II. Abstracts of Presentations 9/1/2006 to 8/30/2007

HOLLISTER, E.B., BOUTTON, T.W., S CHAD T, C.W., AND PALUMBO, A.V. Soil microbial diversity in a mesquite savanna. Department of Rangeland Ecology & Management, Texas A&M University, College Station, TX and Oak Ridge National Laboratory, Oak Ridge, TN.

Land use and land cover changes frequently alter key aspects of ecosystem structure and function, both aboveground and below. The invasion of southern Great Plains (USA) grassland systems by mesquite (Prosopis glandulosa), a nitrogen fixing tree, is widespread and has contributed to altered nutrient cycling and hydrologic regimes, reductions in ecosystem stability, and declining abundance of native grassland species. Mesquite invasion typically leads to increased above- and belowground plant biomass, as well as increased stores of C and N in plants and soils. In addition, this vegetation change often alters soil microbial biomass and activity. Accordingly, we hypothesized that these changes in ecosystem structure and function would be reflected in the composition of the soil microbial community. Soil bacterial diversity and community structure were examined in a temperate savanna system that has undergone mesquite invasion. 16S rRNA was extracted, cloned, and sequenced from soil bacterial communities occurring beneath each of four savanna plant types: C3 perennial grasses, C4 midgrasses, C4 shortgrasses, and mesquite. Clone libraries were analyzed using a suite of community analyses tools to determine the degree of diversity present within and similarity between communities occurring under each plant type. For analyses based upon operational taxonomic units (OTU), clones were grouped at the 97% similarity level. Community statistical analysis tools based on OTU comparisons (e.g., J-Libshuff) were not as successful in detecting differences as were those that took into account the actual sequence (e.g., Treeclimber). For example, no significant differences were found with respect to bacterial community composition or diversity with the OTU methods. However, analyses of community phylogeny using Treeclimber indicated that the bacterial community present under C4 shortgrasses was of a significantly different lineage than those characterized from soils occurring under C4 midgrasses, C3 midgrasses, and mesquite trees (p < 0.05). Differing richness and abundance of Actinobacteria were identified as underlying the differences observed between C4 shortgrass and C4 midgrass community phylogenies, while Alphaproteobacteria were identified as underlying the differences observed between the C4 shortgrass and C3 perennial grass phylogenies. Members of both the Actinobacteria and Acidobacteria were related to differences observed between the C4 shortgrass community and that from beneath mesquite. These results suggest that while there is a substantial degree of overlap in the composition of soil bacterial communities associated with individual plant types in this system, subtle and detectable differences do occur and may be important to consider when choosing and interpreting community analysis statistics.

THE ROLE OF THE ARABIDOPSIS TAPNAC GENE DURING PSEUDOMONAS SYRINGAE LEAF INFECTION

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TapNAC is a transcription factor that belongs to the Arabidopsis NAC gene superfamily (NAM, ATAF1 and CUC1-2). The roles of NAC genes in plants are diverse; for example, NAC genes are involved in the regulation of shoot apical meristem development (Aida et al 1997), environmental stress (Fujita et al. 2004) and regulation of cell wall composition (Mitsuda et al, 2005). The present study attempts to elucidate a new role for NAC genes in plant-pathogen interactions. Microarray
analysis revealed that TapNAC, a tapetal specific NAC gene that is normally not expressed in Arabidopsis leaves, is suddenly activated up to 7 fold upon infection by Pseudomonas syringae pv tomato DC3000; no other abiotic or biotic stress resulted in such a statistically significant increase in TapNAC gene expression in non-floral tissues. Elevated expression of a tapetal gene in a leaf tissue after Pseudomonas infection is intriguing, and could be conducive to identifying the function of TapNAC. We used Pseudomonas syringae pv tomato DC3000 to infect seedlings and rosette leaves of transgenic Arabidopsis plants that overexpress TapNAC, that do not express TapNAC (T-DNA knockout line), or that express a chimeric dominant repressor protein (a specific suppressor of TapNAC target genes) in order to investigate the function of TapNAC in plant – pathogen interaction. The infected plants were examined 5 days after inoculation and differences in the disease symptoms were observed. The results of these studies will be presented.

QTL MAPPING OF MODIFIERS FOR THE GENE opaque-2 AND OTHER GENES AFFECTING ENDOSPERM TRAITS IN MAIZE (Zea mays L.)

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Despite its yield and several other remarkable agronomic traits, maize presents low level of proteins in its grain and the proportion of essential amino acids is less compared with other cereals. In an effort to increase the nutritional value of maize, scientists have identified specific loci able to modify the protein composition of the kernel. In 1964, the recessive gene opaque-2 (o2) was identified playing a major role in storage protein quality. Homozygous o2 kernels have higher levels of essential amino acids, especially lysine and tryptophan. Unfortunately, the associated soft and floury features of the o2 kernels cause low yield and susceptibility to diseases. Additional efforts were needed to restore the desirable hard endosperm in a modified opaque version named Quality Protein Maize (QPM). However, neither the mechanism nor the genetic components by which the modification of the endosperm in QPM lines occurs are well understood. We have developed a population of Recombinant Inbred Lines (RILs) derived from the cross between an opaque-2 line and a QPM line to map the position of Quantitative Trait Loci controlling modification of endosperm and other traits. Field trials have been held in College Station (CS) and Weslaco (WE), Texas. For the trait ‘opacity’, we found recurrent major effect QTLs in chromosomes 3, 6 and 8 accounting together for about 50% of the observed variance. Work is still in progress to better define these loci and investigate the inheritance of the hallmark effects of the gene o2. The characterization of the modifier genes will provide important tools for Marker Assisted Breeding to improve the nutritional value of this crop without negatively affecting other traits.
Quantitative Trait Loci Mapping Heat Tolerance during Reproductive Development in Wheat (*Triticum aestivum*).

