still a big gap in the global virome catalogue of these lower eukaryotes. In order to clearly understand the essential patterns and processes of virus evolution a wider range of organisms, including oomycetes, must be sampled and screened. Based on high-throughput sequencing (RNA-seq) numerous novel species of double stranded (ds), positive (+) and negative (-) single-stranded (ss) RNA viruses have been detected and characterized in different species of *Phytophthora* and *Halophytophthora*. The study of the prevalence of these viruses together with their abundance and diversity indicate that they are regular members of their oomycete host populations. Furthermore, oomycetes appear to commonly host complex multiple viral infections. If these viruses are biological entities and influence their host behavior is still undetermined, but our work represents a step forward understanding the origin, spread and evolution of RNA viruses in oomycete species.

Session 9 - Ecology, Metagenomics and Microbial Interactions

Oral Presentation 9.4.

Investigating the molecular basis of pathogenicity by a *Pseudomonas fluorescens* isolate on oomycetes

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Fresh market production of tomatoes, cucumbers, peppers, lettuce, and spinach is shifting to hydroponic greenhouse operations. In hydroponics systems, *Pythium* species can be introduced by airborne dust particles from neighboring farm fields and cause root rots that results in stunting or yellowing of leaves. To identify bacterial antagonists of oomycetes, we surveyed a collection of pseudomonads from a Lake Erie diatom bloom, known to also contain oomycetes. Using a high throughput competitive plate assay, I have identified and sequenced three strains of *Pseudomonas fluorescens* that exhibit contact dependent killing of *Pythium dissotocum P. oopapillum P. ultimum,* and one strain of *Saprolegnia parasitica*. Bioinformatic analysis of the *P. fluorescens* isolate that we have tested most completely, indicates that it has no ability to degrade cellulose, pectin/starch, and contains no recognizable animal effector proteins. To address the utility of this isolate as a biopesticide, we added 50 ml of bacterial culture to a 6 L tub of contaminated water from our experimental hydroponic system. Aliquots of the water sample were filtered on subsequent days, and the filters were placed on V8 media containing antibiotics and antifungal agents. These assays show the bacteria was capable of depleting Pythium propagules in a complex mixture of water containing other bacteria, algae, and fungal species. To identify the molecular basis of this inhibition, we have implemented a targeted gene knockout strategy, generating a first set of candidate gene markers that resulted in loss of virulence.

Session 10 - Host-pathogen Interaction and Resistance Mechanisms III

Oral Presentation 10.1.

Reprogramming of defense-related trafficking in plants during oomycete infection

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During plant invasion, the Irish potato famine pathogen *Phytophthora infestans* penetrates host cells through hyphal extensions known as haustoria. Haustoria are enveloped by a host derived membrane known as the extra-haustorial membrane (EHM) whose functions and biogenesis are poorly understood. Through this interface, the pathogen secretes effector proteins which comprehensively subvert host immune responses. An interesting group of effectors particularly target the EHM, some of which reprogram the defense-related trafficking in host cells. Using the peri-haustorial effectors as molecular probes and cell biology approaches, we identified a series of host immune processes targeting the haustorium interface as well as pathogen counterstrategies. These processes include: (i) modulation of plant autophagy; (ii) trafficking of plant immune receptors; (iii) recruitment of chloroplasts to the pathogen interface. I will discuss our recent discoveries on these processes, focusing particularly on the multidirectional trafficking of NLR type of immune receptors in infected cells when they are in the resting state or activated state. I will also share our recent findings on spatio-temporal dynamics of chloroplast positioning at the haustorium interface, providing insights into their mode of action during infection. A better understanding of the cellular trafficking pathways and the signaling events at various plant pathogen interfaces would enable novel strategies to tackle filamentous plant pathogens.

Session 10 - Host-pathogen Interaction and Resistance Mechanisms III

Oral Presentation 10.2.

Pythium oligandrum: a biocontrol agent with growth promotion and disease resistance properties which does not inhibit mutualistic interactions in legumes

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Pythium oligandrum is a soil-borne oomycete associated with rhizosphere and root tissue and has been used in different plant systems to control soil-borne pathogens. Although numerous studies have shown the effect *P. oligandrum* on growth promotion and protection, its impacts on mutualistic interactions and root microbial community is yet unclear. Our research aims to understand interactions between *P. oligandrum* and mutualistic organisms as well as overall root microbiota. Thus, we devised a model system using *M. truncatula* and PISUM SATIVUM with *P. oligandrum* strain M1. Our results showed that *P. oligandrum* increased *M. truncatula* biomass and yield and a similar observation was drawn for *Pisum sativum*. *P. oligandrum* inoculation induced multiple defense pathways in *M. truncatula*, as proved by RNAseq and metabolomics indicating production of isoflavonoid defense compounds such as medicarpin and formononetin. For mutualistic interactions, *P. oligandrum* promoted formation of *Ensifer meliloti* colonies around *M. truncatula* roots in early growth stage, while *P.oligandrum* soil inoculation resulted in development of multilobed nodules. In addition, *P. oligandrum* did not negatively affect the formation of arbuscular mycorrhizal symbiosis in *M. truncatula*. Metagenomic analysis showed that *P. oligandrum* mycelium in soil changes the structure of microbial community by reducing the relative abundance of fungal taxa related to phytopathogens such as *Fusarium sp.*, or by promoting relative abundance of other mycoparasite fungal genera like *Trichoderma, Clitopilus* and *Alatospora*. Together, our results open new horizons toward understanding benefits of *P. oligandrum* on microbial and mutualistic interactions and to improve development of *P. oligandrum* products.

Session 10 - Host-pathogen Interaction and Resistance Mechanisms III

Oral Presentation 10.3.

Dissecting components of *Solanum americanum* non-host resistance to *Phytophthora infestans*

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The '*R*-gene destroyer' *Phytophthora infestans* is the causative agent of potato late blight and the pathogen responsible for the Irish potato famine in the 1840s. Field infection results in substantial annual crop losses and multiple fungicide sprays are required to prevent this. The non-tuber bearing potato relative *Solanum americanum (S.am)* exhibits strong "nonhost" resistance (NHR) to *P. infestans. S.am* is the source of two late blight resistance genes; *Rpi-amr1* and *Rpi-amr3* that confer strong resistance against multiple *P. infestans* isolates, and are found in most accessions in our collection. To understand NHR and facilitate its transfer into cultivated potato varieties we aim to identify additional *Rpi* genes from *S.am*.

Using AVRamr1 and AVRamr3 recognised by *Rpi-amr1* and *Rpi-amr3*, we have identified several "non- amr1,3" resistance genes in *S.am*. By combining Resistance gene enrichment sequencing (RenSeq) with bulk segregant analysis, we can rapidly map these genes in our *S.am* reference genomes. I will report on progress towards cloning and characterising these novel genes, as well as their potential for elevating disease resistance in potato. One gene, designated *Rpi-amr5*, maps to a cluster of 16 NLRs. Virus-induced gene silencing of this cluster renders the resistant parent susceptible and implicates one paralogue in resistance. We used CRISPR and stable transformation of *S. americanum* and to verify the identity of *Rpi-amr5*.

Session 10 - Host-pathogen Interaction and Resistance Mechanisms III

Oral Presentation 10.4.

