

Poster 24. High-throughput sequencing to assess the microbial diversity in Hessian fly.

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Recently, oceans and soils have been explored for their large unknown microbial genomic resources. Insects could prove another avenue for genomic innovations as majority of insects are known to harbor the symbionts. The objective of this work was to estimate the microbial diversity associated with the Hessian fly, a serious pest of wheat, using 454 pyrosequencing. Insect fat body and midguts from three different larval stages were dissected out. Following DNA extraction, the V3, the most hypervariable region (corresponding to positions 341-534 in *E. coli*) of the 16S rRNA gene was amplified and sequenced. These 454 tag sequences (total of ~6,000) served as query against reference database (V3RefDB) and the phylotype assignments were made according to V3RefDB sequences that display the minimum distance to the query. The most abundant group associated with Hessian fly is γ -*Proteobacteria* followed by β -*Proteobacteria* and *Bacteroidetes*. For assignment of similarity based operational taxonomic units (OTUs), sequences were aligned and distance matrices were calculated by using ARB software, and clustering was done by DOTUR. At the lowest level of dissimilarity, a total of 951 OTUs were recorded. A relatively large number of different populations dominate all samples, which count for observed phylogenetic diversity.

Poster 25. Recurrent deletions of puroindoline genes at the grain hardness locus in four independent lineages of polyploid wheat.

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Polyploidy is known to induce numerous genetic and epigenetic changes but little is known about their physiological bases. In wheat, grain texture is determined by the hardness (*Ha*) locus consisting of genes *Pina* and *Pinb*. These genes are conserved in diploid progenitors but were deleted from the A and B genomes of tetraploid *T. turgidum* (AB). We now report the recurrent deletions of *Pina-Pinb* in other lineages of polyploid wheat and discuss a physiological basis of this phenomenon. We analyzed the *Ha* haplotype structure in 90 diploid and 300 polyploid accessions of *Triticum* and *Aegilops* species. *Pin* genes were conserved in all diploid species and deletion haplotypes were detected in all polyploid *Triticum* and most of the polyploid *Aegilops* species. Two *Pina-Pinb* deletion haplotypes were found in hexaploid *T. aestivum* (ABD). *Pina* and *Pinb* were eliminated from the G genome, but maintained in the A genome of tetraploid *T. timopheevii* (AG). Subsequently, *Pina* and *Pinb* were deleted from the A genome but retained in the A^m genome of hexaploid *T. zhukovskyi* (A^mAG). Comparison of deletion breakpoints demonstrated that the *Pina-Pinb* deletion occurred independently and recurrently in the four polyploid wheat species. The PIN proteins have α -amylase inhibitor activity and bind to the surface of starch granules in the endosperm. We hypothesize that the sudden gene dosage-driven increase in PIN proteins in a neopolyploid would constrain the embryos obtaining nutrition from the endosperm during seed germination. Therefore, deletions of *Pin* genes would be favored for early stand establishment during polyploid speciation.

Poster 26. The finished genomic sequence of the Septoria tritici blotch pathogen Mycosphaerella graminicola.

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Mycosphaerella graminicola is the haploid ascomycete that causes Septoria tritici blotch, one of the most important diseases of wheat worldwide. This pathogen is phylogenetically distinct from other fungi that have been sequenced and is hemibiotrophic; early infection is biotrophic, followed by a switch to necrotrophic growth just prior to symptom expres-

sion. More than 15 genes for resistance have been identified and named in wheat, some of which have been shown to interact in a gene-for-gene relationship. However, the trigger for the switch from biotrophic to necrotrophic growth of the pathogen and the mechanisms of resistance in the host are not known. To better understand the biology of this pathosystem, the genome of the pathogen was sequenced completely by filling in the gaps in an 8.9× draft sequence. The essentially finished sequence contains 18 chromosomes from telomere to telomere, plus five fragments. Four of the five fragments contain telomeres so they presumably make up two additional chromosomes for a total of 20. A comparative bioinformatics analysis of *M. graminicola* with seven other sequenced fungal genomes revealed that *M. graminicola* possessed fewer enzymes than expected for degrading plant cell walls. Analyses of grass-infecting pathogens *versus* those from other hosts indicated that the suites of cell wall-degrading enzymes were tailored to break down the cell wall compositions of their particular hosts. The frequency of transposable elements in the genome of *M. graminicola* was intermediate between those of other sequenced fungi. Many long (> 10 kb) retrotransposons were identified in the finished genome compared to the draft sequence, indicating the need for finishing of other fungal genomes. Availability of the finished genome for *M. graminicola* should greatly aid research on this organism and will help to understand its interaction with wheat.

Poster 27. Transcriptome analysis of high-temperature adult-plant resistance conditioned by *Yr39* during the wheat-*Puccinia striiformis* f. sp. *tritici* interaction.

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Stripe rust (caused by *Puccinia striiformis* Westend. f. sp. *tritici* Eriks. (*Pst*)) is a destructive disease of wheat worldwide. High-temperature, adult-plant resistance (HTAP) to stripe rust is non-race-specific, inherited quantitatively, and is often more durable than race-specific resistance. Previously, we identified and mapped the single *Yr39* HTAP stripe rust resistance gene in the spring wheat cultivar Alpowa, which was identified on chromosome 7BL and accounted for 64.2% of the variation in HTAP resistance. To identify transcripts associated with *Yr39*-mediated HTAP resistance, we selected two recombinant inbred lines from an ‘Alpowa/Avocet Susceptible’ cross that differed at the *Yr39* locus to represent an incompatible (*Yr39*) and compatible (*yr39*) interaction with *Pst*. Using the Affymetrix Wheat GeneChip, we profiled the transcription changes occurring in flag leaves of these two lines over a time-course after treatment with *Pst* urediniospores and mock-inoculation. This time-course study identified 107 and 10 transcripts that were significantly induced and repressed during *Yr39*-mediated HTAP resistance, respectively. Only one transcript was induced during the compatible interaction. The temporal pattern of transcript accumulation showed a peak at 48 h after infection, which was supported by quantitative PCR assays that showed a rapid increase in fungal biomass after this time in the compatible interaction. Most (64%) of the annotated transcripts specifically induced during HTAP resistance were involved in defense and/or signal transduction, including transcripts associated with pathogenesis-related protein production, phenylpropanoid (lignin) and anthocyanin biosynthesis, and receptor-protein-kinase signalling. As expected for non-race-specific resistance, no transcripts associated with an oxidative burst and/or hypersensitive response were identified. This study represents the first transcript profiling of HTAP resistance to stripe rust in wheat, and we conclude that *Yr39*-mediated HTAP resistance involves substantial gene expression changes associated with known nonspecific defense mechanisms.

Poster 28. Interdisciplinary approaches to understanding the mechanisms of cereal crop–aphid pest interactions using a wheat–greenbug system.

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The greenbug, *Schizaphis graminum* (Rondani), is an important aphid pest of small grain crops, especially wheat and sorghum in the Southern Plains of the U.S. No host resistance gene against aphid pests in cereal crops has been cloned, and the mechanisms of host resistance against aphid feeding are not well understood. At the Texas Agricultural Experiment Station – Amarillo, we have a research program aimed at understanding the mechanisms of interaction between the