

Meetings

Twenty-first century mycology: a diverse, collaborative, and highly relevant science

Mycological Society of America Meeting, East Lansing, Michigan, USA, June 2014

In June 2014, over 250 researchers from a variety of national and international locations attended the annual meeting of the Mycological Society of America (MSA), which was held on the campus of Michigan State University in East Lansing, MI, USA. Talks, posters, and symposia covered an impressive breadth of fungal biology organized around four main research themes: cell biology/physiology, ecology/pathology, genetics/molecular biology, and systematics. The taxonomic diversity of the fungal kingdom was highlighted throughout the meeting, with presentations covering all of the major fungal lineages. While the core of the talks was on the fungi themselves, many emphasized interactions with other species, including a wide range of plants, animals, and bacteria. It was also clear that mycologists have adopted all the latest ‘-omics’ and next generation sequencing methodologies to ask cutting-edge questions about topics such as infectious disease, climate change, and bioremediation. At the same time, in the era where almost anyone can generate a fungal sequence, strong organism-based knowledge has become even more important (see in this issue of *New Phytologist*, Herr *et al.*, pp. 27–31). Fortunately, this was on consistent display throughout the meeting and the MSA has long been a welcoming community to researchers from all disciplinary backgrounds. Below we summarize a handful of the many meeting highlights.

‘... while fungal–insect interactions have long been a central theme of mycological research due to their agricultural importance, it is clear that omics-enabled research will continue to push this area in new and interesting directions’

Plant–fungal interactions: what part is bacterial?

It has been known for many years that fungi can harbor bacteria within their hyphae, although the majority of the research on this topic has focused on arbuscular mycorrhizal (AM) fungi in the

phylum Glomeromycota (Bonfante & Anca, 2009). Two talks provided significant new information about these endofungal symbioses. Jesse Uehling (Duke University, Durham, NC, USA) discussed work on the bacterium *Glomeribacter* sp., which grows inside the hyphae of the soil fungus *Mortierella elongata*. This fungus is associated with roots of a range of host plants and has been shown to consistently enhance their growth. Using a combination of different methods to clear fungi of bacteria, Uehling found that *M. elongata* growth on agar plates was strongly affected by the presence of *Glomeribacter*, with bacteria-free fungal strains growing much more quickly. Interestingly, Uehling presented evidence suggesting the suppression of *M. elongata* growth may be a tradeoff that allows for greater tolerance of stressful environmental conditions, particularly changes of pH. Working in a different system, Melissa McCormick (Smithsonian Environmental Research Center, Edgewater, MD, USA) gave a talk featuring work on the diversity of endofungal bacteria associated with *Tulasnella* fungi, which are themselves involved in orchid mycorrhizal symbioses. She found that some bacteria present in *Tulasnella* hyphae were closely related to those previously found in AM fungal hyphae (*Burkholderia* spp.), but not to those in ectomycorrhizal fungal hyphae. In addition, there were also bacterial lineages unique to *Tulasnella* hyphae, indicating a diverse range of bacteria have become independently involved in endofungal symbioses. Curiously, McCormick found that fungi with reduced loads of bacteria in hyphae grew more slowly, which is the opposite of what Uehling found. The reason for this difference is not immediately obvious and represents an enticing direction of future research. In her talk, McCormick also raised the intriguing question about whether these bacteria are actually the important drivers of orchid–fungal interactions, since the amount of fungal colonization (which is positively correlated with bacterial colonization) is positively correlated with orchid growth (see Márquez *et al.* (2007) for a parallel example involving a plant, fungus, and virus). While more work is needed to establish the frequency of these interactions in field settings, to determine to what extent they are facultative vs obligate, and to develop robust methods to consistently visualize bacteria within fungal hyphae, the application of methods such as comparative transcriptomics (i.e. looking at gene expression in fungi with or without endofungal bacteria) holds great promise for better understanding this exciting third dimension of plant–fungal symbioses.

Insects and fungi: probing deep alliances with new methods

Interactions between insects and fungi were also a topic of several talks at the meeting. Joshua Herr (Michigan State University, East Lansing, MI, USA) presented work on the Asian longhorned beetle (ALB), a recently introduced species causing major tree damage in