Rpi-agf1, a novel broad-spectrum *R* gene against *P. infestans*, reveals the importance of multivesicular bodies during infection

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Late blight is a devastating potato disease caused by the oomycete *Phytophthora infestans*. Wild *Solanum* species have been described to be a source of resistance against this notorious pathogen. In this study we mapped a novel resistance gene, *Rpi-agf1*, present in *Solanum agrimoniifolium* native to Guatemala. This gene provides a broad-spectrum resistance against 26 out of 27 tested *P. infestans* isolates. *Rpi-agf1* is not a classical NB-LRR type of *R* gene; it encodes for lyst-interacting protein 5 (LIP5). LIP5 is a protein conserved in all eukaryotes and is involved in endosomal trafficking via the ESCRT (endosomal sorting complex required for transport) pathway. Particularly, LIP5 is a key regulator of multivesicular bodies (MVBs) biogenesis. MVBs have been described to play a role during both surface and intracellular immune responses. The Rpi-agf1 resistance mechanism is unclear, although we identified helper NRCs as an important downstream signalling component required for the resistance. Additionally, we identified several effectors from distinct *P. infestans* effector families that physically interact with LIP5 via yeast two-hybrid and *in planta*. Apart from describing a novel resistance gene against *P. infestans*, our results emphasize the importance of multivesicular bodies during pathogen infection.

Session 10 - Host-pathogen Interaction and Resistance Mechanisms III

Oral Presentation 10.5.

Phytophthora detected in plants imported to Norway

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Phytophthora species are spreading across borders in Europe, mainly through infected nursery stock, and these alien, invasive organisms are threatening forests, woodlands, and urban greenings. In Norway, several projects have been focusing on plant import. In a surveillance program in 2018 and 2019, a total of 231 soil samples from the rhizosphere of imported, symptomless woody ornamental plants were analyzed for presence of *Phytophthora* using bait leaves for detection. In an ongoing research project, 150 samples from imported plants were analyzed in 2021, and more than 100 samples have thus far been analyzed in 2022.

A total of 19 *Phytophthora* species were detected in 85 out of the 231 samples (37%) during the project in 2018 and 2019. Five of the species detected had not been found in Norway before; *P. amnicola, P. chlamydospora, P. hibernalis, P. parvispora* and *P. occultans*. In the current project, 16 *Phytophthora* species were detected in 65 out of the 150 samples (43%) in 2021, hereof two new species to Norway; *P. multivora* and *P. x stagnum.* By April 2022, four *Phytophthora* species were detected, among them *P. ramorum*.

Of the *Phytophthora* species found, only *P. ramorum* is a quarantine organism in Norway, however, many of the other species are contributing to decline and dieback of several tree species in Norway, e.g. European beech (*Fagus sylvatica*) and grey alder (*Alnus incana*) where *P. xcambivora* is the dominating pathogen on both.

ABSTRACTS

Poster presentations

(According to topics)

Poster topics

1. **Cell Biology, Signalling and Metabolism**
2. **Diversity, Taxonomy and Population studies**
3. **Ecology, Metagenomics and Microbial Interactions**
4. **Effectors, Virulence and Pathogenicity**
5. **Host-pathogen interaction and resistance mechanisms**
6. **Oomycete Genetics and Genomics**

Topic 1: Cell Biology, Signalling and Metabolism

Poster 1.1

The search for markers of oomycete EVs

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The movement of effector proteins and RNAs from pathogen into host cells is known to occur. However, the exact mechanisms facilitating this movement are still being widely studied. One possible explanation is the secretion and uptake of extracellular vesicles (EVs) between organisms.

In this study we are isolating EVs from the oomycete *Phytophthora infestans* with the aim of identifying oomycete-associated EV markers and understanding the cargo of these bodies. This is being achieved by a proteomics approach to identify both secreted and vesicular proteins during *in vitro* growth. We have identified some known EV proteins found in mammalian EV fractions, e.g. enolase, confirming our methodology and approach. We have also identified some oomycete specific proteins that have, as yet, unknown functions. The overall aim of this work is to find markers of EVs that we can use to determine how these EVs are secreted and taken up into the plant cell and whether this is a mode of transport for pathogenicity factors such as RxLR effectors.

Topic 2: Diversity, Taxonomy and Population studies

Poster 2.1

Characterising the UK population of *Bremia lactucae,* the cause of lettuce downy mildew

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Lettuce Downy Mildew (LDM) is a foliar disease caused by the biotrophic oomycete *Bremia lactucae* (Regel). LDM negatively affects crop value through a reduction in yield and visual quality. Disease management strategies are usually preventative, employing resistant cultivars in conjunction with fungicides to preserve crop quality. Little is known about the population of *B. lactucae* but there is an ongoing risk of emergence of new strains which may have reduced sensitivity to fungicide active ingredients or can overcome host resistance genes. Monitoring and characterisation of the *B. lactucae* population would provide evidence in support of integrated LDM management strategies.

A key aim of this study is to examine the genotypic and phenotypic diversity of the UK *B. lactucae* population. Previously developed, and additional, simple sequence repeat (SSR) markers from a local screening of the *B. lactucae* genome, were tested to identify those appropriate for profiling UK isolates collected from commercial lettuce crops. Candidate markers were tested for their ability to identify genotypic (allelic) diversity in UK and other reference isolates. Evidence of discrimination between UK isolates and from reference strains is presented.

Other aims are to a) determine if there are links between genotypic and phenotypic traits, including fungicide sensitivity and virulence profile (race), in relation to geographical location and host cultivar and b) investigate the epidemiological traits of selected genotypically diverse UK isolates under field conditions in mark-and-recapture experiments.

Topic 2: Diversity, Taxonomy and Population studies

Poster 2.2

Decline of *Juglans regia* caused by *Phytophthora* species in Serbia

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When monitoring decline of Persian walnut (*Juglans regia* L.) in Serbia, symptoms typical of *Phytophthora* infections were recorded and included collar rot cankers with staining exudates, bleeding cankers on stems and branches, increased crown transparency and dieback of shoots, and root rot of plants in nurseries. After the isolation, all colonies showed typical *Phytophthora* hyphae under the light microscope. Using classical and molecular identification methods, three species were identified including *P. plurivora*, *P. cactorum* and *P. lacustris*. *Phytophthora plurivora* was the most common species and isolated from both necrotic tissue, and soil samples, while *P. cactorum* and *P. lacustris* were isolated exclusively from soil samples. To test the aggressiveness of the isolated species, two years old *J. regia* and, as comparison, *J. nigra* plants were grown and inoculated under the bark following standardized protocols. In addition, plants were also inoculated with *P. citrophthora, P. gonapodyides, P. polonica, P. pseudocryptogea, P. ×cambivora,* and *P. ×serendipita.* Twelve plants per isolate-host combination were inoculated, and the experiment was finished after 14 weeks. In *J. regia*, *P. citrophthora* and *P. plurivora* were most aggressive, causing mortality of 25% and 17% of plants, respectively. *P. citrophthora* caused the largest necroses, 13 times bigger than in the control. In *J. nigra*, the most aggressive pathogen was *P. ×serendipita*, causing mortality of 58% of plants, followed by *P. plurivora* with 33% mortality. *P. cactorum* and *P. ×serendipita* caused the largest necroses, eight times bigger than in the control. These results showed the high risk posed by *Phytophthora* species to *Juglans* plantings with high importance for both food and wood production. Intriguingly, *J. nigra* was also susceptible in the pathogenicity tests, suggesting that careful monitoring of this species should be performed in both nurseries and planted forests.

