

# Virtual Zürich Mycology Symposium 2022

21<sup>st</sup> January 2022 -

<https://us02web.zoom.us/j/81550065913?pwd=bIRaQ1RORlpaem5jeE5PbnBsLzBoZz09>

8h30-8h40	Welcome address
8h40-8h55	<b>Thierry Kuhn (Laboratory of Microbiology, Unine)</b> - Building the fungal highway
8h55-9h10	<b>Alessio Bernasconi (Integrative Biology, ETHZ)</b> - The role of the plant immune system in virulent-avirulent strain co-infection
9h10-9h25	<b>Chiara Pedrazzini (Molecular Ecology, Agroscope)</b> - The population structure of <i>B. brongniartii</i> , the most prevalent pathogen of the European cockchafer
9h25-9h45	Break
9h45-10h	<b>Stephanie Bräunlich (Arable-Crop Breeding, Agroscope)</b> - Wheat rusts in Switzerland: Presence of genetic groups and resistance breeding
10h-10h15	<b>Fanny Germanier (Department of Biology, Unifr)</b> - Combining antagonistic bacteria with copper to control late blight disease in potato plants
10h15-10h30	<b>Luzia Stalder (Evolutionary Genetics, Unine)</b> - The functional ecology of plant microbiome interactions linked to the wheat plant pathogen <i>Zymoseptoria tritici</i>
10h30-10h45	<b>Markus Schlegel (Biodiversity and Conservation Biology, WSL)</b> - Fungal diversity monitoring through the air? - a case study on a waxcap grassland
10h45-11h	Break
11h-11h45	<b>KEYNOTE - Martin Grube (Institute of Biology, University of Graz)</b> - The symbiotic design of lichens
11h45-12h	Concluding remarks

## **Building the fungal highway**

Thierry Kuhn

University of Neuchâtel

Bacterial-fungal interactions play important roles in shaping complex microbial communities and maintaining their functions. In particular, fungal mycelial networks can facilitate bacterial dispersal in water unsaturated environments via so-called “fungal highways”, whereby bacterial cells move through the liquid film around fungal hyphae. The mechanisms underlying fungal highways are, however, hard to study because of the difficulties in teasing apart the effect of network-facilitated bacterial dispersal and the effect of biological interactions. To tackle these difficulties and promote a better understanding of the biological components of bacterial-fungal interactions, we designed and 3D-printed two devices in which stable liquid films can be established to mimic an artificial fungal highway. In the first device, which we call the “bacterial canal”, a water layer of 1.35 mm in width with a relatively strong directional flow are produced, enabling passive transportation of both motile and non-motile bacteria. In the second device, which we call the “bacterial bridge”, a more stationary water film of 0.135 mm in width is generated, allowing only the dispersal of motile bacteria. The devices were 3D printed with a material that is autoclavable, hydrophilic, and of low cost. These devices allow us to disentangle the roles of fungi in biotic interactions (e.g., via the production of exudates or the modification of the surface properties of the mycelium), and in providing a physical structure that allows bacterial dispersal. This will further our understanding of the mechanisms underlying bacterial-fungal interactions along fungal highways. This study also demonstrates the potential of wider applications of microfabrication technologies including 3D printing in accelerating microbial ecology research.

## **The role of the plant immune system in a virulent-avirulent strain coinfection**

Alessio Bernasconi, Bruce McDonald, Andrea Sánchez-Vallet

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Following the gene-for-gene interaction, a strain that carries an avirulent gene is recognized by the corresponding resistant gene on the plant counterpart leading to an incompatible reaction. Consequently, in a single plant-pathogen system, the avirulent strain of, for example, *Zymoseptoria tritici* is unable to infect and reproduce on that specific cultivar, such as Chinese Spring. However, natural infections are frequently produced by several strains that co-infect simultaneously the same host. How the avirulent strain interacts with a resistant cultivar in presence of a virulent strain in mixed infection remains poorly understood. Recently, we demonstrated the substantial advantage offered by the mixed infection that significantly increases the opportunity of asexual reproduction of the avirulent strain on a resistant cultivar. We additionally showed that tissue or cell damage was sufficient for penetration and the reproduction of the avirulent strain. Nevertheless, in distal infection, the avirulent strain seems to profit from the same advantage, and confocal observation revealed early colonization of the plant tissue by the avirulent strain in co-infection with a virulent strain. We suggest that the repression of the immune system response caused by the virulent strain prevents avirulent strain recognition on a resistant cultivar.

