



# 30<sup>th</sup> Fungal Genetics Conference

**March 12-17, 2019  
Pacific Grove, California**

**Abstract Book**

GENETICS



6120 Executive Boulevard, Suite 550, Rockville, MD 20852, (240) 880-2000  
<http://www.genetics-gsa.org>

**826F Empirical measures of mutational effects define neutral models of regulatory evolution in *Saccharomyces cerevisiae*.** A. Hodgins-Davis<sup>1</sup>, F. Duveau<sup>1</sup>, E. Walker<sup>1</sup>, P. Wittkopp<sup>1,2</sup> 1) Dept of Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, MI; 2) Dept of Molecular, Cellular, and Developmental Biology, University of Michigan, Ann Arbor, MI.

Understanding how phenotypes evolve requires disentangling the effects of mutation generating new variation from the effects of selection filtering it. Evolutionary models frequently assume that mutation introduces phenotypic variation symmetrically around the population mean, yet few studies have tested this assumption by deeply sampling the distributions of mutational effects for particular traits. Here, we examine distributions of mutational effects by measuring the phenotypic consequences of thousands of point mutations introduced randomly throughout the genome. Specifically, we use these mutations to determine the distribution of mutational effects on gene expression for ten genes in the budding yeast *Saccharomyces cerevisiae*. We find that these distributions of mutational effects differ among genes and generally violate the assumption of normality. For example, all of the distributions of mutational effects examined included more mutations with large effects than expected for normally distributed phenotypes. In addition, some genes also showed asymmetries in their distribution of mutational effects, with new mutations more likely to increase than decrease the gene's expression or vice versa. Neutral models of regulatory evolution that take these empirically determined distributions into account suggest that neutral processes may explain more expression variation within natural populations than currently appreciated.

**827W Mating behavior affects the evolutionary success of a killer meiotic driver in fission yeast.** J Lopez Hernandez<sup>1</sup>, R Helston<sup>1</sup>, S Zanders<sup>1,2</sup> 1) Stowers Institute for Medical Research; 2) University of Kansas Medical Center.

According to Mendel's first law, the two alleles at a heterozygous locus have an equal chance to be transmitted to the next generation. Meiotic drive, however, is a phenomenon where an allele biases its own transmission to the next generation. Meiotic drivers are found across eukaryotes, with examples from plants to mammals. Selfishly these genes often impose a fitness cost to the host. For example, in fission yeast, some members of the *wtf* gene family are meiotic drivers. In early gamete (spore) development, a *wtf* driver produces a poison protein that spreads in the spore sac. Subsequently, during spore maturation, the driving gene also produces an antidote protein that rescues only the gametes that carry the meiotic driver. This results in only *wtf*<sup>+</sup> viable gametes produced from *wtf*<sup>+</sup>/*wtf*<sup>-</sup> heterozygote. Population genetics modeling predicts that a driving *wtf* gene will spread in a population due to this transmission advantage. Contrary to this prediction, we observed no change in allele frequency after one generation. Interestingly, we found that inbreeding, facilitated by mating type switching, suppresses transmission of the driver. Surprisingly, the propensity to inbreed greatly varied between natural isolates. The diversity of mating behavior and its influence on drive unveils an intricate path of fission yeast evolution.

**828T Mitochondrial genomes as phylogenetic backbone for evolutionary studies.** B. Brankovics, A. van Diepeningen, C. Waalwijk, Theo van der Lee BU Biointeractions & Plant Health, Wageningen University & Research, Wageningen, Gelderland, NL.

The mitogenome has some unique qualities that make it interesting and worthwhile to investigate for fungal phylogeny. Mitogenomes are present in high copy numbers within cells which makes them easily accessible for molecular techniques such as next generation sequencing and PCR, making it an ideal target for diagnostics and metagenomic analyses. It has a relatively small size and a minimal set of genes, which makes it feasible to study it in its entirety and to identify homologous regions that can be used for phylogenetic inference. The mitogenome could be viewed as an unbiased phylogenetically highly informative housekeeping marker.

Our in-depth investigation of mitochondrial genome diversity and evolution within the genus *Fusarium* has revealed several interesting insights. **1)** The presumed asexual *F. oxysporum* can exchange genetic material with members of other vegetative compatibility groups. **2)** Sequence similarity and phylogeny of parts of the mitogenome indicate gene flow between the *F. fujikuroi* and *F. oxysporum* species complexes, indicating that horizontal transfer has played an important role in the evolutionary history of these two species complexes. This has to be taken into consideration when studying individual gene genealogies within these groups.