Topic 2: Diversity, Taxonomy and Population studies

Poster 2.3

Molecular approaches to detect *Phytophthora infestans* from different varieties of potato seedlings artificially inoculated

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Potato late blight is caused by the oomycete *Phytophthora infestans*, the most destructive pathogen affecting cultivated potato (*Solanum tuberosum* L.). Under favorable conditions, *P. infestans* can infect leaves and tubers and severely spread in field if left uncontrolled. The use of resistant varieties, when available, along with effective detection tools can be considered sustainable strategies to control the disease compared to chemical control. The aim of the present work was to set up a potato seedling assay in order to monitor the colonization by *P. infestans* and test different molecular techniques for the detection of the pathogen. For this purpose, 2-week-old seedlings of different varieties were sprayed with a sporangial suspension of the pathogen and kept under controlled conditions in a phytotron for 10 days. At different stages of the infection leaf samples were collected and analyzed by a targeted PCR approach using *P. infestans* ribosomal ITS specific primers and by real-time qPCR using housekeeping genes primers for the quantification of the plant/pathogen DNA ratio. Both approaches provided a method to detect *P. infestans* in relation to the infection stage and presence/absence of symptoms. Future implementation of this assay will involve the use of Nanopore sequencing so that conventional qPCR will validate both reliability and sensitivity of this next generation sequencing approach as a potential tool for late blight diagnostics.

Topic 2: Diversity, Taxonomy and Population studies

Poster 2.4

Six new *Phytophthora* Clade 9 species from South-East Asian forests

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Species from the genus *Phytophthora* represent a severe threat to the environment and biodiversity worldwide, causing significant economic damage to agriculture, forestry, and nursery activities. *Phytophthora* is an evolving genus with a nearly constant discovery and description of new taxa. Here we report six new species of *Phytophthora* isolated from forest streams and swamps in South-East and East Asia, precisely from Japan, Taiwan and the Indonesian islands of Kalimantan, Sumatra and Sulawesi. These new species were sexually sterile and did not form oogonia in pure culture. Non-caducous, non-papillate sporangia, broad-ovoid, ovoid to pyriform in shape, with short or long pedicels, were produced after incubation for 24-48 h in distilled water with non-sterile soil extract. Chlamydospores were highly variable in dimension, with thin walls. The growth-temperature test confirmed these tropical and subtropical species to be thermophilic, with optimum and maximum growth temperatures being 27.5 °C and 35 °C, respectively. Most isolates ceased growth at 10 °C but resumed growth when brought back to room temperature. Based on preliminary ITS, Btub, HSP90, cox1 and nadh1 phylogenies, these six taxa constitute the *Phytophthora palustris* species complex within *Phytophthora* phylogenetic Clade 9.

Topic 2: Diversity, Taxonomy and Population studies

Poster 2.5

A survey in natural ecosystems of Louisiana revealed a high diversity of previously known and new *Phytophthora* taxa

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In 2020 a survey of *Phytophthora* diversity was performed at 14 locations, natural forests, rivers and marshes across the state of Louisiana. Rhizosphere soil samples and naturally fallen leaves floating in forest streams were collected and transported to the laboratory at Louisiana State University where the isolation tests were performed using a standardized baiting method (Jung et al. 1996). Collected leaves from streams and necrotic leaves from baited soils were plotted on sterilized paper towels, and segments with necrotic spots plated onto selective PARPNH-V8 agar (Jung et al. 1996). Emerging *Phytophthora* colonies were immediately subcultured onto fresh V8 agar and subjected to sequencing of the ITS and cox1 gene regions for species identification. Over 500 isolates containing eight known and seven putatively new *Phytophthora* taxa, together with multiple *Pythium* and *Phytopythium* species were obtained. Most isolates belonged to *P. chlamydospora*, *P.* ×*stagnum* and *P. mississippiae*. In addition, at several sites *P. cinnamomi* and *P. citrophthora* were isolated. Furthermore, seven yet undescribed *Phytophthora* taxa were found, including two taxa each from *Phytophthora* Clades 2 and 7, and three taxa from Clade 10. Morphological studies, temperature-growth tests and multigene phylogenetic analyses are currently underway for the official description of all new *Phytophthora* taxa. Since the pathogenicity and host ranges of the new taxa are still unknown, their occurrence in natural ecosystems could pose a potential risk as does the finding of the aggressive wide-host range pathogen *P. cinnamomi*.

Topic 3: Ecology, Metagenomics and Microbial Interactions

Poster 3.1

Garden waste - a pathway for *Phytophthora* from urban areas to natural environments

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Nineteen different *Phytophthora* species were detected in the rhizosphere of woody ornamental plants imported to Norway in 2018-2019. In total, 231 samples were tested, and *Phytophthora* spp. were detected in 37 % of them. Each sample of 0.5-1 L weighed on average less than one kilogram, thus totally approximately 200 kg soil was tested. This represents only 0.002 ‰ of the around 110 000 tons of imported plant material including attached soil in the period. Based on this, we conclude that urban greenings are commonly infested with *Phytophthora* spp. from imported plants. From such areas these pathogens have several pathways to natural environments. First, *Phytophthora* spores find their way to streams and rivers via drains and surface runoff after rain. Another pathway is transport by, *e.g.* people and vehicles from infested to clean areas, in particular transfer of contaminated soil during construction work. A very common pathway is garden waste with dead and dying plants that are deposited at the edges of urban forests, streams and riverbanks, despite ban on this, *e.g. Thuja*, a major plant genus in the import, which has proven as a common carrier of several *Phytophthora* spp. It is of utmost importance that such plants undergo safe sanitation, *e.g.* aerobic decomposition to achieve high temperatures. At a composting plant in Oslo, we detected *Phytophthora* in incoming garden waste. However, after composting with frequent rotations for up to a year and temperatures reaching approximately 70°C, the final product was free from *Phytophthora*.

Topic 3: Ecology, Metagenomics and Microbial Interactions

Poster 3.2

Diverse previously unknown viruses detected in *Phytophthora* Clade 5 species from Asia

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Recent years have seen a rapid increase in the identification of new viral species among many major taxa of phytopathogenic fungi and in some oomycetes. Novel technologies, such as high-throughput sequencing (HTS), have enabled the detection and description of a number of previously unknown viruses revealing their high abundance and wide taxonomic diversity. Even though viruses residing in fungi typically cause latent infections, there is increasing evidence of virus-induced hypovirulence (reduced virulence) in a number of fungus hosts, drawing considerable attention to their use as potential biocontrol agents. *Phytophthora* phylogenetic Clade 5, an understudied and one of the smallest *Phytophthora* clades, currently includes four species: *P. agathidicida*, *P. castaneae*, *P. cocois*, and *P. heveae*. In this study, high-throughput sequencing (HTS) of total RNA was used to investigate the potential virome of isolates of native populations of *P. castaneae* and *P. heveae* isolated across East and Southeast Asia. The investigation revealed high viral prevalence and diversity of novel viruses with double-stranded RNA (dsRNA), positive-sense single-stranded RNA (+ssRNA), and negative-sense single-stranded RNA (-ssRNA) genomes in the studied isolates. Further study of these viral communities could potentially lead to the development of biocontrol agents for suppression of their phytopathogenic hosts.