## The population structure of *B. brongniartii*, the most prevalent pathogen of the European cockchafer

Chiara Pedrazzini, Jürg Enkerli

Molecular Ecology, Agroscope/ETH

The fungal genus *Beauveria* (Bals.) Vuill. (Ascomycota: Hypocreales) is globally distributed and comprises the insect pathogenic species *B. pseudobassiana* and *B. brongniartii*. While *B. pseudobassiana* has a broad host spectrum, *B. brongniartii* specifically infects *Melolontha* spp. Fabricius, 1775 L. (Coleoptera: Scarabaeidae), a widespread pest throughout Central Europe. *M. melolontha* has a three to four years life cycle, and infested areas are typically occupied by one population, resulting in spatially and temporally shifted populations. *B. brongniartii* is considered the most important antagonist of *M. melolontha* and its presence is linked to *Melolontha* spp. infested sites. A Biological Control Agent (BCA) based on this fungus is commercially available for the control of *M. melolontha* larvae in soil. Although there has been detailed research on the potential and use of *B. brongniartii* as BCA, its population genetic structure has never been investigated, and it is unknown whether it may be affected by the life cycle of *M. melolontha*. The goal of this study was to infer the population genetic diversity and reproduction mode of *B. brongniartii* isolated from *M. melolontha*. A collection of 697 *Beauveria* spp. isolates from infected *M. melolontha* adults from 45 European locations was established. The isolates were analysed using ddRADseq. Phylogenetic analysis identified 413 isolates as *B. pseudobassiana*, suggesting that *B. brongniartii* may not be the most prevalent antagonist of *M. melolontha*. *B. pseudobassiana* isolates either contained the MAT1 or MAT2 mating type indicating a potential for sexual reproduction, whereas among the *B. brongniartii* isolates only MAT1 was detected, which implies clonal reproduction for this species. Further in-depth analyses will provide more information on the distribution and diversity of *Beauveria* genotypes, a prerequisite for developing more specific BCAs of *M. melolontha*.

## **Wheat rusts in Switzerland: Presence of genetic groups and resistance breeding**

Stephanie Bräunlich, Jessica Joaquim, Rachid Majdi, Benjamin Blanchard and Fabio Mascher

Arable-Crop Breeding, Agroscope Changins

Rust diseases are among the most severe threats for global wheat production. Stem and yellow rusts caused by *Puccinia graminis* f. sp. *tritici* (Pgt) and *P. striiformis* f. sp. *tritici* (Pst) respectively, can result in yield losses of up to 40%. Both diseases have received increasing attention in recent years due to the rapid emergence and spreading of new virulent races with concomitant breakdown of many important resistance genes. Even though chemical fungicides are available in Switzerland, disease control relies largely on the deployment of varieties with specific resistance genes. Major resistances are based on the Flor's gene-for-gene model. Therefore, assessment of the presence and the absence of avirulence genes in the Pgt and Pst populations is crucial to estimate the effectiveness of resistance genes. For this, the presence of avirulence genes is assessed by using differential lines, *Triticum* spp. accessions that contain one or a combination of resistance genes. According to their avirulence spectrum, the rust strains are assigned to so-called genetic groups.

In Switzerland, important yellow rust epidemics have occurred in 1999/2001, 2014/2015 and in 2019/2020. We have identified the genetic groups PstS8 and PstS10 (Warrior and Warrior minus race, responsible for the epidemics in 2014/15 and beyond) but also the more recent group PstS14, which is able to overcome most resistant varieties sown in Switzerland. For stem rust, no epidemics have occurred in Switzerland in recent decades, however observations of stem rust became more frequent. In 2021, we recorded for the first time the occurrence of highly virulent strains in Ticino. The strains show similarities to the genetic groups TTRTF and TKKTF, which were previously identified in Sicily and Czech Republic.

The monitoring of stem and yellow rust races together with resistance tests of wheat varieties allows to support breeding efforts to develop varieties that are resistant against novel pathogen races.