The mitogenome offers a phylogenetic backbone, a null hypothesis of the evolutionary past of the organism that could be contrasted to genealogies of effector genes and/or other genes involved in niche adaptation. This would allow the identification of patterns of adaptation such as gene flow or balancing selection. Furthermore, mitogenomes can be used to trace the spread of pathogens and identify their origin.

**829F Phylogenomic resolution and whole genome barcoding of zygomycete lineages with low coverage genome sequencing.** J Stajich<sup>1</sup>, D Carter-House<sup>1</sup>, J Ortanez<sup>1</sup>, J Pena<sup>1</sup>, S Masonjones<sup>1</sup>, Y Wang<sup>1</sup>, A Gryganskiy<sup>2</sup>, G Benny<sup>3</sup>, N Reynolds<sup>3</sup>, M Smith<sup>3</sup>, W Davis<sup>4</sup>, T James<sup>4</sup>, G Bonito<sup>5</sup>, N VandePol<sup>5</sup>, S Mondo<sup>6</sup>, I Grigoriev<sup>6</sup>, A Macias<sup>7</sup>, M Kasson<sup>7</sup>, Y Chang<sup>8</sup>, J Spatafora<sup>8</sup>, ZyGoLife Consortium 1) Microbiology and Plant Pathology, Univ California, Riverside, Riverside, CA; 2) Department of Biology, Duke University, Durham, NC; 3) Department of Plant Pathology, University of Florida, Gainesville, FL; 4) Department of Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, MI; 5) Department of Plant, Soil and Microbial Sciences, Michigan State University, East Lansing, MI; 6) US Department of Energy Joint Genome Institute, Walnut Creek, California, USA; 7) Division of Plant and Soil Sciences, West Virginia University, Morgantown, WV; 8) Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR.

Phylogenetic approaches to classifying species and their relationships has been best served by multigene sequence datasets. Phylogenomics approaches, which apply whole genome data to resolve phylogenies by identifying shared genomic content and the use of molecular phylogenetics has further improved confidence and resolution. The fungal tree of life has benefited from the myriad of efforts which have produced more than 1500 reference quality genome sequences. Low coverage genome sequencing of many species can achieve useful inventory of genes for evolutionary and comparative studies even if chromosome-level assembly is not reached. As part of the ZyGoLife project (<http://zygolife.org>) we have sequenced and assembled low coverage genomes of more than 400 strains of zygomycete fungi at an average of ~5-10x coverage to examine the genetic diversity, cryptic species, and phylogenetic relationships of the Mucoromycota and Zoopagomycota phyla. We have also sequenced and annotated reference genomes from more than 80 species of zygomycetes. The summary statistics for completeness of low coverage assemblies showed BUSCO scores that ranged from 70-98%. Overall we reconstructed the expected relationships for the many of the well studied Mucoralean fungi to allow examination of the multiple origins of mycoparasitism, support of the Endogonales and Umbelopsis lineages as sister to the main Mucorales and paraphyly of the Mucor and Rhizopus genera. We also resolve new phylogeny of the Zoopagomycota and examine genome evolution. Genome sizes in this phylum range from 1Gb in the insect associated Entomophthoromycotina lineages to 18Mb in the Coemansia lineages. To support this work automation tools and workflows for fungal genome assembly (<http://github.com/stajichlab/AAFTF>), annotation (<https://funannotate.readthedocs.io>), and rapid phylogenomic marker extraction and alignment ([https://github.com/stajichlab/PHYling\\_unified](https://github.com/stajichlab/PHYling_unified)) were developed and applied. With costs of 10-fold sequence coverage approaching less than \$30 a strain and the development of rapid, easy to use analysis tools along with robust databases, these data support utility of whole genome barcoding of fungi as feasible approach for fungal species identification and phylogenetics.

**830W Incipient speciation in the rice blast fungus.** M Thierry<sup>1,2</sup>, S Ravel<sup>1</sup>, S Cros-Arteil<sup>1</sup>, H Adreit<sup>1</sup>, J Milazzo<sup>1</sup>, E Fournier<sup>1</sup>, R loos<sup>2</sup>, P Gladieux<sup>1</sup>, D Tharreau<sup>1</sup> 1) UMR BGPI, CIRAD, INRA, Univ Montpellier, Montpellier SupAgro, Montpellier, France ; 2) ANSES, Laboratoire de la Santé des Végétaux-LSV, Unité

de mycologie, Malzéville, France.