Topic 3: Ecology, Metagenomics and Microbial Interactions

Poster 3.3

High diversity of novel viruses in the tree pathogen *Phytophthora castaneae* revealed by high-throughput sequencing of total and small RNA

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Phytophthora castaneae Katsura & K. Uchida (≡ *P. katsurae*, nom. illegit.), an oomycete pathogen native to Southeast and East Asia, causing root and trunk rot of different tree species growing in Asia, was shown to harbor diverse novel viruses from different viral families. Four *P. castaneae* isolates collected from *Chamaecyparis hodginsii* in a semi-natural montane forest site in Vietnam were investigated for viral presence by traditional and next-generation sequencing (NGS) techniques, i.e., double-stranded RNA (dsRNA) extraction and high-throughput sequencing (HTS) of small RNAs (sRNAs) and total RNA. Based on phylogenetic analyses and sequence homology, as well as genome organization the viruses were identified as related to members of the order *Bunyavirales* and families *Endornaviridae*, *Megabirnaviridae*, *Narnaviridae*, *Totiviridae*, and the proposed family "Fusagraviridae." The study describes six novel viruses: Phytophthora castaneae RNA virus 1–5 (PcaRV1-5) and Phytophthora castaneae negative-stranded RNA virus 1 (PcaNSRV1). All six viruses were detected by sRNA sequencing, which demonstrates that the RNA interference (RNAi) mechanism is actively targeting viruses in *P. castaneae*. The highest proportion of virus derived sRNA profiles was 21 nucleotides (nts) in length while 20 and 22 nts peaks were also prominent. To our knowledge, this is the first report of viruses in *P. castaneae* and the whole of *Phytophthora* Clade 5, as well as that of the RNAi system actively targeting viruses in Clade 5 species. PcaRV1 is the first megabirnavirus described outside of the kingdom of fungi.

Topic 3: Ecology, Metagenomics and Microbial Interactions

Poster 3.4

Natural variation in oomycete infection of *Marchantia polymorpha*

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The liverwort *Marchantia polymorpha* and related liverwort species have become emergent models for comparative evolutionary studies in development and plant-microbe interactions. However, most studies of *Marchantia* are on a small number of accessions, and the genomic resources which are available for other more established model organisms are not currently available. With the help of collaborators and a UK-wide citizen science project, we have collected 92 accessions of *Marchantia polymorpha* in axenic tissue culture. We have found natural variation in their resistance or susceptibility to the hemibiotrophic oomycete pathogen *Phytophthora palmivora*. This experimental system opens the door to genome-wide association studies, and the discovery of novel genetic elements which are important in the infection processes of these plants.

Topic 3: Ecology, Metagenomics and Microbial Interactions

Poster 3.5

The microbial BCA *Pythium oligandrum* induces growth promotion in potatoes and causes dynamic changes to the rhizosphere microbiome

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Studies on potato rhizosphere microbiomes have focused mainly on bacterial and fungal spatio-and-temporal dynamics between plant growth stages, genotypes and cropping systems. Very few studies have investigated manipulation of the potato rhizosphere microbiome with the amendment of a BCA, which is of importance for our understanding of the function of BCAs in the environment and their overall impact on soil and plant health. We therefore conducted a series of field trials to investigate whether amendment of the oomycete biocontrol agent *Pythium oligandrum* (P.o) induces growth promotion in potato, and further induces dynamic and temporal changes of the rhizosphere microbiome of the starch potato CV. Kuras.We found a general increase in canopy diameter, and a significant increase in shoot length in potatoes treated with P.o. We further sampled the rhizosphere of the same plants sprayed with P.o or non-sprayed plants as controls, using a non-destructive rhizosphere sampling (NDRS) approach. We sampled at two timepoints (initial growth and senescence). We found that P.o treatment had an effect on both the fungal and bacterial community diversity and we are our exited to present our latest results. We are currently investigating the effectiveness of this BCA to control early blight disease and how either the early blight pathogen or the BCA affect the microbiome and overall plant health.

Topic 4: Effectors, Virulence and Pathogenicity

Poster 4.1

A *Phytophthora* effector targets actin-mediated plastid movement in the host

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Pathogens secrete a variety of effector proteins to act as virulence factors in the apoplast, host cytoplasm and also targeting specific organelles. The mechanisms of effectors include suppressing the host immune response, promoting nutrient acquisition from the host and influencing organelle function. Besides being a central hub in plant metabolism, chloroplasts play a key role in plant immunity by synthesising defence hormone precursors and generating reactive oxygen species and hence tend to be prime targets of effectors. We identified an RXLR effector from *Phytophthora palmivora* that associates with KAC1, a kinesin-like protein. KAC1 is known to regulate actin dynamics during plastid movement in response to light. These data suggest that *Phytophthora* pathogens have evolved effectors that target host cytoskeletal function and chloroplast dynamics to promote infection and disease development. These findings are in line with certain effectors of other plant pathogens found to interfere with the host cytoskeleton and recent evidence showing that chloroplasts alter morphology and accumulate at the pathogen interface. Here, we report our recent results on the effector's influence on cytoskeleton formation and chloroplast movement. The role of chloroplasts in plant immunity and possible reasons for their accumulation at pathogen interface will also be discussed.

Topic 4: Effectors, Virulence and Pathogenicity

Poster 4.2

Aphanomyces euteiches Crinkler effector interferes with RNA silencing

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Crinklers (CRN) effectors are widely distributed among oomycetes and numerous are known to target the host nucleus. Here we address the molecular function of AeCRN5 from the legume root pathogen *Aphanomyces euteiches.* As others CRNs oomycete effectors, AeCRN5 is a modular protein with a conserved Nterminus for plant targeting and a variable Cter domain with an unknown function. We showed that AeCRN5 exhibits dynamic nuclear localisation in plant cells, transiently accumulates in nuclear bodies, triggers cell death and induces major developmental defects when expressed in host root cells. 3D modeling of AeCRN5 Cter sequence using AlphaFold2 predicted a structural homology with *Restriction Endonuclease 5 (REase 5)* family and identified a putative homodimerization of the effector. Co-immunoprecipitation analyses confirmed that AeCRN5 acts as a homodimer within the plant cells. A nucleic acid-protein interaction assay based on FRET-FLIM in *N. benthamiana* leaves revealed the RNA binding ability of AeCRN5 Cter domain. On-going experiments will help to evaluate a putative RNase activity of the effector. Alanine replacement of amino acids predicted to interact with RNA, abolished nuclear bodies accumulation of the AeCRN5 as well as effects on plant development, although the conservation of homodimerization of the effector. While RNA-binding capacity of AeCRN5 mutants are under investigation, we found that AeCRN5 is able to interfere with plant RNA silencing mechanism using a heterologous system. Altogether, these data indicate that AeCRN5 acts through its Cter domain as a plant RNA silencing suppressor probably to facilitate pathogen infection.

