2012 Southern Division Meeting Abstracts

Abstracts presented at the APS Southern Division meeting in Birmingham, Alabama, February 5–6, 2012. The abstracts are arranged alphabetically, by first author's name.

Molecular diversity in field population of *Sclerotinia sclerotiorum* isolates affected common bean in Upper Egypt

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Phytopathology 102:S2.1

The tested isolates of *S. Sclerotiorum* from different localities of Upper Egypt were able to infect common bean Balady cv. causing Sclerotinia rot with different degrees of severity ranging from moderate to severe. They were varied in culture morphology and production of sclerotia and apothecia on PDA medium as well as in soil. Isolates consisted of eight vegetative compatible groups (VCGs); three of them consisted of two different isolates (3EB, 5EB), (3EB, 6ED) and (5EB, 6ED) and the remaining 5 VCGs each consists one isolate, compatible only with itself. Cluster analysis of electrophoresis patterns of proteins obtained by PAGE and SDS-PAGE and isozyme obtained by PAGE were able to differentiate the tested isolates of the pathogen, according to similarity levels, to different groups. Grouping the isolates by PAGE was not related to their virulence or geographic origin, while, grouping them by SDS-PAGE was partially successful in geographical origin. Cluster analysis of random amplified polymorphic DNA (RAPD) banding patterns using two primers was unable to detect appreciable differences in DNA banding patterns of the tested isolates.

Comparison of stream baiting protocols for recovering *Phytophthora ramorum* from streams and nursery discharge

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Phytophthora ramorum, the causal agent for Sudden Oak Death, has devastated the coastal California black oak, tanoak, and coast live oak populations in fourteen coastal counties in California and one county in Oregon. Through the shipping of infected nursery stock from the West Coast, *P. ramorum* has been introduced into nurseries throughout the southeastern United States. The pathogen persists in infected nurseries and is leaving these nurseries in discharge water. As many of the eastern United States woody plants can act as sources of inoculum, there is a plausible pathway for the pathogen to enter the local ecosystems and remain a long-term threat. Streams outside of nurseries in Alabama, Mississippi, Florida, Georgia, and North Carolina have tested positive for *P. ramorum*. The current method for recovering *P. ramorum* is in situ baiting for one to two weeks using mesh

bags and non-wounded rhododendron leaves. In situ baiting allows for the sampling of large volumes of water over an extended time period and has proven effective for capturing of *P. ramorum* (88% detection rate). However, baits are vulnerable to losses from storms and vandalism and require constant immersion in the stream for the entire baiting period. An alternative method of in vitro baiting has been developed using one liter bottles to collect stream water, immediately into which non-wounded and wounded rhododendron baits are placed and then incubated. The in vitro assay, Bottles of Bait (BoB), is nearly equivalent to in situ baiting for pathogen capture (83% detection rate), less expensive to perform, eliminates bait losses, and does not require sustained water flow.

Relative efficacy of nine fungicides applied to peanut early emergence for foliar and soilborne diseases

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Prothioconazole (0.20 kg/ha) was recently found to reduce foliar and soilborne diseases of peanut (*Arachis hypogaea*) at harvest when applied in an 8–10 cm band 21 days after planting (DAP). Four trials were conducted in 2011 to compare prothioconazole with standard broadcast use rates of azoxystrobin, penthiopyrad, tebuconazole, flutolanil/propiconazole, pyraclostrobin, prothioconazole/tebuconazole, fluoxastrobin and picoxystrobin, with all treatments banded at 21 DAP. All trials were oversprayed with chlorothalonil at sprays 3–7 of a normal 7-spray program for leaf spot (*Cercospora arachidicola*) control. In three trials where leaf spot occurred, all fungicides except tebuconazole had less disease than the nontreated control. In three trials with low stem rot (*Sclerotium rolfsii*) incidence there were no treatment effects on disease levels. The fourth trial had 27% incidence in the control, and only prothioconazole and azoxystrobin had less stem rot. Across all trials, only prothioconazole significantly increased pod yield over the nontreated control (5763 vs 5230 kg/ha). Fungicides applied to peanut at early emergence can reduce epidemics of foliar and soilborne diseases at harvest, and prothioconazole is particularly well suited for this application.

Assessing the impacts of cogongrass (*Imperata cylindrica* (L.) Beauv) on root-feeding bark beetle populations associated with Southern Pine Decline

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The non-native, invasive grass *Imperata cylindrica* (L.) Beauv. (cogongrass) is becoming an increasing threat to the diversity of native plant and wildlife species of the southeastern United States. Cogongrass infestations have also been found to affect pine ecosystems by altering fire regimes and limiting regeneration by forming extensive monocultures through rhizomatous growth. Another issue that landowners of southeastern U.S. pine forests are facing is Southern Pine Decline (SPD). The factors associated with SPD include a complex of abiotic and biotic stressors that can cause economically significant premature mortality in mature pine forests. A suite of root-feeding bark beetles, which vector staining fungi associated with SPD, are attracted to

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stressed pines. Cogongrass could be one biotic factor attributing to pine decline by possibly inducing additional stresses to pines, leading to higher infestations of root-feeding bark beetles. Twenty plots were established in a loblolly pine plantation located in southeastern Mississippi, ten of which were placed in areas with an extensive infestation of cogongrass and ten were placed in areas without cogongrass. Insect traps placed within each plot are checked on a bi-weekly basis to observe insect population trends. Current trends show that *Hylastes salebrosus* is the most important species observed and has shown consistently higher populations in plots with cogongrass versus plots without cogongrass. *Hylastes porculus*, *Dendroctonus terebrans*, and *Hylobius pales* have also shown similar trends as *H. salebrosus* but, populations are currently not significantly different between plots with cogongrass and plots without cogongrass for any species. Soil moisture was found to be significantly higher in the top eight inches of the soil horizon for plots with cogongrass versus plots without cogongrass. This may be an indication cogongrass rhizomes are altering moisture availability to loblolly pine roots and causing additional stresses to the trees.

Curative activity of penthiopyrad and pyraclostrobin on infections by *Cercospora arachidicola* of peanut (*Arachis hypogaea*)

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Phytopathology 102:S2.2

Detached leaf experiments were used to evaluate the curative potential of penthiopyrad and pyraclostrobin on infections caused by *Cercospora arachidicola*. In the first experiment, peanut leaves were inoculated with 1.0×10^4 spores/ml of *C. arachidicola*, maintained in an environment conducive for infection, and sprayed with penthiopyrad (10.0 fl oz/acre), penthiopyrad (16.0 fl oz/acre), or pyraclostrobin (9.0 fl oz/acre) at -1, 1, 2, 3, or 5 days after inoculation (DAI). Unsprayed, inoculated leaves were used as a positive control, and unsprayed, non-inoculated leaves were used as a negative control. Incubation period ranged from 13 to 30 DAI. All fungicide treatments significantly suppressed disease for all application treatments, with nearly complete control for the -1, 1, 2, and 3 DAI treatments. For the 5 DAI treatment, infection frequency was reduced by at least 80%. Additional experiments were done to extend the length of the application treatments to include 7, 8, and 9 DAI, and to include chlorothalonil (24.0 fl oz/acre) as a non-curative fungicide control. In these experiments, chlorothalonil significantly reduced infection frequency for all application treatments up to 5 DAI ($P < 0.01$). Penthiopyrad and pyraclostrobin showed significant activity up to 9 DAI ($P < 0.01$). All leaves treated with penthiopyrad or pyraclostrobin had at least a 75% reduction to infection frequency compared to the positive controls. No differences in activity were evident between penthiopyrad and pyraclostrobin for any of the application dates tested ($P \geq 0.13$). These results suggest that spores of *C. arachidicola* may be susceptible to protectant fungicides as late as 5 DAI, and that penthiopyrad and pyraclostrobin are able to kill the fungus after penetration. More research is needed to better understand the stages of fungal development over time in this assay, and to evaluate the activity of these fungicides beyond 9 DAI.

A new mechanism of DMI fungicide resistance in *Monilinia fructicola*

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Monilinia fructicola (G. Wint.) Honey is a widespread causal agent of brown rot, causing serious disease of peach fruit in China and elsewhere. Control of this disease is largely dependent on demethylation inhibitor fungicides (DMIs) but resistance is starting to emerge. Resistance in *M. fructicola* is associated with the mobile genetic element 'Mona' upstream the *cyp51* target gene. In this study we generated and analyzed mutants resistant to the new DMI fungicide SYP-Z048, chemical name (5-(4-chlorophenyl)-2,3-dimethyl-3-(pyridine-3-oxazoline). Resistant mutants were generated by UV (ultraviolet) exposure in the laboratory. The four common target genes of ergosterol biosynthesis inhibitor (EBI) in agriculture were isolated in the sensitive and resistant isolates, including *cyp51*, *erg2*, *erg24* and *erg27*, encoding C-14 demethylase, $\Delta 8 \rightarrow \Delta 7$ sterol isomerase, C-14 sterol reductase, and 3-keto reductase, respectively. The results showed that resistance was correlated with the amino acid (AA) change from tyrosine to phenylalanine at position 136 (Y136F) in *cyp51*. No AA changes were found in *erg2*, *erg24* or *erg27*. The *cyp51* gene containing the mutation was transferred via *Agrobacterium tumefaciens* and expressed using the CaMV35S promoter in DMI sensitive isolates of *Botrytis cinerea* and *Fusarium verticillioides*. The sensitivity of transformants of *B. cinerea* and *F. verticillioides* to SYP-Z048 was reduced, with highest resistance factors of 7.5 and 8.0, respectively, compared with the

parent isolate confirming that the Y136F mutation is indeed associated with DMI resistance. The *M. fructicola* mutants were less fit compared to wild-type isolates suggesting that reduced fitness may have prevented the emergence of isolates with the Y136F mutation in the field.

Regulation of the virulence factors by the TofI/TofR quorum-sensing system in *Burkholderia glumae*: The major pathogen causing bacterial panicle blight of rice

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Phytopathology 102:S2.2

Burkholderia glumae is the major pathogen causing bacterial panicle blight (BPB), an economically important rice disease in the southeastern United States. TofI/TofR quorum sensing system is a key regulatory element for the production of virulence factors of *B. glumae*. To better understand the relationship between quorum sensing and known virulence factors (toxoflavin, flagella and lipase), as well as, putative virulence factors (i.e. extracellular polysaccharide), a series of mutagenic and phenotypic analyses were conducted in this study. Deletion mutants for quorum-sensing genes, *tofI* and *tofR*, were successfully generated by using pBBSacB with a sucrose-sensitive counter selective marker, SacB. The parental strain 336gr-1 and its quorum-sensing-defective derivatives have undergone a series of phenotypic characterizations and quantification tests. We observed toxoflavin production and flagellum-mediated motility in *tofI* and *tofR* mutants on solid media. This observation strongly indicates the presence of additional regulatory component(s) for the virulence factors.