Emerging fungal diseases of plants represent a growing issue accompanying global environmental changes, and there is tremendous interest in identifying the factors controlling their appearance and spread. Our model system is the rice blast fungus *Pyricularia oryzae* (synonym *Magnaporthe oryzae*), a textbook example of widely distributed, rapidly adapting pathogen, causing a major rice disease. We previously showed that rice-infecting *P. oryzae* is subdivided into four main lineages, that diversified about 1,000 years ago, including pandemic clonal lineages and a sexually-recombining Southeast Asian lineage displaying signatures of admixture with multiple sources (10.1128/mBio.01806-17). The aim of our study was to clarify the geographical distribution of rice-infecting lineages and the extent of the co-existence in syntopy, and to identify the factors contributing to reduce or facilitate gene flow between them. Analyses of population structure based on Infinium-genotyping of 5300 SNPs for 970 isolates collected on rice on the five continents confirmed the existence of four major lineages (L1 to L4) within *P. oryzae*. The lineages displayed contrasted population structures (L1: recombinant; L2-L4: clonal) and varied in geographic range sizes (L1 and L2: global distribution; L3 widespread in the south hemisphere; L4: mostly South Asian). The recombinant lineage (L1) was the most genetically diverse, with most diversity distributed within and between regions in East and Southeast Asia. Genealogies of clonal lineages L2-L4 had roots in Asia, consistent with an Asian origin. In vitro measurements of reproductive compatibility revealed widespread intrinsic prezygotic and early postzygotic isolation between all lineage pairs with compatible mating types, with the lower levels of reproductive isolation were measured in mating experiments with lineage L1. Mycelial growth rate measurements suggested no difference in temperature optima but pathogenicity testings showed that the lineages were differentially adapted to different rice subspecies. Our work shows that the fungus has been very extensively moved, and that the success of its different lineages is linked to their reproductive isolation, which allows their maintenance in syntopy and enhances their ability to adapt to new conditions in the face of gene flow.

**831T "Illuminating the dark matter of genomics: Structural variation in mini-chromosomes facilitates adaptation of *Magnaporthe oryzae*."** T. Langner, A. Harant, J. Win, S. Kamoun Kamoun Lab, The Sainsbury Laboratory, Norwich, Norfolk, GB.

Eukaryotic genomes are often organized into compartments to facilitate transcriptional regulation, recombination, and adaptation to changing environments. In many plant pathogens, rapidly evolving genes inhabit gene sparse areas in the genome and are tightly associated with repeats and transposable elements – a phenomenon often referred to as the "two-speed genome". Further, there is growing evidence that compartmentalization of genomic regions including virulence factors into mini- or accessory chromosomes can facilitate adaptive evolution.

The blast fungus *Magnaporthe oryzae* is the most devastating rice disease worldwide. In addition, *M. oryzae* can infect more than 50 different grass species including agronomically important staple crops like wheat, barley, and millet. Host adaptation in *M. oryzae* is tightly linked to presence/absence polymorphisms of genes encoding effector proteins that are translocated to the host cell to facilitate infection. Further, there is evidence that recombination can contribute to exchange of genetic material including effectors. This might lead to new structural variants in populations of *M. oryzae* and can putatively facilitate host jumps and epidemics.

Even though karyotype variation in form of mini-chromosomes has been known since decades, their origin, genetic identity, structural features, and influence on population structure and adaptation are largely unknown.

Here we set out to study the extend and effect of large scale structural variation of mini-chromosomes. We present nanopore assemblies of ten rice blast isolates of a clonal population collected in Italy. In addition, we sequenced and analysed the genetic content and structure of 18 isolated mini-chromosomes comprising 2.5% - 10% of the total genome of individual isolates.