Topic 4: Effectors, Virulence and Pathogenicity

Poster 4.3

Comparative sequence, structure and functional insights of a pectinesterase from *Phytophthora infestans*

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Microbial pectin methylesterase (PME) plays a crucial role in pathogen's invasion of plant tissues. The PME enzyme is found in all higher plants as well as microorganisms, including fungi, oomycetes, bacteria, and archaea. Here, we have studied the comparative sequences, structural and functional aspects of a pectin methylesterase from the oomycete pathogen *Phytophthora infestans*. The Pectin methylesterases enzyme acts on the O6-methyl ester groups of the homogalacturonan component of pectin, resulting in its deesterification into pectate and methanol. The *Phytophthora infestans* pectin methylesterase (*Pi*-PME) exhibited maximum activity at basic pH, and broad range of temperature. The *Pi*-PME structure comprised mainly of β-strands and arranged as a β-helix made up of three parallel β-sheets interconnected with loops. The loop regions in the vicinity of the active site are extended as compared to plant and fungal PMEs, but less as compared to bacterial and insect PMEs. Further, molecular dynamics simulations of different *Pi*-PME-substrate complexes (fully, partially, and de-methylated) revealed different behavior in the active site. Interestingly, partially de-methylated pectin is the preferred substrate for catalysis. Those structural characterization, substrate binding and their dynamics behaviour of *Pi*-PME can be further helpful in fields such as plant virulence, pre-processing of pectin degradation and other industrial applications.

Topic 4: Effectors, Virulence and Pathogenicity

Poster 4.4

Defining genomic signatures of *Phytophthora sojae* effectors to better exploit soybean genetic resistance

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The use of soybean varieties carrying *Rps* (Resistant to *Phytophthora sojae)* genes has long been recognized as a very effective way to manage this disease. This method relies on effector-triggered-immunity (ETI), where an R gene product from the plant (*Rps*) recognizes a specific effector from the pathogen, encoded by an avirulence gene (*Avr)*. To better exploit the efficacy of each *Rps* gene, it is crucial to be able to identify the pathotypes of the isolates of *P. sojae* found in the fields, based on the haplotypic diversity of their corresponding *Avr* genes. It was found that for seven of these *Avr* genes recognized by the seven most common *Rps* genes, genomic signatures can be used as accurate predictors of phenotypes. Subsequently, a molecular assay that reveals the different alleles of these seven *Avr* genes was developed in order to diagnose with precision the pathotypes of *P. sojae* isolates. Constant adaptation of *P. sojae* makes the average durability of *Rps* genes to last 8-20 years, leading breeders to deploy new resistance genes, such as *Rps8*. This led us to investigate the interaction of *P. sojae* with this resistance gene and by using the CRISPR/Cas9-mediated genome editing method, we demonstrated that complete knockout of the *Avr3a* gene in *P.sojae* induced a virulence gain towards *Rps8*. This whole project shed light on the complexity of *Avr* genes, and enabled the development of a concrete solution to effectively exploit the use of already deployed *Rps* genes as well as those to come.

Topic 4: Effectors, Virulence and Pathogenicity

Poster 4.5

Identification of *Phytophthora* effectors targeting intracellular organelles in plants

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Phytophthora species are notorious phytopathogens that reduce yield and quality of agricultural production and also affect our ecosystems. In order to obtain specific and sustainable solutions for plant protection, we need a better understanding of the molecular interactions between plants and pathogens. Intracellular organelles such as chloroplasts play a pivotal role in plant immunity and have recently been shown to remodel and relocate upon pathogen encounters. Pathogen effectors, therefore, often directly target plant organelles to interfere with immune responses. Our project aims to identify novel effector proteins targeting plant organelles. Identification of these organelle-targeting effectors will provide more insight in plant-pathogen interactions and the involvement of organelles in the plant's molecular response to biotic stresses.

Topic 4: Effectors, Virulence and Pathogenicity

Poster 4.6

Understanding the evolution of LWY effector repertoire in *Phytophthora*

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RxLR effectors are deployed to manipulate host cellular processes by *Phytophthora*. Previous genome analyses revealed widespread WY/LWY motifs in RxLR effectors. Each WY or LWY unit forms a conserved 3-5 α-helical bundle. Recently, some of these effectors were found to have the WY1-(LWY)n arrangement in which neighboring WY-LWY/LWY-LWY units are concatenated by a conserved, rigid linkage, resulting in an overall non-globular shape (hereafter LWY effectors or LWYs). Despite the structural conservation, the (L)WY units show divergence in surface-exposed residues. We hypothesize that shuffling of (L)WY repeat units may contribute to the functional differentiation in the *Phytophthora* effector repertoire.

Here we developed a new bioinformatic pipeline based on Hidden Markov Model to predict LWY effectors from *Phytophthora* genomes to investigate potential (L)WY domain shuffling events. Generally, we found 112 and 135 LWY effectors from *P. infestans* and *P. sojae*, respectively. A comparison of *P. infestans* LWYs to its sister species *P. mirabilis* revealed a *P. mirabilis* effector PmRxLR1 as a potential recombined product of two *P. infestans* LWYs at the junction of two LWY units. Further analysis of three *P. capsici* strains suggested potential domain shuffling events in seven pairs of LWY effector candidates. These findings indicate that the recombination-based mechanism could promote *Phytophthora* effector evolution.

This study laid the foundation for understanding the modularity-driven diversification of LWY effectors. Future functional analysis of specific (L)WY units or unit combinations will offer novel insight into virulence mechanisms employed by *Phytophthora* and facilitate the deployment of sustainable resistance.

Topic 5: Host-pathogen interaction and resistance mechanisms

Poster 5.1

Biocontrol and growth inhibition of plant pathogen *Phytophthora cactorum*

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The control of plant diseases that are caused by pathogenic fungi or oomycetes depends mainly on synthetic pesticides. However, a long-term and extensive use of chemical pesticides is an overall burden on the environment, and intensive applications increase the risk of pathogens with an evolved resistance to available commercial products. It is of utmost importance to limit the application of agrochemicals and introduce new antimicrobial agents that would be more effective against pathogens and safe for both the environment and humans. Microbes and plants are promising sources of novel natural metabolites with antimicrobial activity for developing new strategies for plant disease control. Here, putative antimicrobial substances were tested against *Phytophthora cactorum*, a representative of the Oomycota phylum. Significant inhibition of *P. cactorum* growth was observed in response to endophytic fungus and extracts from fungal mycelium or orchid tubers.

Topic 5: Host-pathogen interaction and resistance mechanisms

Poster 5.2

Effects of a future climate change scenario on three European temperate tree species infested with soilborne *Phytophthora* species

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Invasive pathogens well-established in forests are usually associated with severe impacts due to the lack of co-evolution between the host and the pathogen but these damages might be exacerbated by new environmental conditions associated with climate change. Each particular host-pathogen interaction might be affected differently. This assay pretends to compare how the interactions between seedlings of different European tree host species and *Phytophthora* may change under current climate conditions and future climate scenarios. Approximately 12 replicates of three temperate tree species per climate scenario were infested with two *Phytophthora* species: *Castanea sativa* inoculated with *P. castanetorum* and *P. cinnamomi*, *Quercus robur* inoculated with *P. plurivora* and *P. quercine* and *Fagus sylvatica* inoculated with *P.* ×*cambivora* and *P. tubulina*. Plants were allocated in two climate chambers, one set for current climate conditions and another chamber set for future climate conditions according to the intermediate RCP 4.5 predictions for the year 2100. Weekly assessments of mortality rates and measurements of gas exchange, chlorophyll fluorescence, leaf spectral reflectance and pigment content were performed and at the end of the trial examination of the root systems was also accomplished. Preliminary results will be presented and discussed.