Peanut cultivar improvements in field resistance to *Tomato spotted wilt virus* in the southeastern U.S.

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Phytopathology 102:S2.2

When spotted wilt, caused by thrips-vectored *Tomato spotted wilt virus* (TSWV), emerged as a threat to peanut (*Arachis hypogaea* L.) production in the southeastern U.S. in the early 1990's, identifying genotypes with resistance to the TSWV and development of resistant cultivars rapidly became an essential objective of all peanut breeding programs in the region. The cultivar 'Southern Runner' was developed and released before spotted wilt became a problem, but was found to have a moderate level of field resistance compared to 'Florunner', the predominant cultivar grown at the time. By 1995, several genotypes had been identified with similar or slightly better resistance than that of Southern Runner. In 1996 the cultivar 'Georgia Green' was released. It has moderate resistance similar to that of Southern Runner, and responds well to other cultural and chemical factors in an integrated spotted wilt management program. In 1999, 'C-99R' was released with slightly better resistance than Georgia Green. Since then, several cultivars have been released with even better resistance than that of Georgia Green or C-99R. Resistance in cultivars such as 'Georgia-06G', 'Florida-07' and 'Tifguard' is sufficient to allow much more flexibility with other practices that suppress spotted wilt than was possible with Georgia Green. Cultivars 'Georgian' and 'Georgia-10T' have the best resistance to TSWV of currently released cultivars. However, genotypes have been identified that under extremely heavy pressure exhibit even better resistance than these. Therefore, there is potential for further improvements in resistance to TSWV in the future.

Infection of sweetpotato by fungal end rot pathogens prior to harvest

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The end rot disease complex, caused mainly by the fungi *Fusarium solani* (Fso) and *Macrophomina phaseolina* (Mp), has become an important postharvest disease in sweetpotato storage roots. The disease develops a few months after the storage roots are harvested and stored. However, isolations attempted after harvest have shown that the pathogens can be present asymptotically inside the storage roots long before the disease manifests. To determine how and when these fungi enter the storage roots, a greenhouse experiment was done using tissue culture-derived plants free of Fso and Mp. Plants were grown in non-infested soil or in soil infested with each fungus alone or combined. Isolations were attempted from different parts of the plants at intervals after planting. Fso and Mp were recovered from roots and plant stems below the soil line, at the soil line, and five centimeters above the soil line at each sampling time. This suggests that these fungi invade the sweetpotato plants after planting and systemically colonize the plant. Furthermore,

many other species of *Fusarium* were also isolated from plants cultivated in non-infested soil, in soil infested with Mp, and in soil infested with both fungi. However, such species were rarely isolated from plants cultivated in soil infested with Fso alone. These findings indicate that infection with Fso and Mp can occur prior to harvest. More research is necessary to elucidate the ways these fungi enter the plants.

Effects of environment and genotype on charcoal rot development on soybeans

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Charcoal rot of soybean, caused by the soilborne fungus *Macrophomina phaseolina*, is a disease associated with high soil temperatures and low soil moistures. Above-ground symptoms, which can be difficult to distinguish from drought symptoms, include reduced vigor, dead leaves that remain attached to the plant, early senescence and yield loss. Irrigation limits damage, but does not prevent colonization. No soybean line is immune to *M. phaseolina*, but a few lines may have moderate resistance. The effect of drought, genotype, and infestation by *M. phaseolina* were determined in a two year study. The soybean genotypes, DT97-4290 and Delta Pineland 4546 (moderately resistant), LS980358 (susceptible), and R01-581F (drought tolerant) were grown in microplots that were infested or not infested with *M. phaseolina* and water-stressed or not stressed in a factorial randomized complete block design experiment with five replications. Soil moisture and temperature, and rainfall were monitored throughout the season. Root colonization and plant growth was assessed at growth stages V2/V3 and V4/V5. Stomatal conductance was measured with a porometer. Canopy temperature was measured with an infrared sensor and used to calculate Crop Water Stress Index (CWSI). Yield and *M. phaseolina* colonization were determined at harvest. When differences among treatments occurred, plants in infested plots had lower stomatal conductance and higher CWSI than those in non-infested plots. In 2009 across stress treatments, infested LS980358 and R01-581F plants had 38% and 49% lower yields respectively than non-infested plots, whereas DT97-4290 and Delta Pineland 4546 did not differ significantly. The results from this study show that infection by *M. phaseolina* affects soybeans at all growth stages and that some genotypes are more sensitive to colonization.

Identification of microsatellites from *Bipolaris sorokiniana*, a pathogen of switchgrass

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Bipolaris sorokiniana, a pathogen of several cereal crops and turfgrasses, is commonly found on switchgrass, causing spot blotch of leaves and root rot. Molecular characterization of this pathogen is limited, and co-dominant markers to study the genetic diversity of this species are lacking. In this work, we developed and characterized microsatellites or simple sequence repeats (SSR) for *B. sorokiniana*. One hundred and ninety-six microsatellite loci were identified from sequencing three hundred and eighty-five positive clones from a small insert genomic library enriched with a biotin-labeled GT₁₂ motif. Primers for one hundred and five loci were developed, and from these, sixteen polymorphic loci were optimized and characterized using fourteen *B. sorokiniana* isolates collected from infected mature switchgrass plants and seeds from several locations in the U.S. The number of alleles observed per locus varied from 2 to 7, with corresponding allelic richness of 0.143 to 0.500. Fifteen of the polymorphic loci from *B. sorokiniana* cross amplified at least one isolate of *Bipolaris victoriorae*, *Bipolaris spicifera* or *Bipolaris oryzae*. High species transferability of SSRs indicates that the microsatellites isolated from one *Bipolaris* species may be useful in genetic studies of closely related species. The development of these co-dominant markers will be a valuable tool for genetic characterization and epidemiological studies of *B. sorokiniana* that infects switchgrass and other host plants. These markers are also expected to be useful in studying the genetic structure of other species in the genus *Bipolaris* as well as species in related genera.

The role of cymoxanil and famoxadone in the management of bacterial spot on tomato and pepper

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Phytopathology 102:S2.3

Chemical control strategies employed in disease management commonly include applications of mixed pesticides. Cymoxanil and famoxadone, the

components of Tanos 50DF (25% a.i. of each product, E.I. du Pont de Nemours and Company, Wilmington, DE) were examined alone and in combination with copper hydroxide and/or mancozeb on the in vitro growth of copper resistant strains of *Xanthomonas perforans*, *X. euvesicatoria*, and a copper sensitive strain of *X. campestris* pv *vitians*. The treatments were also evaluated for disease control of bacterial spot of pepper and tomato, and bacterial leaf spot of lettuce in greenhouse trials. Neither Tanos, nor its individual components had any bactericidal activity. However, the addition of Tanos to copper hydroxide had a modest toxic effect on in vitro growth of a copper-resistant strain of *X. perforans*. It appears that both components are required since the combination of copper hydroxide with either of the components of Tanos did not induce a significant bacterial reduction in comparison to copper alone. Increased disease control was not obtained in greenhouse trials. In most of these trials, the mixture of copper and mancozeb provided a similar level of disease control in comparison to different copper hydroxide mixtures containing, Tanos, cymoxanil, or famoxadone. Thus, mancozeb might not be an essential component in the management of copper-resistant strains of xanthomonads.

Identification of putative TSWV resistance genes and development of gene-specific marker in peanut

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Tomato spotted wilt virus (TSWV) is one of the most destructive viral diseases threatening peanut production in the Southeastern U.S. Among different strategies of controlling this disease, the use of resistant cultivars is more efficient. The objective of this study is to develop putative TSWV resistance gene-specific marker(s) based on peanut EST sequence homology to tomato Sw-5. One peanut EST sequence with approximately 37% amino acid similarity to tomato Sw-5 was further investigated. Two full length cDNA sequences were achieved using 3'- and 5'-RACE, named AhsW-1 and AhsW-2, respectively. Southern blot analysis reviewed that there were at least 4 copies of AhsW genes in the allotetraploid peanut genome. RFLP polymorphisms have been observed among four parental lines of two RIL populations, suggesting PCR-based gene-specific markers could be developed. Three primer pairs have been designed and used to amplify the four parental lines for polymorphism and the RILs for mapping. One PCR-based marker from the AhsW-2 gene appeared to fit 1:1 segregation ratio. Further study will conduct spatio-temporal expression patterns to confirm the possible relationship with the resistance to TSWV. More PCR primers will be used such as recent published Sw5 gene-based markers in tomato. These markers will be used in mapping population and QTL analysis.

Distribution and molecular characterization of *Botrytis cinerea* isolates resistant to respiration inhibitors

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Botrytis cinerea Pers., is one of the most important plant-pathogenic fungi responsible for gray mold diseases on more than 230 plant species worldwide, including strawberry, one of the most widely grown fruit crops in the U.S.A. Control of gray mold disease is largely dependent on the use of fungicides; however, *B. cinerea* represents a classical high-risk pathogen to develop fungicide resistance. Several fungicides are currently labeled for gray mold control of strawberry in the Southeast, including Pristine (boscalid + pyraclostrobin). During the spring-summer 2011, two hundred and sixteen *B. cinerea* isolates were obtained from strawberry fruit with symptoms resembling gray mold disease from 11 commercial fields in North and South Carolina. Boscalid and pyraclostrobin were used separately for *in vitro* fungicide sensitivity assays based on conidial germination. Among the 216 isolates, 133 (61.6%) were resistant; 61 (28.2%) were sensitive, and 22 (10.2%) were highly sensitive to boscalid. With regard to pyraclostrobin, 144 isolates (66.7%) were resistant and 72 (33.3%) were sensitive. A total of 133 isolates were cross-resistant to both fungicides. Resistance to boscalid was correlated with two point mutations at codon 272 (H272R and H272Y), affecting the third Fe-S cluster region of the iron-sulfur protein (sdhB) target of SDH inhibitors. Pyraclostrobin-resistant isolates possessed a single point mutation at amino acid codon 143 (G143A) of the cytochrome *b* protein. The results of the study suggest that frequent applications of respiration inhibitor fungicides such as pyraclostrobin, azoxystrobin and boscalid have selected for resistance in strawberry fields of the Carolina's indicating that rotation with other fungicide classes need to be implemented.