the north-eastern United States. Herr described both metagenomic and metatranscriptomic work characterizing the bacterial and fungal communities present in the gut of this beetle (Scully *et al.*, 2013). Many ascomycete yeasts were shown to have fluctuating abundances in the gut but only one filamentous fungus was found. The latter was identified as a species closely related to *Fusarium solani* (*Nectria haematococca*). It was always found in the midgut of all the sampled ALB, including those raised on sterile diets. This result may relate to its importance in lignin and cellulose processing when the beetle feeds on tree hosts. Herr also described efforts to sequence the entire genome of this ALB isolate, its shared synteny with *N. haematococca*, and the presence of supernumerary chromosomes that harbor novel genes. Kerry O'Donnell (USDA Agricultural Research Service, Peoria, IL, USA) presented new research of *Fusarium euwallacea*, which is the cause of a recently emerged tree disease with at least five introductions in California and Florida. It is an obligate mutualist found only in association with ambrosia beetles on their mycangium and in active beetle galleries in infested trees. O'Donnell explored the specialization of these fungi with the ambrosia beetles by reconciling phylogenetic trees of the fungus and host and found multiple examples of host switching in the history of the species. For the MSA presidential address (traditionally the last duty of an outgoing MSA president), Joseph Spatafora (Oregon State University, Corvallis, OR, USA) presented a broad synthesis of his research on one of the most well-known insect-associated fungi, the groups *Cordycipitaceae* and *Ophiocordycipitaceae*. He described a range of detailed systematics and genomics work that led to the splitting of 'Cordyceps' into these two separate lineages (Sung *et al.*, 2007). Spatafora also highlighted work on the genome and transcriptome of the beetle larvae pathogen, *Tolyposcladium inflatum*, and how genomic approaches can be used to understand evolution of secondary metabolite producing clusters (Bushley *et al.*, 2013). While fungal–insect interactions have long been a central theme of mycological research due to their agricultural importance, it is clear that omics-enabled research will continue to push this area in new and interesting directions (Hibbett *et al.*, 2013).

Defining fungal lifestyles

Fungi that grow in close association with other organisms, that is symbiotic fungi, are typically classified as mutualists, pathogens, or commensals depending on their effects on their host/partner. Defining the category to which many members of endophytic fungi belong, which are a highly diverse group that grow inside plant tissues, has often been a challenge (Rodriguez *et al.*, 2009). Romina Gazis (Clark University, Worcester, MA, USA) presented a genomics-based approach as part of a larger collaboration with the 1000 Fungal Genomes Project (<http://1000.fungalgenomes.org/>) to help decipher to which category a newly identified fungal endophyte, *Xylona heveae*, may belong. She found that unlike related species in the phylum Ascomycota, including other endophytes, this fungus lacked genes involved in both fungal entry (e.g. formation of appressorium, cutinases) and had a reduced copy number of genes involved in cell wall degradation (e.g. cellulases, hemicellulases, pectinases). This combination suggests that

X. heveae is not likely to be a pathogen, but instead exists either as a commensal or a mutualist. Although this kind of approach is just getting started, it seems very likely that with the continually decreased costs of genome sequencing, genomics will become an increasingly common way to assess fungal lifestyle. An important juxtaposition to this single species-based work was a talk by Georgiana May (University of Minnesota, Minneapolis, MN, USA), which stressed the need for examining fungal lifestyle categories in the context of ecological and evolutionary interactions. She presented work on two symbiotic fungi that frequently co-colonize the leaves of maize (*Zea mays*): *Fusarium verticillioides* and *Ustilago maydis* (Estrada *et al.*, 2012). Both act as pathogens when growing alone on their host, but when grown together *in vitro* (i.e. on agar plates), May showed that *F. verticilloides* grew four times better and significantly inhibited *U. maydis*. This result suggests that rather than being a plant pathogen, *F. verticilloides* may act as a defensive mutualist of plants. To assess this finding *in vivo*, the two fungal species were co-inoculated onto maize and *F. verticilloides* again inhibited *U. maydis* growth. Remarkably, the abundance of *F. verticilloides* when co-inoculated with *U. maydis* was 14 times higher than when grown alone on corn, suggesting the plant may stimulate its growth as an indirect form of defense against attack by *U. maydis*. While the net outcome of this three-way interaction appears to represent a shift from parasitism to mutualism for one pair of partners, this shift is not driven by altruism, but rather by a greater fitness for *F. verticilloides* when co-infecting with *U. maydis*. Given the highly diverse nature of nearly all fungal assemblages, it seems possible that this example may be the 'tip-of-the-iceberg' in terms of shifts among different lifestyle categories depending on local growth neighborhood.

The common language of taxonomists and ecologists: sequence data

Taxonomists and ecologists have historically worked in distinct sub-fields of mycology, with taxonomists focused on systematics and curation of specimen collections and ecologists focused on population dynamics and community structure. As the 'molecular revolution' has swept over the field in the past 20 yr, however, much of the data used by both of these groups is now the same – DNA sequences. Fortunately, there is clear recognition from members of both disciplinary areas that the two can be mutually informative and the symposium entitled 'Sequence-based Identification of Fungi' showed that fungal taxonomists and ecologists are already working together in many constructive ways. Two independent research groups from the University of Tartu, Estonia, have built molecular-sequence databases that have been gaining increasingly widespread use in the mycological and greater ecological community. The UNITE database, which is based on ITS (internal transcribed spacer) sequence data of fungi, contains a number of new features, including a species hypothesis function that allows researchers to annotate the fungal OTUs (operational taxonomic units) used in ecological analyses with a unique identifier that can be tracked across multiple studies (Kõljalg *et al.*, 2013). The MaarjAM database, which includes AM fungal sequence data for multiple gene regions (small subunit (SSU),