Topic 5: Host-pathogen interaction and resistance mechanisms

Poster 5.3

Gene regulatory networks in sugar beet defense responses to *Aphanomyces cochlioides*

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Sugar beet (*Beta vulgaris* L.) is one of the main sources of sucrose, providing nearly 30% of sugar production worldwide. The viability of the crop is threatened by the attack of pathogens that cause various diseases, resulting in severe yield losses. The oomycete *Aphanomyces cochlioides* is one of the most problematic pathogens, due to its worldwide distribution and the ability to induce infection at any stage of sugar beet lifecycle, causing seedling damping-off and chronic root rot. During the early stages of sugar beet cultivation the infection can be controlled by the application of chemical fungicides. However, no major control measures are available for the disease management on mature roots. The genetic basis of resistance is still unclear, therefore the identification of genes associated with sugar beet defense responses is an important step for sustainable disease control. In this study, we performed transcriptome analysis of partially resistant and susceptible sugar beet breeding lines infected with *A. cochlioides.* The aim was to elucidate the transcriptional regulatory networks underlying host-pathogen interactions. Differential expression analysis combined with gene ontology enrichment analysis revealed the presence of genes involved in the defense mechanisms during the early stages of infection. The findings of this study shed a light on the gene regulatory processes activated in response to *A. cochlioides* and can be used to assist sugar beet breeding in developing resistant varieties.

Topic 5: Host-pathogen interaction and resistance mechanisms

Poster 5.4

LIFE-FAGESOS- *Phytophthora*-induced decline of Fagaceae ecosystems in Southern Europe exacerbated by climate change: preserving ecosystem services through improved integrated pest management

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This new EU LIFE project aims to address and remediate one of the most severe threats associated with Global Changes, i.e. climate and biological invasions. The outbreak of Alien Invasive Plant Pathogens, adversely impacting natural and semi-natural forest ecosystems. *Phytophthora* diseases are increasing their impact and distribution range in evergreen oak and chestnut ecosystems of the Mediterranean basin, boosted by temperature increases and extreme weather events such as flooding and drought. Scarce public awareness, sensible human impact on forest areas, and the new EU regulation on fertilizers, that limits the use of K-phosphonate in natural and seminatural ecosystems, further increase the risk for Fagaceae ecosystems. Challenged forest ecosystems need improved tools and strategies to enhance their adaptation to the outlined issue, finally ensuring their preservation as important natural carbon sinks.

The project FAGEOS, by joining 13 multi-actor partners, among which the Universities of Tuscia, Sassari (Italy), Cordoba (Spain), Trás-os-Montes e Alto Douro (Portugal), will develop, test, and transfer knowledge and technologies to contain *Phytophthora* epidemics, specifically addressing:

i) The delivery of regional maps for risk- and impact assessment of *Phytophthora* diseases in the Mediterranean basin in diverse current and predicted climatic scenarios;

ii) The development, validation, implementation, and dissemination of Integrated Pest Management (IPM) protocols, tailored to the specific target ecosystem;

iii) The delivery of fully accessible monitoring protocols, based on validated, innovative models and technologies.

Topic 5: Host-pathogen interaction and resistance mechanisms

Poster 5.5

Phytophthora infestans pathogenicity and the efficiency of plant resistance inducers under elevated CO_2

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Climate change brings uncertainties upon agriculture and one concern is that plant protection measures to safeguard food security will be affected, including the late blight of potato caused by *Phytophthora infestans*. The spraying of fungicides and the introgression of resistance genes from wild Solanum species are the most common strategies of disease control, but *P. infestans* has a great capacity of developing resistance to fungicides, as already shown for metalaxyl, and can break down plant resistance in few growing seasons. Integrated disease management is therefore highly encouraged, with plant resistance inducer (PRIs) as a good alternative since they work indirectly by controlling the pathogen through the plant's own immune system, thus having less impact on the environment. However, their performance in a future climate with elevated CO_2 remains unanswered. Since PRIs often impose a mild stress reaction, there is a possibility that such a stress reaction leading to improved resistance can be masked when the plants are grown in an environment that already imposes an elevated stress level. This remains untested in crop species and is the main objective of this study. The efficiency of PRIs under elevated CO_2 as well as the aggressiveness of *P. infestans* in potato plants are being tested under controlled environment and in the field using Free Air CO_2-Enrichement (FACE). We already have preliminary data showing an increased expression of plant defence-related genes in potato cultivars due to increased CO_2 levels and application of PRIs, as well as their impact on resistance against *P. infestans*.

Topic 5: Host-pathogen interaction and resistance mechanisms

Poster 5.6

Phytophthora resistance mechanisms in diverse plant species

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There are several distinct barriers protecting plants against pathogens and pests. Some mechanisms are species-specific, and others are evolutionary conserved. Here, we analyzed the interaction between *Phytophthora* and its host, using several models and complementary 'omics' approaches. Our analyses included *P. infestans* and *Solanum* accessions, *P. cactorum* and *P. plurivora* interaction with hybrid poplar plants, and *P. cinnamomi* and *Castanea sativa*. We employed an artificial inoculation and analyzed the response at plant proteome and metabolome levels. Additionally, we selected samples for lipidome analyses, hormone determination, and enzymatic activity detection to validate the observed changes. Collectively, our data demonstrated differences and similarities in plant defense strategy against *Phytophthora*.

Topic 5: Host-pathogen interaction and resistance mechanisms

Poster 5.7

Reprogramming of defence-related selective autophagy through surface immune activation during *P. infestans* infection

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The Irish famine pathogen *Phytophthora infestans* penetrates host cells and forms haustoria that enable the translocation of effector proteins. Invaded plant cell counters with a spatially confined cell-autonomous defence response known as the focal immunity, a poorly understood process implicated in the concentration of immune responses around pathogen contact sites. We recently discovered that a selective form of autophagy-mediated by the autophagy cargo receptor Joka2—targets the haustorium interface to contribute to the plant focal immunity. Joka2 also contributes to defence against bacteria and viruses, whereas pathogens have evolved effectors to manipulate it. How Joka2 contributes to immunity and how it gets activated by immune signalling are unknown. Here, we discovered that Joka2-mediated autophagy is slowed down upon pathogen recognition, suggesting a non-degradative role of Joka2 in plant immunity. Consistent with this view, autophagic degradation of Joka2 is substantially reduced upon PAMP (Pathogen associated molecular patterns) elicitation. Our proteomics screen and biophysical assays revealed that Joka2 interacts with MAPKs implicated in plant immune signalling. Deletion of the specific Joka2 domain that mediates MAPK interactions impaired Joka2's immune function. Confocal microscopy of fluorescently tagged protein kinases co-expressed with Joka2 uncovered co-localisation to Joka2-labelled foci that condense around the *P. infestans* haustoria. Our further functional, biochemical and cell biology assays suggest an unprecedented mode of action of Joka2 in functioning as a signalling scaffold to regulate immune signalling. These results implicate selective autophagy in plant focal immunity and immune signalling, suggesting more complex functions for autophagy than the widely known degradative roles.