Identification and analysis of differentially expressed maize WRKY transcription factors in response to *Aspergillus flavus* colonization of resistant and susceptible germplasm

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Phytopathology 102:S2.4

Successful colonization of maize (*Zea mays* L.) by the opportunistic fungal pathogen *Aspergillus flavus* results in the contamination of maize kernel tissues with carcinogenic mycotoxins known as aflatoxins. Aflatoxins pose a significant threat to human and animal health and are the cause of heavy crop losses annually. Maize resistance to such fungal pathogens has been shown to be controlled by a complex network of regulatory pathways resulting in the expression of many genes coding for resistance-associated proteins (RAPs). One such regulatory network is the WRKY transcription factor network. WRKY transcription factors are characterized by their distinctive N-terminal amino acid sequence of 'WRKY' which contributes to the DNA binding specificity of this class of transcription factors. This DNA binding domain recognizes highly conserved sequences referred to as W-boxes, which have been found within the promoter regions of some RAP-coding genes. Recently, these transcription factors have been found to be differentially expressed in kernels of resistant and susceptible germplasm colonized by *A. flavus* using oligonucleotide microarray studies. In this study, the expression of eight WRKY genes was further investigated under field control and *A. flavus* inoculation conditions using quantitative/real-time polymerase chain reaction (qPCR). These genes were previously shown to be induced in *A. flavus* infected kernels. Preliminary results of this study have revealed that several of these genes are significantly regulated in both the susceptible and resistant germplasm. Further analyses of these expression patterns and the degree of homology of these select WRKY transcription factors to those from other model organisms, such as rice (*Oryza sativa*), may also shed light on the specific mechanisms resulting in the expression of maize RAPs in response to *A. flavus* colonization.

Comparative genomic analyses of the rice pathogenic *Burkholderia glumae* strains reveals plasticity among the genomes

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Burkholderia glumae, the causal agent of bacterial panicle blight of rice (*Oryza sativa* L.), is a major pathogen that reduces the yield and quality of rice, world over. To better understand the genome-scale characteristics particularly that of its pathogenicity, the genome of a highly virulent strain found in Louisiana, 336gr-1 was sequenced using Illumina Genome Analyser II. It was then aligned and analyzed with the genome of a previously sequenced Korean strain, BGR1. Comparative analyses with the East Asian strain BGR1 strain revealed unique regions present in the two geographically isolated genomes. These unique regions and also the genes for major virulence factors in both these strains were compared to identify important regions of genomic variation. The unique regions of 336gr-1 genome were annotated and the protein-coding genes were identified. PCR analysis using primers specific for the unique regions and other genes of interest further validated these results. Significant evidence of evolution by genome plasticity within *B. glumae* has been observed. Comparative genomic analyses of these pathogenic strains give an insight into how they adapted to different ecological niches and host plants, particularly the rice varieties. These findings may help explain the diversity of *B. glumae* genotypes and its host ranges, and augment elaborate characterization of its ecology and pathogenesis.

Evaluation of ICRISAT varieties for resistance to foliar peanut diseases in Haiti

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Peanut rust and late leaf spot, caused by *Puccinia arachidis* and *Cercosporidium personatum*, respectively, are important foliar diseases of peanut (*Arachis hypogaea* L.) throughout Haiti. A varietal trial using genotypes developed by ICRISAT was conducted in Haiti during 2010 (Bois Rouge) and 2011 (UCNH and Isle Adam) to investigate the effectiveness of resistance to reduce disease intensity and yield loss. Fifteen genotypes from ICRISAT were evaluated in the experiments each year. Susceptible standards for each trial included Tamsan (Bois Rouge), Tifguard (UCNH), and local runner and

Valencia types (Isle Adam). Peanuts were hand planted according to local standards, and grown under rainfed conditions; no fungicide applications were made. In 2010, all ICRISAT lines had similar levels of leaf spot and rust, and all had less disease than Tamsan. All 15 had higher yields than Tamsan, and the highest yields were from ICRISAT lines ICGV 99036 and ICGV99046. In 2011 (UCNH), only peanut rust was present and all 15 ICRISAT lines had less disease than Tifguard; ICGV 99036 had less rust than any other genotype. Yields were again significantly higher in ICGV 99036 and ICGV99046. At Isle Adam, all ICRISAT lines had much lower leaf spot ratings than the local lines. Rust ratings were also significantly less for all ICRISAT lines, although the local Valencia was much more susceptible than the local runner. This study verifies that the high levels of resistance to foliar diseases reported in ICRISAT lines are effective in Haiti. These lines also have much greater yield potential than improved cultivars from the United States when grown without fungicides in Haiti.

Evaluation of soybean recombinant inbred line (RIL) derived sister lines for resistance to *Phakopsora pachyrhizi*

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Phakopsora pachyrhizi, the causal agent of Asian soybean rust, was first discovered in continental U.S. in late 2004. *P. pachyrhizi* has the potential to cause severe yield losses as all U.S. commercial soybean varieties are susceptible. In this study, ten recombinant inbred line (RIL) derived sister lines of two different populations (RN06-32-2 and RN06-16-1), which had previously been evaluated against Florida soybean rust isolates, were evaluated for differences in resistance to infection by *P. pachyrhizi* (Louisiana isolates) under both growth chambers and greenhouse conditions. For each line, sixteen plants were evaluated at R1 stage through inoculation with 200 µl of rust spore suspension (3×10^4 spores/ml) per leaf on the upper surface. For growth chamber detached leaf assay, soybean leaves at R1 stage were inoculated in the same manner. Fifteen days after inoculation, plants in the greenhouse and the detached leaves in growth chamber were evaluated for lesion appearance, pustule formation, and pustule eruption and density. Significant differences were observed among sister lines in their responses to *P. pachyrhizi* infection under both conditions. The lines 15-b and 16-c of population RN06-16-1 and 94-c line of RN06-32-2 population, which showed immune reaction to Florida rust isolates, exhibited a resistant response against Louisiana rust isolates. Whereas, the 8-a and 8-b lines of population RN06-32-2 showed the same immune response to Louisiana rust isolates as to Florida rust isolates. Lines 15-c and 16-b of population RN06-16-1 showed moderately resistant response. Lines 94-a and 94-b of population RN06-32-2 showed the same susceptible and resistant response, respectively, against Louisiana rust isolates as to the Florida rust isolates. Currently, protein profiles of these lines are being compared to understand the compatible and incompatible host-pathogen interactions at the molecular level using proteomics.

Characterization of a recombinant inbred line (RIL) population for resistance to late leaf spot in cultivated peanut (*Arachis hypogaea*)

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Late leaf spot (LLS), caused by *Cercosporidium personatum*, is an important disease of peanut (*Arachis hypogaea*). A field study was conducted in 2011 to characterize leaf spot resistance in a recombinant inbred line (RIL) population developed from a cross between Gregory, a Virginia type cultivar and Tifguard, a runner type cultivar. Gregory has been previously characterized as susceptible and Tifguard is reported as moderately resistant to the disease. This biparental population consisted of 78 F_{7.8} lines and was grown with the parents in three replications in a non-sprayed field trial. The leaf spot intensity was assessed four times during the season using the Florida 1-to-10 severity scale. Final leaf spot severity ratings ranged from 4.25 to 8.33. The genotypes showed a nearly normal distribution curve for leaf spot resistance, which documents the quantitative nature of the trait. The final rating showed Tifguard, the moderately resistant parent, as the most resistant genotype and the lines - CB 15A and CB 23A as the most susceptible genotypes among the 80 lines. Interestingly, nearly 67 percent of the population was found to be more susceptible than the susceptible parent Gregory. This observation can be attributed to transgressive segregation and the possible inheritance of susceptible alleles from the resistant parent. This RIL population will be genotyped using SNP (single nucleotide polymorphism) and SSR (simple sequence repeat) markers. The genotypic and the phenotypic data will help generate marker-trait associations for genetic mapping and marker development for LLS resistance in elite U.S. cultivars of peanut.

Fungal occurrence associated with foliar lesions on giant miscanthus in Mississippi

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Giant miscanthus (*Miscanthus × giganteus* Greef et Deu.) is a tall perennial grass currently in production stages as a biomass feedstock crop for alternative energy sources. Long term production and sustainability depends on low-input management to generate positive net-energy gains. Consequently, potential pathogens may threaten the sustainability of giant miscanthus. In the summer of 2010 foliar diseases displaying variable symptoms were observed in giant miscanthus yield trials in Starkville, Mississippi. This study was initiated to better understand the fungi associated with giant miscanthus. A survey was completed in 2011 to monitor fungal frequency in 'Freedom', 'Illinois' and 'Nagara' giant miscanthus. Lesions were excised from mature leaves and surface disinfested with 0.6% NaOCl and rinsed with sterile distilled water prior to plating on 1.5% water agar amended with 100 mg/L of chloramphenicol and streptomycin sulfate. Fifty random lesion segments were plated per giant miscanthus plot. Lesions were incubated under continuous fluorescent light on a laboratory bench to promote sporulation. Following 21 days, fungi were identified based on the presence of fruiting structures using light microscopy. Hyphal or spore transfers were made to potato dextrose agar. The internal transcribed spacer (ITS) regions of ribosomal DNA of selected fungi were amplified with PCR with primers ITS1 and ITS4 to confirm identity. Fungi most frequently isolated from giant miscanthus were *Alternaria*, *Phoma*, and *Stagonospora*. Species of *Bipolaris*, *Colletotrichum*, *Leptosphaeria*, *Pithomyces*, and *Nigrospora* were also isolated, however at lower frequencies. Pathogenicity of these fungi on giant miscanthus is currently being investigated.