large subunit (LSU), ITS and others), allows AM ecologists to create phylogenetically-based virtual taxon units at roughly the species-level, which also greatly facilitates cross-study comparisons (Öpik *et al.*, 2010). Teaming with the originally prokaryote-based Ribosome Database Project (RDP), Andrea Porras-Alfaro (Western Illinois University, Macomb, IL, USA) gave a talk outlining the development of an automated fungal identification classifier for ITS- and LSU-based sequences. A key resource in this latter database is a bootstrapping function that gives confidence intervals for each level of the sequence taxonomic assignments designated. Although each database offers different advantages, all are in a period of transition from Sanger to next-generation sequencing and the ability to query sequences generated by the latter methodology is not yet fully integrated. Despite this lag, this collection of databases gives fungal taxonomists and ecologists an excellent range of tools to use depending on the fungal guild and gene region being studied.

Fungi and agriculture: a double-edged sword

The distinguished Karling lecture, named in honor of the American mycologist John Karling, was given by Bruce McDonald (ETH Zurich, Switzerland). McDonald focused on the intended and unintended consequences of domestication on plants, animals, and fungi. He presented the idea of 'domestication syndromes', which includes the tame behavior of animals and ease of shattering or access to seeds from grasses (maize and rice). He suggested that fungi also have domestication syndromes, including rapid conversion of sugars, strong competitive ability in fermentation settings, and loss of mycotoxins. In addition, McDonald posited that the domestication of fungi was dependent of the development of plant and animal-based agriculture. He highlighted the fact that modern agroecosystems, which are dominated by crop monocultures, serve as active incubators for fungal plant pathogens and that fungal evolution has tracked the host domestication. In particular, he explored the hypothesis that the accessory chromosomes in fungal pathogens such as *Zymoseptoria tritici* may facilitate their 'domestication' to already domesticated plants (Stukenbrock *et al.*, 2010). Overall, the talk provided an excellent integration of large-scale theory and detailed molecular biology about how fungi both affect and are affected by agricultural domestication.

New approaches for studying fungal biology

The meeting also featured important methodological breakthroughs or improvements that will facilitate future research on fungi. In a larger study of the macrofungal communities on the tropical south Pacific island of Moorea, French Polynesia, Todd Osmundson (University of Wisconsin-La Crosse, La Crosse, WI, USA) presented a R-based search algorithm that can be used to determine the most likely geographic origin of fungi found in any given sampling location. Importantly, because the representation of samples from different geographic regions is heavily uneven in the NCBI GenBank database, Osmundson used a random draw technique that queried the database to generate a

null distribution of frequency for each region. He then looked at geographic origin of each of the closest matches to his Moorea samples and found that Pacific rim locations were significantly overrepresented for all matches at $\geq 98\%$ sequence similarity. This result indicates that fungal dispersal, at least in this system, is primarily regional, with geographically distant locations not contributing significantly. Cedar Hesse's (Los Alamos National Laboratory, NM, USA) talk focused on a meta-transcriptomics study of the microbiological communities in maple forest litter in plots with and without nitrate addition. Previous genomics work in this study system indicated that there were shifts in fungal community composition between these two types of plots and his new work showed that nitrate addition appears to both suppress fungal lignocellulolytic enzyme expression and most strongly affect fungi in the phylum Basidiomycota. To obtain these results, Hesse stressed the importance of using an RNA treatment that selectively removed rRNA, as rRNA often dominates total RNA extraction pools. Specifically, he found in untreated vs treated RNA samples from the same plot, the amount of mRNA increases from 5 to 62% and from 1.2 million to 10.5 million reads. This dramatic increase suggests that for environmental transcriptomic studies, which include considerable amounts of rRNA from both prokaryotic and eukaryotic organisms, this additional step will greatly improve the representation of both fungal and prokaryotic mRNA for subsequent ecological inferences that can be drawn.

Looking forward: a bright and collaborative future

Although meeting attendance was slightly lower than usual due to the co-occurring 10th International Mycological Congress in Bangkok, Thailand, this year, the smaller size facilitated productive interactions with colleagues in conference hallways, the mushroom foray, and poster sessions. In upcoming years, MSA will be involved in a number of joint meetings with the Botanical Society of America (2015 in Edmonton, Canada), the International Mycological Association (2018 in San Juan, Puerto Rico), and Mexican Society for Cellular and Molecular Biology (2019 in Ensenada, Mexico). These international meetings represent exciting opportunities to facilitate interactions between mycologists and researchers in other fields of plant science, and continue the increasingly collaborative approach to studying fungi.

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