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High temperature during reproductive development is a major problem to wheat production and end-use quality in the Southern Great Plains and to wheat production in many environments worldwide. We have initiated a project to integrate genotypic (QTL), phenotypic, and transcript level data to identify genes controlling reproductive stage heat tolerance in heat tolerant genotypes of wheat as it relates to yield and end-use quality maintenance. Efforts have initially focused on building recombinant inbred lines (RILs), mapping of QTL linked to reproductive stage heat tolerance, and development of cDNA libraries enriched, through suppressive subtractive hybridization, for genes induced by the heat stress. The selected tissues for library construction included wheat heads and flag leaves isolated from plants subjected to heat stress at 10 days after pollination. RILs were derived from a cross between heat tolerant spring wheat cv Halberd, and a susceptible winter wheat cv Cutter or cv Karl92. These parents were also used as models to define the two adaptive responses to heat stress (heat avoidance (susceptible) and heat tolerance). Over 1920 unique ESTs have been sequenced. These genes include some potential regulatory proteins, heat shock proteins, lipid-transfer protein as well as many novel genes that may belong to uncharacterized pathways involved in response to heat stress. For example, a lipid transfer protein and an alpha amylase inhibitor remained stable during heat shock in heat-tolerant cultivar Halberd. Reproductive stage heat tolerance QTL mapping results will also be presented.

Impact of High Nighttime Temperature on Rice (*Oryza sativa* L.) Growth, Development and Physiology

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The presence of seasonally high nighttime temperatures (HNT) along the United States Gulf Coast and in regions of similar climate, occurring during the critical stages of development, reduces rice (*Oryza sativa* L.) yield and quality. The objective of this study was to determine the effects of HNT and chemical preventive treatments on growth, development, physiology and yield of rice plants. Plants were grown under ambient nighttime temperature (ANT) (27 °C) or HNT (32 °C) in the greenhouse. They were subjected nightly to a HNT through use of continuously controlled infrared heaters, starting from 2000 h until 0600 h. The HNT did not affect plant growth and development except for leaf number and total leaf area. Elevated nighttime temperatures did not hasten crop development rate, as indicated by the dates of panicle emergence. Plants grown under HNT showed a 54% decrease in panicle dry weight (PDW) compared to ANT. Reduction in PDW appeared to be due to insufficient photosynthate supply to the grain as a result of a decrease in net leaf photosynthesis (Pn) and an increase in respiration rates. Rice plants pre-treated with the salicylic acid or glycine betaine generally showed an increase in PDW (up to 107% increase) and Pn, and a decrease in respiration rate compared to untreated plants, when grown under HNT.
Plants Defective in Calcium Oxalate Crystal Formation Have More Bio-available Calcium

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Bio-available calcium affects bone formation and calcification. Here we investigate how a single gene mutation altering calcium partitioning in the forage crop Medicago truncatula affects calcium bioavailability. Previously, the cod5 M. truncatula mutant was identified which contains identical calcium concentrations to wild-type, but contains no oxalate crystals. In this study, equal number of male and female mice (n=120) were randomly grouped and then fed one of four 45Ca-containing diets: M. truncatula extrinsically or intrinsically labeled and cod5 extrinsically or intrinsically labeled. Absorption of the tracer was determined in the legs one day after consumption. The absorption was similar in the M. truncatula and cod5 extrinsically diets; however, in the intrinsically labeled diets, calcium adsorption was 22.87% (p<0.001) higher in mice fed cod5. Our study presents the first genetic evidence demonstrating the functional impact of removing oxalate crystals from foods.

Gene Expression in an Association of Loblolly Pine

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As part of the Plant Genome project “Association Genetics of Natural Genetic Variation and Complex Traits in Pine” we are analyzing expression of approximately 200 genes involved in xylem development and disease responses in loblolly pine. An association population of 500 genotypes from across the natural range was developed at North Carolina State University. We, at Texas A&M University, are using real-time qPCR to assay expression levels in 426 of these genotypes. We hope to relate expression data to geographic origin, to identify genes that are coordinately expressed, and to associate variation in expression to genotypic and metabolic data being produced at UC-Davis and to phenotypic data being produced at U. Florida and North Carolina State. Preliminary expression data for the first 23 genes involved in lignin biosynthesis has been gathered and will be presented here. Other xylem-related genes to be analyzed include those involved in cellulose and cell wall biosynthesis, cell wall proteins, and genes involved in signal transduction.