Topic 5: Host-pathogen interaction and resistance mechanisms

Poster 5.8

The cell surface barrier of defense against *Bremia lactucae* in lettuce

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The biotrophic oomycete *Bremia lactucae* is the causal agent of downy mildew disease in lettuce and one of the major threats to its cultivars. Studies of the lettuce-*Bremia* interactions focused on the identification and characterization of intracellular immune receptors with the central nucleotide-binding domain (NLRs) that can recognize effector proteins secreted by Bremia and mount strong defense associated with cell death. Because *Bremia* can quickly overcome resistance in lettuce (a new race evolves every couple of years), there is a need to better understand the *Bremia*-lettuce interactions. Instead of focusing on NLR-based effector-triggered immunity, we investigate activity of the cell surface receptors that recognize pathogen-associated molecular patterns (PAMPs) and can lead to quantitative broad-spectrum resistance. This project is part of the large public-private partnership LettuceKnow and aims to find receptors of PAMPs in lettuce and study how PAMP-triggered immunity leads to the activation of defense and intersects with hormone-elicited defense. We will discuss our experimental approaches such as association mapping and dense time-series expression profiling of transcriptional reprogramming. We will also discuss how to leverage the information on PAMP-triggered immunity for making lettuce a less hospitable host for downy mildew.

Topic 6: Oomycete Genetics and Genomics

Poster 6.1

New Insights into mycoparasitism in oomycete-oomycete interactions revealed through comparative genomics and transcriptomics

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Pythium oligandrum and *Pythium periplocum* are mycoparasitic oomycetes with potential as biocontrol agents. Traits important for successful mycoparasitism are being revealed by comparative genomic and transcriptomic analyses of the hyper-aggressive mycoparasite *P. oligandrum* versus the weaker mycoparasite *P. periplocum* on oomycete and fungal hosts. These diploid, compact, repeat-less genomes lack the typical 2-speed architecture of their phytopathogenic counterparts. Along with the expansion of several families of CAZy enzymes, that we have previously reported, ABC transporter subfamilies are also rapidly evolving in mycoparasitic oomycetes in a strikingly similar manner to mycoparasitic fungi. Dual interaction transcriptomics with the potato late blight pathogen *Phytophthora infestans* as the prey species, reveals that mycoparasitic Pythium species rely on the secretion of an arsenal of trypsin proteases and elicitin-like proteins during mycoparasitic attack, and microscopic studies show that mycoparasitic hyphae directly interact with prey hyphae via the formation of hooks and coiling. Several trypsin proteases are highly differentially expressed during mycoparasitism of *P. infestans* and are tandemly duplicated within the corresponding Pythium genomes. Putative effectors, with similarity to effectors from phytopathogenic oomycetes such as RXLR and CRN genes may also play important roles in oomycete mycoparasitism. During attack, *P. infestans* attempts to defend itself, with the differential expression of genes encoding RXLRs, protease inhibitors and other genes involved in detoxification and cell wall degradation and remodelling.

Topic 6: Oomycete Genetics and Genomics

Poster 6.2

Targeting a tubulin gene in different downy mildews using small RNA inhibits spore germination

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Small RNAs (sRNAs) including microRNA (miRNA) and small interfering RNA (siRNA) are involved in the regulation of gene expression via both transcriptional and posttranscriptional RNA silencing. Exogenously applied sRNAs can enter pest and pathogen cells, either directly or via the hosts, and silence target genes. Downy mildews contain important obligate crop pathogens, which are not amenable to genetic transformation. Recently, we focused on the use of small RNA to decipher functions of genes that are specifically involved in processes that are poorly understood in obligate oomycetes. To gain information on the properties of sRNA-mediated gene silencing, optimise and test in different downy mildew pathogens including *Hyaloperonospora arabidopsidis* (*Hpa*), *Perenospora vicia f. sp. pisi* (*PVP*) and *Bremia lactucae* (*Bl*), we targeted a tubulin encoding gene. We used *Hpa-814031* and identified its orthologues in *PVP* and *Bl*. We then designed 30-nt long siRNAs against the gene. As a negative control, a siRNA targeting the *Hpa* effector gene *Hpa-HAC1* was used. An *in vitro* spore germination assay optimised for *Hpa* and *PVP* was used to test the synthesised siRNAs. Both *Hpa* and *PVP* spore germination was totally inhibited by siRNAs targeting tubulin encoding gene. However, siRNA targeting *Hpa-HAC1* did not inhibit spore germination in either pathogen. Currently, these tubulin targeting siRNAs are being tested against *Bl* and gene silencing is being investigated by RTqPCR. Latest results will be presented.

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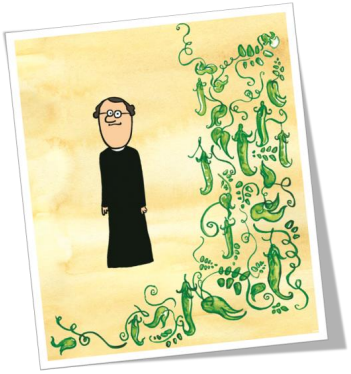


Oomycete Molecular Genetics Network

The oomycete molecular genetics community has a strong culture of collaboration and communication and sharing of techniques and resources. In the 1990's the community started to meet regularly, initially every two years at the Asilomar Fungal Genetics Conference in a concurrent session dedicated to oomycetes. From 1998 onwards annual meetings became a tradition and shortly thereafter the *Phytophthora* Molecular Genetics Network was launched, later renamed Oomycete Molecular Genetics Network (OMGN).

With the blossoming of genetic and genomic tools for oomycetes, many new investigators, from a variety of backgrounds, have become interested in oomycetes. One of the goals of the network is to facilitate the integration of these investigators into the community and to further strengthen the cooperative culture of this community.

The **goals** of the network are

1. Promote communication and collaboration, and minimize duplication of effort, within the worldwide oomycete research community.
2. Promote the entry and participation of new investigators in the field, particularly junior faculty.

The annual meeting serves to meet these goals.

Membership is free and open to all researchers with an interest in oomycetes and from disciplines ranging from molecular genetics, genomics, biology, population biology, ecology, either at an experimental or a computational level. To subscribe to the mailing list 'omgn-users@lists.oregonstate.edu' contact Nik Grunwald (grunwaln@science.oregonstate.edu)

The network is governed by a **Steering Committee** (SC) that has twelve members and two ex officio members.

- The SC has to be diverse in terms of gender, age, location, expertise, ….
- Every year three new members join the SC (six-year term), two of which are elected at the annual meeting, and one is invited by the SC.

The **role** of the SC is:

- Appoint Scientific Chairs and Organizers of the annual meeting.
- Provide feedback to the Scientific Chairs and Organizers on the program, keynote speakers and funding amongst other things.
- Solicit requests for proposals for upcoming meetings.
- Decide on the location of upcoming meetings.

OMGN Steering Committee

As shown in the overview below six SC members were formally at the end of their six-year term in 2020 but due the COVID the term has been extended with two years. Three of six will rotate off this year and the other three next year.

Nominations for new SC members are welcome!