Ecology and epidemiology of bacterial pathogens

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Interpretation of epidemiologic patterns of disease can be used to identify life-cycle events and develop management recommendations for phytopathogenic bacteria. For example, seed was originally suspected as the primary source of inoculum for center rot of onion (*Allium cepa*), caused by *Pantoea ananatis*. However, higher disease incidence in dry years, disease gradients, and lack of secondary spread from inoculated plants in the field did not fit patterns associated with seedborne diseases. Because of the epidemiologic patterns observed, additional research was conducted and thrips were shown to transmit the bacterium. Eventually, recommendations of insect control and eradication of nearby weeds were developed for management of center rot. Seed health assays are a key component of management strategies for many bacterial diseases. However, seedborne bacteria continue to plague growers despite improvements in seed health assays. Populations of phytopathogenic bacteria recovered from individual seeds show that seedborne bacteria occur in highly-skewed, inverse-Gaussian distributions. To maintain a 95% confidence level for a seed health assay, seeds harboring lower concentrations of bacteria require larger sample sizes due to detection threshold limits of laboratory-based assays. Low inoculum densities also could impact grow-out tests by extending incubation periods. The impact of low levels of inoculum in seed is much the same as that described for inoculum surviving sanitation, i.e. the benefit, in terms of a delay in disease development, is proportional to the logarithm of the sanitation ratio if one assumes an exponential increase in disease over time. Reduction in initial inoculum must be enormous to significantly impact polycyclic diseases, one possible explanation of why seedborne diseases continue to be a problem. Finally, a five-step management program based on fertility, weed control, copper-sprays, field-curing and storage practices to manage bacterial streak and bulb rot of onion, caused by *Pseudomonas viridiflava*, will be presented.

Genetic diversity of *Phytophthora capsici* recovered from pepper in Southern France

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Phytophthora capsici is a soil-borne oomycete plant pathogen that attacks important vegetable crops. Both sexual and asexual reproduction plays a significant role in *P. capsici* population dynamics and disease epidemiology. The genetic structure varies depending on the region under investigation. Our objective was to investigate the genetic diversity and survival mechanisms of the pathogen obtained at two locations in Southern France. A total of 63 *P. capsici* isolates were recovered from roots or crowns of pepper grown in high

tunnels in the Provence-Alpes-Côte d'Azur region. Both mating type were obtained from both locations and none of the isolates showed resistance to mefenoxam. Genotypic analysis using 10 SNP loci revealed a total of 35 multi-locus genotypes, of which ten were clonal lineages and 25 were unique genotypes. Clonal lineages were confined to the same locations and collection period and it appears that sexual and clonal reproduction play an important role in *P. capsici* disease epidemic development.

Production practices impact anthracnose severity and biomass yield of selected sorghum varieties

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A study was conducted on a site in 2009 that had been cropped to sorghum in 2008. A strip-split plot design with 4 replications: (1) non- or irrigation regimes, (2) tillage systems (conventional and conservation), and (3) 3 sorghum varieties. Varieties were 1990 (a photoperiod sensitive forage sorghum), SS506 (forage), and NK300 (a silage/grain sorghum). Conservation plots were in-row subsoiled with a narrow-shanked subsoiler, while conventional plots were disked/leveled using a tandem disk harrow. Biomass samples were collected at 18 and 24 weeks after planting, where 1.5 m row length was hand harvested from each of the two middle rows. Anthracnose severity was assessed on Aug 10 and Sep 10 on a 1 to 9 scale based on the % of leaf area blighted (where 9 is high). As indicated by a significant sorghum variety × tillage interaction, anthracnose severity on Aug 10 was higher on all sorghum varieties under conservation than conventional tillage, with SS506 having the highest disease ratings at both rating dates. While higher anthracnose ratings were noted for the irrigated than rainfed 1990 sorghum on Sep 10, equally high severity ratings were noted for SS506 under both irrigation regimes. At the 18th wk harvest date, biomass yield was impacted by variety selection and irrigation but not tillage, while variety selection but not irrigation and tillage influenced yield at 24 wk.

Genetic dissection of the regulatory network that controls virulence genes of *Burkholderia glumae*

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Burkholderia glumae causes bacterial panicle blight (BPB), which is one of the most serious rice diseases in the southern United States including Texas, Arkansas, Mississippi and Louisiana. Because the importance of this pathogen was recognized recently, its virulence mechanism has been understudied compared to other major rice pathogens. ToxI/ToxR quorum-sensing system mediated by LuxI/LuxR-family proteins is known as the key element that controls all the known virulence factors of *B. glumae* including the phytotoxin, toxoflavin, and lipase. We initiated and have performed molecular genetic and genomic studies of *B. glumae* to gain a comprehensive knowledge on the virulence system of this pathogen. Novel regulatory factors discovered from our research include PidS/PidR two-component system, Clp protease and NtpR. *pidS* and *pidR* encoding PidS/PidR two-component system positively regulate the production of toxoflavin, while *clp* and *ntpR* encoding Clp protease and NtpR, respectively, negatively regulate it. Functional characteristics of these newly found regulatory factors and their relationship with known regulatory factors, particularly ToxI/ToxR quorum-sensing system, are currently being investigated. New regulatory pathway(s) which controls the production of virulence factors in a ToxI/ToxR quorum-sensing independent manner was also discovered recently, indicating that the regulatory circuit controlling *B. glumae* virulence genes is far more complex than previously known. In addition, comparative genomic and transcriptome analyses using an Illumina high-throughput sequencing platform are being conducted to better understand genomic variations among strains having different geographic and phenotypic backgrounds as well as transcriptional dynamics dependent on various regulatory factors and environmental conditions.

Evaluation of nitrogen rate effects on cotton yield in soil management zones treated and untreated with Telone® II

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Root-knot nematodes (*Meloidogyne incognita*) and reniform nematodes (*Rotylenchulus reniformis*) commonly occur in cotton fields in the mid-south. Control recommendations are based on nematode species present, soil types,

nematicide efficacy, economics, and other variables. Research results show that the use of soil maps and EC_a data can be used effectively to identify low and high risk, or “responsive” zones. As a result, input costs can often be reduced by 30–40% by using this technological approach to assist in nematode management. This study was initiated to evaluate cotton yield responses when varying rates of nitrogen were applied in soil management zones treated and untreated with Telone® II. Nitrogen was applied at the 3 different rates to treated and untreated 6 row strips across a field. Yield monitor data from this trial demonstrated the positive effects and value of the Telone II application in all nitrogen levels and soil management zones tested. ®Trademark of Dow AgroSciences LLC. Telone II is a Restricted Use Pesticide. Always read and follow label directions.

Host and non-host infection by *Cercospora zae-maydis* and *Cercospora beticola*: Host sensing, stomatal tropism and initiation of stomatal penetration

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Grey leaf spot of maize caused by *Cercospora zae-maydis* (*Czm*), and *Cercospora* leaf spot of beet caused by *Cercospora beticola* (*Cb*) are economically important plant diseases found worldwide. Both pathogens penetrate stomata to cause disease, and initial observations suggest they exhibit tropism toward stomata. To more thoroughly characterize these processes, specific biological parameters underlying infection were quantified. Hypha from germinating conidia grew more slowly and formed fewer branches in the first two days of infection on host leaves compared to conidia on non-host leaves and plastic leaf replicas, suggesting an early response to the leaf environment. Between 3–7 days after inoculation, both pathogens grew extensively on host and non-host leaf surfaces, and re-oriented hyphal tip growth or initiated lateral hyphal branches directed toward stomata within a distance of approximately 50 µm of the stomate being targeted for infection. Upon encountering stomata of either host, *Czm* formed appressoria and *Cb* penetrated stomata directly at approximately 75% efficiency, but perception of stomata appeared to be abolished on plastic leaf replicas. Initiation of infection through host and non-host stomata suggests the presence of a broadly conserved stomatal sensing mechanism that precedes the host-specific necrotrophic growth habit common among *Cercospora* spp. This research represents the first quantification of stomatal tropism in *Cercospora* spp., and provides a framework to dissect early events underlying infection at the molecular level.

Tracking asymptomatic foliar infections of Pecan Anthracnose

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Colletotrichum gloeosporioides causes anthracnose of pecan (*Carya illinoensis*) and infects leaves after long wet periods and temperatures of 25–30 °C. These infections remain dormant until favorable conditions allow them to form necrotic lesions. To expose these latent infections, pecan (cv. Desirable) leaves were placed in –10 °C for 5 hours in order to kill the tissue. They were then placed in 0.5% sodium hypochlorite solution for 1 min., rinsed in distilled water, placed in 70% ethanol for 1 min., and then rinsed again. The treated leaves were incubated in moist chambers for two weeks, then rated as to the number of sporulating lesions. Bi-weekly samples from two sites in Georgia showed that there were no foliar lesions until early August when the average number of lesions per leaf went from 0 to 5.9, and to 33.3 two weeks later. The pecan cultivars Amling, Stuart, Pawnee, Excel, Desirable and Zinner had 0.7, 4.7, 5.5, 20.0, 61.0 and 65.6 latent infections per leaf, respectively.

Investigating effective management strategies for *Ustilaginoidea virens* on *Oryza sativa*

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Rice false smut, caused by *Ustilaginoidea virens* (teleomorph *Villosiclava virens*) is now established in all rice growing counties of Arkansas. The disease cycle for false smut on rice was poorly understood. However, recent histological and PCR evidence show that *U. virens* is both soil and seed borne and that it colonizes rice seedlings soon after emergence. Plants infected at this stage maintain this association, without causing symptoms, until eruption of sporophores on panicles at heading. It has been proposed that screening rice seedlings for the presence of *U. virens* with PCR could provide a rapid and effective tool to identify effective seed treatments. The main objective of our study was to investigate the efficacy of various seed treatments in the field and greenhouse. In this study, we tested demethylation inhibitors (DMI),

quinone outside inhibitors (QoI) and Cu in *in-vitro* petri-plate growth assays, greenhouse studies and in field studies to identify effective materials and concentrations of those materials. In 2011, replicated experiments were conducted with infested seeds treated with selected fungicides in soils from Newport and Pine Tree, Arkansas. In these tests, seedlings were harvested three weeks after emergence from each replicate of all treatments grown in the greenhouse and field. The sampled seedlings were then screened for the presence of *U. virens* using PCR as a diagnostic tool as previously reported. Infection levels based on PCR results were determined from the number of seedlings infected (with positive amplicons [= 232 bp bands]) from each treatment. In the field, disease levels were determined as the number of infected heads per m². Preliminary data suggest that an effective disease management strategy may include using seed treatments or seed treatments followed by foliar applications under prevailing field conditions.