## **Combining antagonistic bacteria with copper to control late blight disease in potato plants**

Fanny Germanier, Laure Weisskopf

University of Fribourg

Potato being one of the most consumed crops worldwide, it appears as essential to guarantee its safe and abundant production. Unfortunately, this crop counts many different enemies ranging from insect pests to viruses, including one of the most devastating ones: *Phytophthora infestans*, causal agent of potato late blight. Even if the comprehension of this oomycete's biology is growing day by day, *P. infestans* is still difficult to manage and often requires the use of large amounts of fungicides. In Switzerland, only copper-based products are allowed for organic production of potatoes. However, this heavy metal can accumulate in the soil and presents a toxicological danger for non-target organisms at high doses. Many researchers are currently working on the use of living organisms as natural enemies of different pests, aiming at developing environmentally friendly alternatives. However, biocontrol agents (BCA) tend to show lesser reliability when applied under field conditions in comparison to the laboratory assays, which is likely due to the variability of natural conditions and subsequent stress the BCA have to manage. This project thus aims at finding reliable control strategies by combining biocontrol treatments and reduced doses of copper, to ultimately diminish the total amount of copper and fungicides used whilst maintaining sufficient protection against late blight. Therefore, as a first step, more than 200 bacterial strains were screened *in vitro* against *P. infestans* to select the strains showing the highest antagonistic potential. Then, detached leaf experiments were conducted by applying a mixture of the best bacterial candidates with a reduced concentration of copper as foliar treatments against the oomycete. A selection of the best mixtures was then tested on whole plants grown in the greenhouse. Finally, a field experiment was carried out with the five best bacteria-copper combinations. The assays showed encouraging results especially under greenhouse conditions. Unfortunately, the infection of the field experiment was extremely severe due to the climatic conditions, but, despite the absence of significant results, some promising tendencies could be observed.

## **The functional ecology of plant microbiome interactions linked to the wheat plant pathogen *Zymoseptoria tritici***

Luzia Stalder, Daniel Croll

University of Neuchâtel

Plant-associated microbiomes promote plant health in natural environments and can confer resistance to pathogens. Within these microbial networks, plant-beneficial and plant pathogenic strains are often closely related. Hence, monitoring potential pathogenic and plant beneficial microbes at the strain level is critical for our understanding of microbiome functions. However, comprehensive strain level monitoring has been challenging. Here, we characterize strain level microbiome interactions of *Zymoseptoria tritici*, the main pathogenic fungus of wheat. We use the PacBio Sequel II system to sequence newly developed and highly polymorphic amplicons that allow to characterize pathogen strain diversity in environmental samples. We have developed a joint set of amplicons to monitor the bacterial diversity of the *Pseudomonas* genus, which includes many potential agonistic and antagonistic species. Alongside, we sequence the full-length 16S and fungal ITS loci to generate deep insights into crop microbiomes. We apply our set of amplicons to an extensive hierarchical set of wheat samples spanning the growing season, different plant genotypes, as well as replicated leaf and root compartments. The deep sequencing allows us to track *Z. tritici* strain communities and their microbial environment at very high granularity. We will use evidence for co-occurrence or exclusion of individual genotypes to validate previously unknown synergistic and antagonistic microbiome interactions under controlled conditions. This will enable us to identify biotic and abiotic factors determining the ecological niche of *Z. tritici* and reveal principles of competitive exclusion and persistence of crop pathogens in general. Overall, our work introduces an innovative model system to comprehensively investigate plant microbiome interactions.

## **Fungal diversity monitoring through the air? - a case study on a waxcap grassland**

Markus Schlegel, Jenny Panziera, Veronika Zengerer, Jonas Brännhage, Andrin Gross

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Despite fungi being indispensable for most ecosystems, important basics for their effective protection, conservation and monitoring are still largely missing. In the course of the revision of the red list of macrofungi, efforts are taken to obtain more knowledge on fungal populations in Switzerland. Apart from extensive field surveys and the analysis of citizen science data, the aim is to incorporate information from environmental DNA (eDNA). Especially the analysis of spores collected from the air is a promising tool, which we wanted to evaluate in a pre-study: We collected fungal spores from the air on a waxcap grassland site and analysed them using eDNA metabarcoding. In parallel, all fruit bodies from waxcap and coral-like fungi were assessed in field surveys and their positions determined accurately using GPS. An average spore sample contained more than thousand potential species, and a majority of the species observed during the field surveys were found as well. Only some rare species were not detected. Spore dispersal models suggested that a large majority of spores were deposited in close proximity to the fruit bodies (< 50 m). Considering the red list status of the species, an average spore sample contained approximately up to 26 % of the non-threatened species of low concern (LC), 11 % of vulnerable (VU), 7 % of endangered (EN) and 3.4 % of the critically endangered (CR) species. Taken together, our results suggest that eDNA analysis of air samples to monitor fungi is feasible and has the potential to capture an enormous number of species from a broad taxonomic range. Since spore traps were able to capture the surrounding local diversity, we think that the application of this method for the upcoming revision of the red list of macrofungi of Switzerland will contribute valuable data on species occurrence.