Nominate a colleague or yourself by sending an e-mail to the SC co-chairs Francine Govers (francine.govers@wur.nl) and John McDowell (johnmcd@vt.edu) **before 23 August** or by contacting Francine at the meeting. Persons not attending the meeting can also be nominated. The elections will be held on during the meeting.

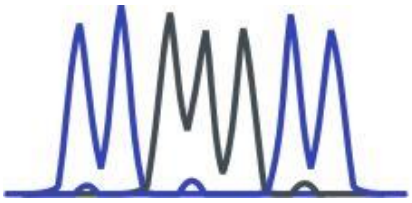
OMGN Steering Committee

Year	2027	2026	2025	2024	2023	2022	2021	2020	2019	2018	2017	2016	2015	before 2015
Meeting Location				Asilomar	Lyon, France	Brno, Czech Republic	NO MEETING		Oban, Scotland	Tai'an, China	Asilomar	Malmo	Asilomar	
	6 year term ends													
	6 year term ends													
	6 year term ends													
		6 year term ends							*Eleonor Gilroy*					
		6 year term ends							*Claire Gachon*					
			6 year term ends						Sebastian Schornack	*Sebastian Schornack*				
			6 year term ends						Miaoying Tian	*Miaoying Tian*				
				6 year term ends					Erica Goss	Erica Goss	*Erica Goss*			
				6 year term ends					Wenbo Ma	Wenbo Ma	*Wenbo Ma*			
						6 year term ends			Francine Govers (chair)	Francine Govers (chair)	Francine Govers (chair)	Francine Govers (chair)	*Francine Govers*	
						6 year term ends			John McDowell (chair)	John McDowell (chair)	John McDowell (chair)	John McDowell (chair)	*John McDowell*	
						6 year term ends			Sophien Kamoun	Sophien Kamoun	Sophien Kamoun	Sophien Kamoun	*Sophien Kamoun*	
						6 year term ends			Elodie Gaulin	Elodie Gaulin	Elodie Gaulin	Elodie Gaulin	*Elodie Gaulin*	
						6 year term ends			Yuanchao Wang	Yuanchao Wang	Yuanchao Wang	Yuanchao Wang	*Yuanchao Wang*	
						6 year term ends			Jean Ristaino	Jean Ristaino	Jean Ristaino	Jean Ristaino	*Jean Ristaino*	
									Brett Tyler	Brett Tyler	Brett Tyler	Brett Tyler (interim chair)	Brett Tyler (interim chair)	Brett Tyler (interim chair)
									Pieter van West	Pieter van West	Pieter van West	Pieter van West	Pieter van West	Pieter van West
										Manual Ospina-Giraldo	Manual Ospina-Giraldo	Manual Ospina-Giraldo	Manual Ospina-Giraldo	Manual Ospina-Giraldo
									Howard Judelson	Howard Judelson	Howard Judelson	Howard Judelson	Howard Judelson	Howard Judelson
											Nik Grunwald	Nik Grunwald	Nik Grunwald	Nik Grunwald
											Laura Grenville-Briggs	Laura Grenville-Briggs	Laura Grenville-Briggs	Laura Grenville-Briggs
ex-officio (Asilomar meeting coordinator, treasurer)											Paul Morris	Paul Morris	Paul Morris	Paul Morris
ex-officio (support logistics, website, travel grants etc)						Joel Shuman			Joel Shuman	Joel Shuman	Joel Shuman	Joel Shuman	Joel Shuman	Joel Shuman
Meeting organizers					Elodie Gaulin Bernard Dumas	Thomas Jung Marília Horta Petra Doleželová et al			Claire Gachon Yacine Badis	Xiuguo Zhang Chunyuan Zhu	Aurélien Tartar Lina Maria Quesada	Laura Grenville-Briggs	Howard Judelson Wenbo Ma	
International Scientific Advisory Committee										John McDowell Francine Govers Sophien Kamoun Brett Tyler Laura Grenville-Briggs Erica Gross				

Overview of network meetings

Year	Date	Location	Scientific chairs	# participants
OMGN meetings				
2023		Lyon, France	Elodie Gaulin; Bernard Dumas	
2022	22-25 August	Brno, Czech Republic	Thomas Jung; Marília Horta, et al.	90
2022	16 March	Asilomar - lunchmeeting	Francine Govers	15-20
2021	**NO MEETING - COVID**			
2020	**NO MEETING - COVID**			
2019	10-13 july	Oban, Scotland	Claire Gachon; Pieter van West; Yacine	111
2018	8-12 April	Tai'an, Shandong Province, China	Xiuguo Zhang; Chunyuan Zhu	200
2017	11-14 March	Asilomar, CA, USA	Aurélien Tartar; Lina Maria Quesada	80
2016	14-17 June	Malmo, Sweden	Laura Grenville-Briggs; et al	93
2015	14-17 March	Asilomar, CA, USA	Wenbo Ma; Howard Judelson	108
2014	2-4 July	Norwich, UK	Mark Banfield; Sophien Kamoun	147
2013	10-12 March	Asilomar, CA, USA	John McDowell; Mark Gijzen	115
2012	26-28 May	Nanjing, P.R. China	Yuanchao Wang; Daolong Dou; et al	154
2011	13-15 March	Asilomar, CA, USA	Paul F. Morris; Manual D. Ospina Geraldo; Vipaporn Phuntamart	67
2010	6-8 June	Toulouse, France	Arnaud Bottin; Elodie Gaulin; Bernard Dumas	151
2009	15-17 March	Asilomar, CA, USA	Kurt Lamour; Mark Gijzen	74
2008	6-8 May	Birnam, Scotland, UK	Paul Birch; Steve Whisson; Pieter van West; Jim Beynon	98
2007	18-20 March	Asilomar, CA, USA	??	??
2006	4-7 May	Wageningen, Netherlands	Francine Govers; et al	84
Phytophthora Molecular Genetics Network				
2005	13-15 March	Asilomar, CA, USA	Jean Ristaino; Jim English	??
2004	21-23 May	New Orleans, LA, USA	Francine Govers; Paul Morris	43
2003	16-18 March	Asilomar, CA, USA	??	47
2002	1-2 Aug	Milwaukee, WI, USA	??	36
Oomycete Genetics				
2001	16-17 July	Wooster, OH, USA	Sophien Kamoun	36
Phytophthora Molecular Genetics Symposium; Beyond Y2K				
1999	13-14 Nov	Wooster, OH, USA	Sophien Kamoun	??
Advances in Phytophthora Molecular Genetics				
1998	15 Aug	Edinburgh, Scotland, UK	David Cooke; Paul Birch	94
Concurrent sessions dedicated to oomycetes at the Asilomar Fungal Genetics Conference				
2007	Advances in oomycete research		Brett Tyler; Francine Govers	
2005	Recent advances in oomycete research		Pieter van West; Jim Beynon	
2003	*no dedicated oomycete session*			
2001	Oomycete Molecular Genetics		Brett Tyler	
1999	Oomycete Genetics ad hoc workshop		Howard Judelson	
1997	Oomycetes		Brett Tyler	
1995	Workshop Genetics and Molecular Biology of Oomycetes		Francine Govers	

External Partners of the OMGN2022 meeting



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Group photo – OMGN2022