Genetic and phenotypic variations among *Burkholderia glumae* strains, the causal agent of bacterial panicle blight of rice

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The genus *Burkholderia* shows considerable amount of variations ranging from plant and animal pathogenic bacteria to non-pathogenic soil inhabiting species. Genetic and phenotypic variations are evident within the strain level as well. *Burkholderia glumae* is primarily a rice pathogen causing significant yield reduction if the rice flowering season coincides with favorable weather conditions. In the southern rice producing United States, *B. glumae* is becoming a leading rice pathogen. Previously, more than 240 *B. glumae* strains were isolated from rice fields in Louisiana, Arkansas and Texas and tested for their virulence. In this study, phenotypic and genotypic variations among *B. glumae* strains were examined by virulence assays on rice and onion, pigments production, pulse field gel electrophoresis (PFGE) and BOX-PCR and antifungal activity. Considerable level of variation on virulence assay was observed among the strains ranging from avirulent to hyper-virulent. Here we report that, there is variation in pigments production of *B. glumae* strains in certain nutritional conditions, including King's B and CPG media. Our PFGE and BOX-PCR results showed that even *B. glumae* strains showing similar phenotypes are different genetically. The investigation of antifungal activity of *B. glumae* strains against *Rhizoctonia solani* also showed significant variations among strains tested. Some of the strains showed promising results as a future agent for bio-control.

Impact and management of fungal foliar diseases of cotton in Georgia

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Historically, foliar diseases of cotton in the southeastern United States have been considered to be of only minor importance and have not been treated with fungicides. Beginning with interest in Fusarium hard lock in Florida and later with the development of pyraclostrobin and azoxystrobin, attempts have been made over the past decade to reduce the severity of foliar diseases affecting cotton and to improve yields with judicious use of fungicides. Based upon the results from multiple studies reported here, use of fungicides has not been shown manage *Stemphylium* leaf spot; this disease is clearly linked to a nutrient deficiency, typically potassium. Fungicides have been successfully used to manage areolate mildew in eastern Georgia but with unclear impact on yield. In 2008, significant defoliation of the cotton crop was attributed to a newly recognized disease of cotton in Georgia, *Corynespora* leaf spot. Subsequent field trials over three years have documented that post-bloom applications of pyraclostrobin, 6 fl oz/A, can significantly reduce the severity of *Corynespora* leaf spot on leaves, reduce premature defoliation, and significantly improve yields when symptoms are severe. When symptoms are mild, the impact of the disease on yield is less certain. Based upon these studies, applications of pyraclostrobin and perhaps azoxystrobin are likely appropriate for growers in Georgia where risk to *Corynespora* leaf spot is considered elevated.

Effect of planting date, fungicide timing and varietal tolerance on development of narrow leaf spot of rice

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Narrow brown leaf spot of rice is a fungal disease caused by *Cercospora janseana* L. Miyake. It causes long narrow spots on leaves, sheaths, glumes and seeds of rice plants. Investigations on epidemiological aspects of *C. janseana* have been sparse due to low incidence of disease in past few decades. Recent increases in disease pressure have been reported and are a serious concern for growers and researchers. Studies have been conducted to determine the effect of planting date, fungicide timing and varietal tolerance on development of narrow brown leaf spot disease at the Rice Research Station, Crowley. To accomplish the objectives, five varieties; Della, Cheniere, CL111, CL131 and CL151 were planted in Mid-April and Mid-May. Fungicide treatments of propiconazole (Tilt at 6 fl.oz/acre), plus an untreated check, were applied at panicle initiation, early boot and late boot stages. Weekly observations were recorded on disease incidence, disease severity in lower, middle and upper canopy. Preliminary results of this study have shown significant differences among varieties, treatments and planting dates. Disease severity was significantly higher in rice of the May planting date as compared to the April planting date. Among the varieties, disease severity in all levels of the canopy was found higher in CL111 and lowest in Della at both the planting dates. Significantly lower yield loss was found at the April planting date with treatment at the early boot stage and on the second planting date at panicle initiation stage.

Are emaraviruses mite viruses? The Rose rosette paradigm

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Rose rosette (RR) is a devastating rose disease, first described in the 1940s. RR is prevalent east of the Mississippi River and is characterized by elongated stems, malformed flowers and leaves, excessive thorns and witches' broom culminating in plant death in a few years. In the Midwest and the Southern United States, the main host is an invasive species, *Rosa multiflora* Thunb., though cultivated roses are also afflicted. RR has previously been shown to be transmitted by the eriophyid mite *Phyllocoptes fructiphilus* and has been proposed as a biological control agent for *R. multiflora*. Recently, a new member of the genus *Emaravirus* (family *Bunyaviridae*), named Rose rosette virus (RRV), has been found in close association with RR symptomatic roses. Surprisingly, little diversity was seen in virus populations collected from eight different states. Given the low diversity and the fact that the genus *Emaravirus* belongs to the family *Bunyaviridae*, members of which replicate in their arthropod vector, it has been hypothesized that RRV replicates in its vector, *P. fructiphilus*. In order to facilitate testing this hypothesis, a quantitative RT-PCR assay was developed for both the genomic and virus complementary RNA. *P. fructiphilus* mites were collected from virus-positive *R. multiflora* and total nucleic acids were extracted. The RNA was then subjected to quantitative RT-PCR for the genomic or virus complementary RNA strands. Additionally, the presence of capped virus mRNA in the mites was investigated using 5'RACER. The replication of RRV in its vector makes using RRV as a biological control agent unwise and care should be given to prevent the spread of RRV and its vector outside its current geographic range.

The in vitro evaluation of fungicide systemicity for the control of *Sclerotium rolfsii* on tomato with soil and foliar applications

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Two experiments were conducted under greenhouse conditions to evaluate acropetal and basipetal movement of several fungicides. Fluopyram, penthiopyrad, flutolanil, prothioconazole, fluazinam, pyraclostrobin, and pentachloronitrobenzene were applied once, either as a foliar spray or as a soil drench, at recommended rates to greenhouse grown tomatoes. Several tissue types were excised and inoculated with *Sclerotium rolfsii* five days after application to determine fungicide movement. Inoculated tissues were incubated at 30°C and lesions measured after 48 hours. As a soil drench, flutolanil, pyraclostrobin, fluazinam, and penthiopyrad significantly reduced colonization at the crown by 100%, 90%, 89%, and 68%, respectively, compared to the control. Prothioconazole and pentachloronitrobenzene reduced colonization by 39% and 29%, respectively, but were ineffective in reducing colonization of other tissues. Flutolanil significantly reduced colonization on the main stem (94%), branches (100%), and leaves (74%). Fluopyram did not reduce colonization when applied as a soil drench. As a foliar spray, flutolanil and fluazinam significantly reduced colonization on leaves (94% and 93%), branches (100% and 62%), stems (84% and 77%), and crowns (80% and 73%) compared to the control. Penthiopyrad significantly reduced colonization on leaves (80%), branches (76%), and main stem (68%), but was ineffective at reducing colonization at the crown compared to the control. Pyraclostrobin and prothioconazole reduced colonization of leaves (61% and 50%), branches (70% and 37%), main stem (50% and 42%), and

crowns (49% and 34%). Fluopyram significantly reduced colonization on leaves and branches 48% and 35%, respectively, but did not significantly reduce colonization on the main stems and crowns. Pentachloronitrobenzene failed to reduce colonization of all tissue types when applied as a foliar spray. Results indicate that flutolanil and fluazinam can reduce disease in the lower canopy when applied to the foliage. Similarly, flutolanil may also provide suppression of foliar diseases when applied as a soil drench.

Characterization and detection of a new *Botrytis* species from blackberry

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Botrytis spp. cause blossom and fruit diseases on many crops, including blackberry. In 2011, 404 isolates from blackberry and strawberry were collected from North and South Carolina and single spore colonies were characterized. Two distinct species, *B. cinerea* and another undescribed species, were identified based on examination of the coding G3PDH, HSP60 and RPB2 genes and based on morphological characters. The new species formed pale yellow to white colonies with short aerial mycelia and produced black sclerotia on PDA at 20°C. Phylogenetic analysis based on combined DNA sequence data showed that the novel *Botrytis* sp. is most closely related to *B. fabiopsis*, the causal agent of gray mold disease of broad bean, and *B. galanthina* from snow drop. Its conidia, however, are smaller than conidia of *B. fabiopsis* and *B. galanthina* and sequence analysis of genes encoding necrosis and ethylene-inducing proteins (NEPs) also indicated that the novel *Botrytis* spp. is distinct. The new species is pathogenic on broad bean leaves, indicating a broad host range and providing further evidence for its biological distance from *B. galanthina*. Inoculation of blackberry fruit with conidia caused typical gray mold symptoms but compared to *B. cinerea* the latent period was significantly longer. A PCR method was developed to distinguish *B. cinerea* from *B. caroliniana*, however, some cultural characteristics can also be used for rapid distinction.

Identification and characterization of expressed resistance gene analogs (RGAs) from peanut (*Arachis hypogaea* L.) expressed sequence tags (ESTs)

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Cultivated peanut (*Arachis hypogaea* L.) is an important food legume grown worldwide for providing edible oil and protein. However, due to scarcity of genetic diversity, peanut is very vulnerable to a variety of pathogens, such as rust (*Puccinia arachidis* Speg.), early leaf spot (*Cercospora arachidicola* S. Hori), late leaf spot (*Cercosporidium personatum* Berk. & M.A. Curtis), and Tomato spotted wilt virus (TSWV). Large number of peanut ESTs (225,264 ESTs by 11-11-2011) has been developed in recent years by peanut community, providing resources for gene identification and marker development. Based on the homology to typical pathogen resistance genes and the conservative domains/features, we identified 401 resistance gene analogs (RGAs), using a stringent BLAST search. By using 54 known R-gene protein sequences, we screened peanut EST database and identified 995 ESTs targeting different classes of known R genes. Among the 995 ESTs, 559 (56%) are from cultivated peanut (ten genotypes) and 436 (44%) are from three wild peanut genotypes. After assembly, 401 unigenes including 152 contigs and 249 singletons were identified and considered as peanut expressed RGAs. These expressed RGAs are comprised of 75, 196, 89, 27, and 14 RGAs representing for NBS-LRR, protein kinase (PK), LRR-PK/TM, Toxin reductase, and other domain encoding R genes, respectively. The expressed RGAs identified in this study provide a large sequence dataset for RGA-tagged potential markers that could be developed further for use in mapping and ultimately cloning disease resistance genes in peanuts.

