



32nd

Fungal

Genetics Conference

March 12-17, 2024

ABSTRACT BOOK

GENETICS



GSA

G3
Genes | Genomes | Genetics

similar to protective, non-self surveillance systems in plants, animals, and bacteria. Understanding similarities of how different organisms across kingdoms respond to non-self requires leveraging existing model systems and their genetic toolkits. We leveraged two model systems, *Neurospora crassa* and *Pseudomonas syringae* DC3000 (pstDC3000) to dissect fungal response to bacteria. PstDC3000 preferentially surrounds *N. crassa* germlings on a solid surface, causing Propidium Iodide (PI) vital dye uptake, indicative of a cell death response, as early as ten minutes post bacterial proximity. Inoculating *N. crassa* with heat-killed pstDC3000 abolished the PI uptake. Deletion mutants of common or proposed cell death regulating genes in *N. crassa* and pstDC3000 did not abolish PI uptake including; multiple HET genes, VIB1, PhcA, T3SS, and eleven of the seventeen proposed NLR-like genes in *N. crassa*. To try and dissect initial cellular signaling events, we performed transcriptomics on *N. crassa* after pstDC3000 inoculation at ten minutes and one hour. Our study provides insight into an early transcriptional response in filamentous fungi exposed to bacteria alongside surveying fungal NLR-like deletion mutants.

646A A look into the *Pyrenophora teres f. teres* colonization strategies on barley using a transformation-free staining and confocal microscope analysis Ashley C Nelson¹, Gayan Kariyawasam¹, Nathan Wyatt², Janine Haueisen^{3,4}, Eva H. Stukenbrock^{3,4}, Pawel Borowicz⁵, Zhaohui Liu¹, Timothy L. Friesen² ¹Plant Pathology, North Dakota State University, ²Edward T Schafer Agricultural Center, USDA-ARS, ³Evolutionary Biology, Max Planck Institute, ⁴Environmental Genomics, Kiel University, ⁵Animal Sciences, North Dakota State University

Laser scanning confocal microscopy is an invaluable tool in assessing plant microbe interactions at a cellular level. Here we use a transformation-free staining technique with propidium iodide (PI), which stains RNA and DNA, and wheat germ agglutinin labeled with fluorescein isothiocyanate (WGA-FITC), which stains chitin, to visualize fungal colonization of plants. Showcasing this, in tandem with the fungal pathogen *Pyrenophora teres f. teres* (*Ptt*) infecting barley, we show how high resolution images shed light on fungal colonization strategies and infection structures of fungal pathogens. In the *Ptt*-barley interaction, intracellular vesicles develop in epidermal cells directly below penetration points and serve as branching points for hyphal growth into the plant's mesophyll layer. Infected plant mesophyll layers are full of deliberate intercellular hyphal growth that maximizes its surface areas to grow around the individual mesophyll cells, exhibiting patterns we characterize as encasement, mesophyll cell trapping, thick layering, and branching. Encasement is the growth of hyphae in the mesophyll where it surrounds the cells on two opposing sides. Mesophyll cell trapping begins as encasement, but the hyphae continue to grow around the whole mesophyll cells surrounding it on all sides. Thick layering is the layered parallel growth of multiple hyphae and branching is the perpendicular growth of hyphae through numerous layers of mesophyll cells. We analyzed morphological differences between avirulent and virulent isogenic strains of *Ptt* and used the growth patterns mentioned above to assess their success in-planta. Hyphae of virulent strains were most intent on growing parallel to the length of the leaf, through the mesophyll layer as rapidly as possible, followed by lateral branching, explaining the net like lesions characteristic of this disease. Cell death was only observed behind the growing point of the fungus, where mesophyll cells were surrounded by the fungal hyphae. Comparatively the avirulent isogenic isolate was able to grow in-planta but had a fitness deficiency that inhibited its quick takeover of the leaf tissue. We believe the pathogen is maximizing fungal biomass to absorb nutrients at a high efficiency while delaying plant defenses before cell death is an advantage to the pathogen. *Ptt* has shown the potential of this technique to relook at the strategies of fungal pathogens and work in tandem with quantitative and molecular analysis.

647A The key role of the biotic component in kiwifruit vine decline syndrome (KVDS) in Italy, an emerging multifactorial syndrome Micol Guaschino^{1,2}, Marco Garelli^{1,2}, Luca Nari³, Davide Spadaro^{1,2} ¹Dept of Agricultural, Forest and Food Sciences (DISAFA), University of Torino, ²AGROINNOVA, University of Torino, ³Fondazione per la Ricerca, l'Innovazione e lo Sviluppo Tecnologico dell'Agricoltura Piemontese, Agrion

Kiwifruit vine decline syndrome (KVDS) is considered a multifactorial syndrome where abiotic and biotic stressors are involved. In this study, the microbial communities of kiwifruit soil, rhizosphere and root were characterized together with their associations with abiotic stressors. Several soilborne oomycetes belonging to the genus *Phytophthium* were previously associated to the syndrome onset. Association networks unveiled the correlation of *Phytophthium* spp. with the diseased status of orchard soils, whereas in the rhizosphere the oomycete ASVs were negatively associated with growth promoting fungal genera and AM fungi found in kiwifruit. Network analyses conducted for the rhizosphere communities mainly showed that associations were established between different *P. vexans* ASVs, thus revealing strain specific characteristics which require further investigation. The dynamic emerging from the analysis of the ecological processes driving rhizosphere community assembly, highlights the possibility of a dysbiosis phenomenon in the rhizosphere, driven by deterministic processes in the oomycete community. Differently, fungal and bacterial microbiotas showed mainly stochastic processes. The study highlights the importance of considering multifactorial aspects and their interactions in emerging pathosystems, where climate change plays a role in syndrome onset. The combination of different omics techniques is needed for a wider comprehension of oomycete pathogenesis in complex systems.