**832F Variation and function of the AVR-Pita genes among clonal lineages of *Magnaporthe oryzae* in the United States.** S. Park<sup>1,2,3</sup>, C. H. Khang<sup>4</sup>, J. Correll<sup>5</sup>, Y.-H. Lee<sup>2</sup>, S. Kang<sup>3</sup> 1) Department of Plant Medicine, College of Lice Science and Natural Resources, Suncheon National University, Suncheon 57922, Korea; 2) Department of Agricultural Biotechnology, Center for Fungal Pathogenesis and Center for Fungal Genetic Resources, Seoul National University, Seoul 08826, Korea; 3) Department of Plant Pathology and Environmental Microbiology, The Pennsylvania State University, University Park, PA 16802, USA; 4) Department of Plant Biology, University of Georgia, Athens, GA 30602, USA; 5) Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701, USA.

New pathogen races emerge through various change in avirulence (AVR) genes. AVR-Pita1 and AVR-Pita2, members of the AVR-Pita gene family, but not AVR-Pita3, prevent the rice blast fungus *Magnaporthe oryzae* from infecting rice cultivars carrying resistance (R) gene *Pi-ta*. To understand the mechanism underpinning race variation caused by changes in this gene family, we characterized its members among 23 isolates that represent eight clonal lineages of *M. oryzae* in the United States. Screening of their genomic DNA using three AVR-Pita genes as probes revealed multiple haplotypes, but little variation was observed within individual clonal lineages. All isolates seem to lack AVR-Pita2 but carried a single copy of AVR-Pita3. The copy number of AVR-Pita1 varied from 0 to 3. Novel members of the family may be present too. At least six distinct AVR-Pita1 products are encoded. Three of them confer AVR function, making the strains expressing them avirulent to rice containing *Pi-ta*. Comparison of the AVR-Pita1 protein sequences, in light of virulence of the strains carrying them, and site-directed mutagenesis showed that tyrosine residue at the position 192 is essential for AVR function. Unlike the previously characterized AVR-Pita1 genes, which are located very close to the telomere, most AVR-Pita1 genes in the US isolates might not be telomeric.

**833W Wheat blast and gray leaf spot co-evolved in Brazil through repeated admixture among several host-specialized forms of *Magnaporthe oryzae*.** Mark Farman<sup>1</sup>, Bradford Condon<sup>1</sup>, Mostafa Rahnama<sup>1</sup>, Li Chen<sup>1</sup>, Sebastian Martinez<sup>2</sup> 1) Department of Plant Pathology, University of Kentucky, Lexington, KY; 2) Laboratorio de Patología Vegetal, INIA, Treinta y Tres, Uruguay.

The fungus *Magnaporthe oryzae* (synonymous with *Pyricularia oryzae*) is best known as the "rice blast fungus" due to its historical and worldwide impact on the rice crop. More recently, however, it has garnered attention as a potential threat to global wheat production. Wheat blast is a recently emerged disease and was first identified in 1985 in the Brazilian state of Paraná and has since spread to neighboring states and countries. Another *Magnaporthe*-incited disease of recent origin is gray leaf spot (GLS) which was first found in 1971 on annual ryegrasses growing in the U.S. states of Louisiana and Mississippi. In the mid 1990s, the central U.S. saw widespread outbreaks of GLS in perennial ryegrass and the related grass, tall fescue. Naturally, there is intense interest in determining the origin of these two new diseases. In 2016, the authors of a 10 marker study proposed that certain wheat blast isolates belonged to a new species, *Pyricularia graminis tritici* (*Pygt*) (Castroagudin et al. 2016). However, a subsequent comprehensive phylogenomic study provided compelling evidence that: i) wheat blast is not genetically isolated from other host-specialized forms of *M. oryzae*; and ii) *Pygt* is an artificial construct (Gladioux et al. 2018). Despite the overwhelming whole-genome evidence against *Pygt*, its proponents continue to defend its existence, although they have repeatedly and surreptitiously redefined the species boundary (Castroagudin et al. 2017; Ceresini et al. 2018a; Ceresini et al. 2018b). Using an all-inclusive set of isolates, we now show that Castroagudin et al. made serious errors in isolate sampling, as well as data analysis and interpretation; and that these errors seriously undermine their conclusions. Furthermore, we will present evidence that: i) wheat blast and GLS co-evolved in Brazil as a result of repeated admixture among several host-specialized forms of *M. oryzae*; and ii) admixture very likely continues to occur.

Citations: Castroagudin et al 2016. *Persoonia* 37:199; Castroagudin et al 2017. *BioRxiv* doi.org/10.1101/203455; Ceresini et al 2017 *Annu Rev. Phytopathol.* 56: 427; Ceresini et al 2019 *Molec. Plant Pathol.* in press.