Characterization and genetic analysis of rhizobacteria antagonistic to plant pathogens

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Phytopathology 102:S2.7

Numerous antagonistic bacteria have been isolated from suppressive soils in Mississippi. These bacteria showed significant inhibitory activity to some plant pathogenic fungi and bacteria. Preliminary analyses indicated that these bacterial isolates belong to the genera *Burkholderia*, *Pseudomonas*, *Bacillus*, *Enterobacter*, and *Streptomyces*. Further characterization and genetic analysis of the bacteria are under way. Strain MS14 was determined to be a member of

Burkholderia contaminans. Genetic analysis revealed that a 56-kb *ocf* gene cluster is required for production of antifungal activity by *B. contaminans* strain MS14. The gene cluster includes the genes encoding nonribosomal peptide synthetases, the bacterial LuxR regulatory proteins, an ATP-binding cassette, and a xylosyltransferase. Mutagenesis and chemical analysis revealed that an oligopeptide, occidiofungin, is responsible for the antifungal activity of strain MS14. The backbone of occidiofungin is composed of eight amino acids and a xylose, and four variants have been found from the MS14 culture. Purified occidiofungin showed a broad range of pathogenic fungi and its mode of action was predicted to be disruption of biosynthesis of the fungal cell wall. The findings of the project will provide valuable insights to developing novel fungicides and antibiotics for agricultural applications.

Transposon mutagenesis of *Burkholderia glumae* 336gr-1 and screening of mutant derivatives identified *ntpR*, a novel negative regulator of toxoflavin production

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Phytopathology 102:S2.8

Burkholderia glumae is the major causal agent of bacterial panicle blight of rice, a disease that can cause severe yield losses, and is becoming an increasingly important pathogen worldwide. Several virulence factors, including toxoflavin, lipase, flagella, and a type III secretion system, have been shown to be regulated by a quorum-sensing system and are required for full virulence in *B. glumae*. In an effort to identify additional virulence factors and regulatory factors of how this pathogen causes disease, mutant derivatives of a virulent *B. glumae* strain, 336gr-1, were screened on various culture media following transposon mutagenesis. A putative open reading frame encoding a LysR-type family protein was found to be disrupted in two independent mutants showing enhanced toxoflavin production and virulence. In order to verify the function of this gene, named *ntpR*, that is a novel negative regulator of toxoflavin production in *B. glumae*, additional independent mutants of this gene will be created via homologous recombination and then complemented with a functional *ntpR* clone. Further characterization of the regulatory function of *NtpR* on other known virulence factors, such as lipase, flagella, and a type III secretion system, and its relationship with other global regulatory factors, such as a quorum-sensing system, will also be determined.

Mapping the extent of Loblolly Pine Decline in Alabama and Georgia

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Phytopathology 102:S2.8

Pine Decline is a decline complex that poses a threat to the forests and economy of the southeast United States. This slow mortality is not new to the area, but it has just begun to be fully understood and proper management assessed. A comprehensive view of predisposing site factors needs to be developed to act as a tool in managing forests and investments. Once the symptoms are visible, the forested stand is at risk of continued mortality. Previous research has identified the symptomology, fungi, insect vectors, and predisposing factors of this decline. My project will utilize this information to further understand slopes and aspects predisposing stands to this decline, and create a comprehensive map of these sites in Alabama and Georgia, where the decline is most prevalent. This map will be created using a GIS, Forest Inventory Analysis Data (U.S. Forest Service), spatial analyst models, and current data collected from the Forest Health Dynamics Laboratory at Auburn University. It will serve as a valuable tool to understand sites that are already at risk for Pine Decline and thus the proper allocation of resources for management practices. It will also serve as a guide for proper tree species placement to reduce future Pine Decline.

Visual and electronic comparisons of detached strawberry leaves inoculated with two *Colletotrichum* species

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Phytopathology 102:S2.8

Inoculation of detached strawberry leaves with *Colletotrichum* species may provide an accurate, rapid, non-destructive method of identifying anthracnose resistant germplasm. The purpose of this study was to compare two methods for screening anthracnose resistance. Detached leaves of 60 susceptible and 25 resistant strawberry clones were inoculated with one *Colletotrichum gloeosporioides* isolate and two *C. fragariae* isolates. Anthracnose disease symptoms were assessed quantitatively via computer based image analysis and qualitatively by two independent raters and the assessments were compared statistically. There was a strong positive correlation between

percent lesion area determined by the computer analysis and the visual disease scores of the two raters. Image analysis provided a precise measurement of percent lesion area of infected leaves while visual assessment provided rapid results.

Relationships among defoliation by leaf spot, canopy reflectance and yield in new runner-type peanut cultivars

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Phytopathology 102:S2.8

Early and late leaf spot caused by *Cercospora arachidicola* and *Cercosporidium personatum*, respectively, can cause severe losses on susceptible peanut (*Arachis hypogaea*) cultivars. Losses to leaf spot have been correlated with defoliation late in the season and canopy reflectance measured in the near-infrared wavelengths. Recently, several new peanut cultivars have been released with excellent yield potential and field resistance to tomato spotted wilt virus. However, the relationships among late-season defoliation, canopy reflectance and yield have not been characterized for these cultivars. To examine this relationship, field experiments were conducted in 2011 in Attapulgus and Tifton, GA. In each experiment, four cultivars, Florida-07, Georgia-06G, Georgia-07W, and Tifguard, were combined in factorial arrangement with four fungicide treatments, 7, 4, and 3 applications of 1.1 lb ai/A of chlorothalonil with initial applications ca. 35 days after planting, and subsequent applications ca. 14 day intervals. Applications of 1.0 lb ai/A of flutolanil were made at ca. 60 and 90 days after planting in each trial to minimize effects of *Sclerotium rolfsii* on yield. Multiple visual leaf spot ratings were made to estimate the levels of defoliation. On each evaluation date, canopy reflectance in the visible and infrared light spectrum was measured using a Crop Circle crop scanner with an ACS-210 active sensor. Late leaf spot was the predominant foliar disease in both trials. All of the cultivars evaluated were able to maintain good yields even with moderate to heavy defoliation, with percent yield losses to leaf spot less than previously reported from late leaf spot on the previous standard cultivar, Florunner. Percent defoliation was correlated with canopy reflectance in the infrared and visible bands and with the normalized difference vegetative index calculated from those two measurements.

Evaluating peanut rust resistance in newly developed peanut breeding lines

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Phytopathology 102:S2.8

Host resistance is the best option for managing the peanut (*Arachis hypogaea*) rust disease, caused by the fungus *Puccinia arachidis* Speg, especially in low-input peanut producing countries with warm, tropical climates. In 2011, the field resistance of newly developed peanut CRSP breeding lines was evaluated in two locations: Gainesville, FL and Tifton, GA. The peanut rust severity was assessed using the 1-9 ICRISAT rating scale. The disease pressure in Gainesville was low, which resulted in few differences in rust severity among genotypes. Genotype BayoGrande was the only genotype with a consistent susceptible score compared to 2010. In Tifton, the disease epidemic started relatively late for the disease pressure to be high enough to distinguish between genotypes. There was consistency in rust severity scores between the two locations, however. The components of resistance to *P. arachidis* to explain the mechanism behind the resistance, was carried out using a detached leaf study. Detached leaflets were placed on moist sterile paper in sterile Petri dishes and inoculated with *P. arachidis* spores, after which they were incubated for 31 days at 25°C, with a 12 hour photoperiod. Genotypes 97x36-HO2-1-B2G3-1-2-2, BOL19-b5 and 99x33-1-B2G-13-1-1 were among the genotypes with the lowest numbers of pustules at 31 days after infection. Additional results on the components of resistance will be added.

Antagonistic activities of rice-associated bacteria against *Burkholderia glumae* and *Rhizoctonia solani*

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Phytopathology 102:S2.8

Bacterial panicle blight (BPB) and sheath blight (SB), caused by *Burkholderia glumae* and *Rhizoctonia solani*, respectively, are the major rice diseases in the southern United States and can cause severe yield losses. There are no

effective control measures for BPB. Only a few partially resistant cultivars are currently available. Seed treatment and foliar sprays with oxolinic acid have been used for the management of BPB in Japan, but oxolinic acid is not labeled for use in the U.S. In addition, *B. glumae* has been shown to develop resistance to oxolinic acid. Similarly, fungicides have been used to control the SB which has harmful residual effects on environment. For this reason, biological control is a promising venture for BPB and SB disease control. In an effort to isolate bacterial strains that may be useful biological control agents against *B. glumae* and *R. solani*, rice flag leaves from several rice cultivars were collected at the rice heading stage and were used in leaf prints on potato dextrose agar. Several endophytic bacterial strains isolated from the leaf prints were explored for their potential use as biological control agents of the both pathogens. Out of 127 strains screened *in vitro* for their antagonistic activity against *B. glumae* and *R. solani*, 29 showed promising activities. These strains were retested to confirm antibacterial and antifungal activity, *in vitro*. Several strains showed differential antagonistic activity against *B. glumae* and *R. solani*. Seven strains showing prominent antagonistic activity against *B. glumae* and *R. solani* were identified as *Bacillus* spp. by DNA homology of the 16S rRNA sequence. Sequencing of the 16S rRNA is currently underway to identify the remaining strains. Strains showing antibacterial and antifungal activity *in vitro* are also being tested for their ability to reduce BPB and SB symptoms in the greenhouse.

Development of Single Nucleotide Polymorphism (SNP) markers for *Phytophthora colocasiae*

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Phytopathology 102:S2.9

Taro (*Colocasia esculenta*) is a major staple food in many areas in the world. Taro Leaf Blight (TLB), caused by *Phytophthora colocasiae*, is one of the most important diseases limiting production. Our goal is the development of novel markers to characterize natural populations of *P. colocasiae*. During 2010 and 2011, over 400 isolates were collected in Hawaii, Vietnam, and Hainan Island in Southern China. A focused next generation sequencing approach known as Restriction-Site Associated DNA (RAD) was used to identify potential Single Nucleotide Polymorphism (SNP) markers in two isolates from Vietnam. Almost 9M reads were produced and aligned to the *P. capsici* genome (a close relative) revealing >1000 potential SNP sites. A subset of the putative SNP markers are currently being applied to *P. colocasiae* populations. Results of the marker development and our efforts to determine population structure will be discussed.

Variation in resistance of *Pinus taeda* and *P. elliotii* families against *Leptographium* root infecting fungi

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Phytopathology 102:S2.9

Pine decline is a serious threat to forest production sustainability in the southern United States. Complex interactions of biotic and abiotic factors are involved in this decline. *Leptographium* and *Grossmannia* species are the major contributing factors associated with southern pine decline. This study was conducted to determine the variability in resistance of *Pinus taeda* (loblolly pine) and *P. elliotii* (slash pine) families against *Leptographium* and *Grossmannia* species. Bare root seedlings from 23 *P. taeda* and 5 *P. elliotii* families were screened for resistance using an artificial inoculation method. Seedling responses such as seedling survival, lesion presence, lesion length and occlusion of vascular tissues were measured twelve weeks after inoculations. Stems exhibited dark brown lesions and resinous occluded tissues. Within each family, different *Leptographium* and *Grossmannia* species showed similar response without significant differences from each other and no family $\times$ fungal treatment interaction. Therefore, fungal treatments were pooled and families were categorized according to their level of resistance. Families L-5, L-20, L-8, and, L-13 developed consistently smaller lesions, while families L-1, L-2, L-3 and L-4 were found to have larger lesions overall for each fungus. Slash pine families were found to be more resistant than the loblolly pine families. This study shows that family differences exist and that specific families may perform better in high risk sites.

Molecular and biochemical characterization of resistance to *Botrytis cinerea* in *Solanum lycopersicoides*

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The necrotrophic fungal pathogen, *Botrytis cinerea*, causes grey mold on a wide range of hosts, and is a major economic concern for tomato (*Solanum*

lycopersicum; *Sl*) production. Grey mold can result in lesions on the stems, leaves, and flowers of infected tomato plants, as well as fruit rot both pre and post-harvest. Although all known cultivars of tomato are susceptible to grey mold, *Solanum lycopersicoides* (*Slo*), a wild relative of tomato, is extremely resistant. The objectives of this study are to characterize the molecular and biochemical mechanisms of resistance of *Slo* to grey mold, as well as to gain new insight into host responses to necrotrophic pathogens. To this end, next generation sequencing (454) was used to generate transcriptomics data from *Slo* prior to and 24 and 48 hours after inoculation with *B. cinerea*. Bioinformatics analysis of the transcriptomics data revealed numerous genes in *Slo* that are upregulated in response to infection by *B. cinerea*. Additionally, unbiased metabolic profiling techniques utilizing mass spectrometry were developed to identify compounds differentially expressed in *Sl* and *Slo* before and during infection. Finally, confocal microscopy was used to observe infection of *Sl* and *Slo* plants inoculated with a green fluorescent protein expressing-strain of *B. cinerea*. By combining transcriptomics, metabolic profiling, and histology, we will be able to substantially refine the current understanding of defense against *B. cinerea*. Ultimately, this research will identify candidate genes and pathways for utilization in transgenic approaches to increase resistance to necrotrophic pathogens.

***Aspergillus flavus* genetic diversity of corn fields treated with non-toxicogenic strain Aflaguard® in the southern U.S.**

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Phytopathology 102:S2.9

Aspergillus flavus genetic diversity of corn fields treated with the non-toxicogenic strain Aflaguard® was determined for 14 locations within 6 states in the southern U.S. Preliminary Elisa tests determined low levels of toxigenic strains. Sequencing of selected strains within each location will determine if isolates possess the aflatoxin gene cluster, and the type of mutations/deletions that they possess.

Comparison of methods for isolation and quantification of *Rhizoctonia* spp. from field soil

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Phytopathology 102:S2.9

Isolation and quantification of *Rhizoctonia* spp. has been performed using diverse methods, including direct plating, baiting, direct observations, and molecular techniques. In spring 2009, large scale spatial sampling of fields in Arkansas having rice and soybean rotations was initiated using a multiple-pellet soil-sampler (MPSS), toothpick baiting (TB), elutriation of organic matter from soil, and plant sampling. First year sampling indicated the most sensitive method was TB combined with plant sampling at soybean growth stage V3. However, the quantification of soil populations using TB is problematic. Based on the literature, five toothpicks are reported to displace 0.375 cm³ therefore assaying 0.375 g soil at bulk density (BD) of 1.00 g/cm³. The authors felt this did not take into account the saprophytic growth of the fungus. In experiments using artificially infested soil assays with dilutions of 1.00, 0.500, 0.250, and 0.125 propagules (p) per gram *Rhizoctonia solani* AG4 with the MPSS and TB methods, it was determined that TB was assaying a greater volume of soil than the MPSS. After equilibrating the number of isolates recovered with the known quantity of soil assayed by the MPSS at BD = 1.30 g/cm³ toothpicks assayed a volume of soil greater than its displacement with mean soil volume of approximately 15.74 cm³ and radius of approximately 4.50 mm with no significant differences calculated among volume or radii at any of the four soil dilutions. *In situ* field comparisons of TB versus the MPSS indicated a greater level of detection by MPSS where mean recovery was 18.98 p/kg versus 4.57 p/kg by TB. However, large scale sampling by TB in these fields indicated the majority of *R. solani* AG1-IA, *R. solani* AG11, and *R. oryzae* activity in the top 2.0 cm of the soil profile likely explaining the discrepancy between artificially and naturally infested soils and highlighting the limitations of each technique.

Degradation of wheat straws under different soil moisture and temperature conditions and their ability to support perithecia development by *Fusarium graminearum*

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Phytopathology 102:S2.9

Wheat scab, also known as *Fusarium* head blight (FHB) is a destructive disease of wheat worldwide. Scab is caused by a group of *Fusarium* species among which *F. graminearum* is the most common. This disease presents a

major challenge to wheat production throughout most of the U.S.A., relating to multifold effects on yield and quality losses. Although scab has not been reported as major problem of wheat in Alabama, it has been observed in wheat fields in northern parts of this state in recent years. However, the disease has not been found in the southern parts of the state. Plant residues of some crops such as wheat, barley, corn, and soybean are the major sources of inoculum of *Fusarium* species. Development and survival of the fungus is affected by the degradation of these residues. Greater residue degradation will decrease the development of the fungus, and high temperature coupled with high moisture accelerates residue decomposition in the field. We hypothesize that due to higher annual rainfall and temperatures, residue degradation is greater and fungal survival lower in southern fields than northern fields. To validate our hypothesis, we have been conducting laboratory experiments of degradation of wheat straws in native soils maintained at different levels of temperature and moisture. After incubation, straws are disinfected then inoculated with *F. graminearum* and the numbers of perithecia that develop are counted. Our preliminary study with soil at room temperature yielded a lower number of perithecia on straws incubated with higher than with lower soil moisture. Future studies will evaluate the development of perithecia on straws incubated at different temperatures and soil moisture levels.

Use of hot-water baths as non-chemical treatments for Rhizopus soft rot on sweetpotatoes

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Phytopathology 102:S2.10

Soft rot caused by *Rhizopus stolonifer* is the most destructive postharvest disease of sweetpotato (*Ipomoea batatas*). *R. stolonifer*, a zygomycete fungus, is ubiquitous in the soil and air and infects sweetpotato storage roots through wounds, causing total breakdown in several days. *Rhizopus* soft rot can be controlled by minimizing injury to the root, along with a prophylactic application of fungicides. As the demand for organic produce has increased, there is a need for non-chemical control of *Rhizopus* soft rot. Hot-water baths were explored to control *Rhizopus* soft rot on two cultivars of sweetpotato, Evangeline and Beauregard. For each treatment, 20 roots of each cultivar were bruised with an elastic band driven dowel and brushed with 1×10^6 sporangiospores per ml inoculum. The sweetpotato roots were then submerged in 48, 50, 52, and 54°C hot-water baths for 2 to 30 minutes. After 7 days of storage at 21°C, the number of rotten sweetpotatoes was determined. Treating the sweetpotatoes with the hot-water baths decreased the incidence of *Rhizopus* soft rot from 74% rotten in the inoculated control to 41% rotten in the water baths ($P < 0.01$). Among the overall temperature treatments, soft rot reduction was only significant in the 50°C water bath (26% rotten, $P = 0.061$). However, the 10 and 15 min duration 52°C hot-water baths had the lowest soft rot incidence of any time and temperature combination: 28% rotten ($P = 0.023$) and 16% rotten ($P = 0.042$), respectively. Even though the hot-water baths did not provide the same level of control as fungicides, they may be useful as part of an integrated program to manage *Rhizopus* soft rot without relying on prophylactic use of synthetic fungicides.

Effect of initial fungicide application timing and cultivar on peanut pod rot on the High Plains of Texas

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Phytopathology 102:S2.10

The pod rot complex, caused by *Rhizoctonia solani* and several *Pythium* spp., is one of the most damaging and costly diseases to manage in Texas peanut. Field trials were conducted during the 2010 and 2011 growing seasons to compare fungicide regimes where initial fungicide applications were delayed. In 2010, delaying initial applications increased pod rot by 3.6 and 2.0% for mefenoxam and azoxystrobin, respectively. Similarly, the percentage of kernels exhibiting pod rot symptoms was greatest where initial fungicide applications were delayed. Applications of azoxystrobin and mefenoxam initiated approximately 70 days after planting (DAP) increased yields by 203 and 297 kg/ha compared to the non-treated control, whereas yields from plots where initial applications of either fungicide were delayed until 84 DAP were similar. Additional studies were conducted to evaluate the performance of commercially available cultivars in fields with a history of pod rot. Pod rot incidence was lowest ($<1\%$) for the cultivars Tamrun OL07 and ACI 149 compared to the commercial standard Flavorrunner 458 (4.5%). Yields were greatest for Tamrun OL07 (3127 kg/ha); whereas, Flavorrunner 458 and ACI 149 yielded 2256 and 2457 kg/ha, respectively. Grade parameters for Tamrun OL07 are generally 1 to 2% lower than Flavorrunner 458. While this attribute affects crop value, increased hull thickness may limit infections from occurring. Both fungicide timing and cultivar selection can impact pod rot development. Additional research evaluating the interaction between these factors is needed so that producers can maximize profitability.

Diagnosis of centipedegrass anthracnose caused by Colletotrichum eremochloae sp. nov.

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Phytopathology 102:S2.10

Centipedegrass (*Eremochloa ophiuroides*) is a medium-textured, stoloniferous warm-season turfgrass used as home lawn and utility turf. It is well-adapted to a wide range of soil conditions but grows best in sandy, acidic soils of low fertility and can withstand some shade. Diseases arise when centipedegrass is over-managed with fertility and irrigation inputs and low height of cut. In spring transition, centipedegrass home lawns exhibit symptoms of leaf chlorosis and sheath blight, and irregular, chlorotic patches associated with thinning turf. The patches range in size from a few meters isolated in the turf to the entire sward. Microscopic observations in affected leaf sheaths revealed a fungus with light gray mycelium, prominent septation, and abundant hyphal appressoria with acervuli lacking. Cultures on potato dextrose agar were grey in appearance, produced few conidia, and developed a yellow exudate at the perimeter of the colony, similar to *Colletotrichum sublineola* the causal agent of sorghum (*Sorghum vulgare* Pers.) and johnsongrass (*S. halepense* (L.) Pers.) anthracnose. Sequence markers, *Apn2*, *Mat1*, *Sod2*, and ITS was 98% similar to *C. sublineola* but was an identical match to an unidentified fungus present in a centipedegrass herbarium sample that was intercepted from China in 1923, resulting in a new species *Colletotrichum eremochloae*.

Effect of Acidovorax citrulli localization in watermelon seeds on pathogen detection and seed-to-seedling transmission of bacterial fruit blotch

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Phytopathology 102:S2.10

Bacterial fruit blotch, caused by the gram negative bacterium *Acidovorax citrulli* is a serious threat to cucurbit production worldwide. The pathogen is seedborne and seed transmitted in many cucurbit species and under typical transplant house and field conditions, the disease can spread rapidly and cause significant economic losses. *A. citrulli*-infested seeds represent the most important source of primary inoculum and there is no BFB resistance in commercial cucurbit cultivars. Hence, seed health testing and seed treatments are widely employed to exclude the pathogen. Despite this, 2011 BFB outbreaks were reported in many states including in Georgia, Virginia, Missouri and Kentucky. This suggests that more detailed information is needed about the host pathogen interactions that lead to seed infection and seedling transmission to improve BFB management. Using watermelon as a host, we observed that the localization of *A. citrulli* cells in seeds varied depending on the method of ovary inoculation. Inoculation of the ovary pericarp resulted in superficial contamination of seeds while pistil inoculated resulted in contamination of the seed embryo. Both inoculation techniques resulted in seedlots that were 100% infested. However, while 81% of the pericarp inoculated seedlots were detected by a diagnostic assay that relied on washing seeds; only 13% of the pistil-infested seedlots were positive. The location of *A. citrulli* in seeds also affected the rate of initial BFB seedling transmission and our data suggest that type II secreted enzymes may contribute to the early stages of seed-to-seedling transmission of BFB.

Effect of citrus juice on viability of 'Candidatus Liberibacter asiaticus' in liquid suspension

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Citrus Greening Disease, or Huanglongbing (HLB), has recently become a threat to citrus crops in the United States. The bacterium '*Candidatus Liberibacter asiaticus*' (LAS) is the suspected causal agent for HLB in the U.S., and has not yet been successfully cultured. Without the ability to culture the bacterium, it is difficult to develop a treatment for infected trees. Experiments are being conducted to determine the influence of different mixtures of commercial grapefruit juice and pummelo juice from LAS-infected pummelos, and synthetic media on the viability of LAS over time. Pummelo fruits (*Citrus grandis* 'Mato Buntan') infected with LAS were collected in Florida, their seeds were pulverized to create an inoculum, and their pulp was used to prepare pummelo juice for use as a media component. Pummelo juice and commercially available grapefruit juice were filter-sterilized, mixed in different ratios with synthetic media, and inoculated with the seed-based

inoculum. The viability of LAS over time was determined using ethidium monoazide (EMA) treatment in conjunction with quantitative PCR (qPCR) to compare the number of viable cells to the number of nonviable cells at different time points. EMA is a photoactive stain that binds to DNA from nonviable cells, rendering bound DNA undetectable by qPCR and making it possible to quantify DNA from viable and nonviable cells. Viability of LAS in each different treatment over time was determined by comparing qPCR data from EMA-treated and non-EMA-treated samples. Results indicate that treatments containing juice allow LAS cells to remain viable for several days longer than the synthetic media control. There is also an indication that higher concentrations of juice in the media increase the viability of LAS. While LAS has still not been cultured, this research allows insight into conditions conducive for LAS viability, which is important in the search for a culture method.

Efficacy of the fungicide fluopyram and cultivar performance for management of Sclerotinia blight of peanut in Texas

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Sclerotinia blight, caused by *Sclerotinia minor* Jagger, is an economically important disease of peanut. Field studies were conducted in 2010 and 2011 to evaluate 1) the efficacy of the fungicide fluopyram and 2) the performance of peanut cultivars in fields with a history of the disease. Initial applications of fluopyram were made 70–85 days after planting with sequential applications made on a 30 day interval. Disease incidence in plots treated with fluopyram was lower (10.4%) than the non-treated control (44.8%), and did not differ from plots treated with fluzinam (13.2%). On average, fluopyram increased yields by 874 kg/ha over the non-treated control. Both susceptible (Flavorrunner 458 and Tamrun OL02) and partially resistant (Tamrun OL07, Tamrun 11, and Red River Runner) responded to applications of fluopyram. When averaged across fungicide treatments, the partially resistant cultivars exhibited 60% less disease than the susceptible cultivars. Yields for Tamrun11, Red River Runner and Tamrun OL07 ranged from 3438 to 4313 kg/ha and 1896 and 2141 kg/ha for Tamrun OL02 and Flavorrunner 458, respectively. The registration of fluopyram provides producers with an additional tool to manage Sclerotinia blight in peanut. This in conjunction with the development and release of partially resistant cultivars will help sustain production in fields infested with *S. minor*.

Host plant, aphid species and virus infection status influence on acquisition and transmission of Sweet potato feathery mottle virus

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Sweetpotato, *Ipomoea batatas*, production is affected by Sweetpotato feathery mottle virus (SPFMV), a non-persistent stylet-borne virus known to also infect wild morning glory plants. It is vectored by various aphid species, among them the green peach aphid, *Myzus persicae* and the cotton aphid, *Aphis gossypii*. SPFMV frequently occurs in co-infections with other viruses, for example, Sweet potato virus G (SPVG) and Sweet potato virus 2 (SPV2) in Louisiana. Our objective was to determine the effect of host plant, aphid species and infection status on transmission of SPFMV. Sweetpotato cultivars Beauregard and Evangeline and two morning glory species *Ipomoea hederacea* and *I. cordatotriloba* were infected with SPFMV in single and mixed status. Two aphid species *A. gossypii* and *M. persicae* were used to transmit SPFMV from source leaves to test plants of *Ipomoea nil* cv. Scarlet O'Hara by single probes. SPFMV titers in source leaves were quantified using quantitative reverse transcription PCR (qRT-PCR). SPFMV transmission from *I. hederacea* with a natural mixed infection by *A. gossypii* (39%) was significantly greater than in other host-virus combinations. Successful transmissions by *A. gossypii* were greater compared to *M. persicae* in all host-virus combinations. Virus titers in source leaves were significantly greater in *I. hederacea*, and *I. cordatotriloba* singly infected compared to other host-virus combinations. There was a significant positive correlation between virus titer and transmission by both aphid species. These results suggest that under controlled conditions, SPFMV is more readily transmitted from infected morning glory plants compared to sweetpotato. Additionally, mixed infected plants are better virus sources for aphids than singly infected, and *A. gossypii* is a more efficient vector than *M. persicae* under laboratory conditions.

Epidemiology of soybean rust on kudzu and soybean

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Phakopsora pachyrhizi, the causal agent of soybean rust (SBR), is a potentially devastating disease of soybean in the U.S. Each year, during the winter months, the pathogen is limited to the southeastern states on the alternative host kudzu. This source is critical to providing the initial inoculum for the epidemic the following year. To better understand the SBR epidemic in north Florida, which can potentially produce inoculum for other soybean producing areas of the country, a theoretical disease model was developed. This model utilized estimates of disease development and spore escape (Pivonia and Yang, 2006; Andrade et al., 2009) coupled with data from detached leaf assays assessing sporulation to compare SBR development on kudzu and soybean. From experimental data, SBR sporulation per lesion is the main difference of disease development between kudzu and soybean. Based on our results 30 days after disease onset, the number of infections from 1 hectare of kudzu is only one tenth of infections from 1 hectare of soybean. Similarly, the number of spores escaping into the atmosphere each day from 1 hectare of kudzu is 250,000 less than that from soybean. While kudzu is an important inoculum source for SBR, it most likely only has a substantial impact on initial soybean infections. It is these infections on soybean that will propel the epidemic in north Florida and throughout the U.S. These results and their implications will be presented and discussed.

Thinning affects on incidence of ophiostomatoid fungal species in decline-impacted *Pinus taeda* stands

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Ophiostomatoid fungi such as *Grossmannia* spp. and *Leptographium* spp. are known as contributing factors to southern pine decline in the southeastern U.S. This study was developed to quantify fluctuations in ophiostomatoid fungal incidence as a response to traditional thinning in decline-impacted *P. taeda* stands in central Alabama and Georgia. Six forest health monitoring research plots were established on five industrial *P. taeda* timberlands. Plots were divided evenly into thinning and control treatments. Roots of *P. taeda* were excavated and assayed for ophiostomatoid fungal infection from both pre- and post-treatments. The dominant isolated fungal species was *Leptographium procerum* compared to other ophiostomatoid fungi including *L. terebrantis*, *L. alarcris*, *G. huntii* and *Ophiostoma ips*. Statistical analysis indicated that incidence of ophiostomatoid fungal species had a positive correlation with thinning treatments.

An emerging problem for *Celtis* spp. - a novel ampelovirus associated with mottling symptoms

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The genus Ampelovirus, family Closteroviridae, includes viruses that cause significant diseases of several fruit and ornamental crops. Here we report a new ampelovirus closely associated with virus-like symptoms on sugarberry (*Celtis laevigata*), a common landscape tree. Distinct ringspots and yellow mottling patterns have been observed on sugarberry trees in the southern part of the U.S. Such symptoms first present in late spring and become more evident as the season progresses. The novel virus has a monopartite genome which consists of single-stranded RNA of approximately 17kb. Protein pairwise comparisons and phylogenetic analysis revealed its close relationship with Grapevine leafroll-associated virus-3, the type member of the genus Ampelovirus, family Closteroviridae. The amino acid identities between the two viruses range from 26% for the coat protein to 53% for the RNA dependent RNA polymerase homolog, indicating that the new sugarberry virus is a novel member of the genus. Using newly-developed detection protocol, the presence of the virus was confirmed not only in symptomatic sugarberry, but also in multiple hackberry trees purchased from an East Coast mail-in nursery. A great number of mealybugs, recognized vectors of ampeloviruses, are often observed feeding on affected trees, and transmission studies are underway to determine whether they are the potential vector of the new virus.