



2002 APS Annual Meeting Abstracts of Presentations

Abstracts submitted for presentation at the APS 2002 Annual Meeting in Milwaukee, Wisconsin, July 27–31, 2002. The abstracts are arranged alphabetically, by first author's name.

Advances in the integration of morphological and molecular characterization in *Phytophthora* benus: The case of *P. kelmania* and other putative new species. Z. G. ABAD (1,3), J. A. Abad (3), and T. Creswell (2,3). (1) Plant Pathogen ID. Lab.; (2) Plant Disease and Insect Clinic; (3) Dept. Plant Pathology, North Carolina State Univ. Phytopathology 92:S1. Publication no. P-2002-0001-AMA.

Phytophthora with over 70 spp is one of the most important Genera of plant pathogens. Considerable advances in the taxonomy of this stramenopile using morphological and molecular characterization have been accomplished. Yet, correct identification to species levels and characterization of new taxa is still a major challenge. Using our innovative morphological/molecular pictorial key new hosts and 3 new species have been determined including: *P. glovera*, *P. bisheria* (semipapillated), and *P. kelmania* (non papillated, named to honor Dr. A. Kelman). Additional unnamed new species are presented, including: Ph2471, Ph3552, (papillated), Ph9-001, Ph2529, Ph5212 (non papillated). An updated phylogenetic tree (ITS1-2 rDNA) including the recently described *P. europaea*, *P. pistaciae*, *P. psychrophila*, *P. ramorum*, and *P. uliginosa* maintains the consistency of 2 main groups (papillated and non-papillated) and verifies the identity of our eight putative new *Phytophthora* species.

Predominance and pathogenicity of fungi and stramenopiles associated with Black Root Rot of strawberries. Z. G. Abad (1,2), F. J. LOUWS (2), G. E. Fernandez (3), and L. M. Ferguson (2). North Carolina State Univ., Raleigh, NC 27695, (1) Plant Pathogen ID. Lab.; (2) Dept. of Plant Pathology; (3) Dept. of Horticulture. Phytopathology 92:S1. Publication no. P-2002-0002-AMA.

BRR is an important disease of strawberry around the world. From 1998 to 2001 about 1300 isolates (I) were collected in 6 localities (L) in NC. Sixty taxa were identified. *Rhizoctonia fragariae* AG-A (119-I, 4-L), AG-G (55-I, 3-L), AG-I (24-I, 4-L), *Pythium irregulare* (285-I, 5-L), *Pythium* Group F (14-I, 3-L), *Pythium* HS (10-I, 3-L), *Phytophthora cactorum* (74-I, 2-L), *Fusarium solani* (47-I, 5-L), *F. oxysporum* (45-I, 5-L), *F. acuminatum* (21-I, 5-L), *Phoma* sp. (12-I, 5-L) were the most common. Other known pathogens included: *Ascochyta fragariae* (1-I, 1-L), *Macrophomina phaseolina* (5-I, 3-L), *Phomopsis obscurans* (1-I, 1-L). *Rhizoctonia binucleate* AGN (3-I, 1-L) and *Thielaviopsis basicola* (1-I,1-L) are new reports on strawberry. Pathogenicity tests indicate the importance of *R. fragariae* AGA, G, I, *P. irregulare*, *Pythium* HS, *P. cactorum*, *Phoma* sp., *F. oxysporum* and *F. solani*. The diversity of Fungi and Stramenopiles in BRR disease highlights the importance of this complex in the strawberry production areas of NC.

Cultural and analytical methods to determine aflatoxin production by *Aspergillus* species from the Mississippi Delta. H. K. ABBAS (1), R. M. Zablutowicz (2), B. W. Horn (3), W. Xie (4), and W. T. Shier (5). (1) USDA-ARS, CG & PRU, Stoneville, MS; (2) USDA-ARS, SWSRU, Stoneville, MS; (3) USDA-ARS, NPRL, Dawson, GA; (4) University of Minnesota, Minneapolis, MN; and (5) University of Minnesota, Mineapolis, MN. Phytopathology 92:S1. Publication no. P-2002-0003-AMA.

Several methods were used to detect aflatoxin production by various *Aspergillus* species. Isolates (n = 786) were obtained from crops (including corn, peanut, cotton) and soils from the Mississippi Delta. Cultural methods for assessing aflatoxin production on potato dextrose agar included yellow pigment (YP) production and color change with exposure to ammonia vapor (AV). The presence of aflatoxins in culture extract was confirmed by TLC and LC-MS and quantified by ELISA and HPLC. Fifty-one percent of isolates produced YP, and all were positive for the presence of aflatoxins. Forty-seven percent of isolates changed color with AV, and all were positive for aflatoxins. Thus, positive YP and AV responses were highly correlated with aflatoxin production. However, both methods failed to detect aflatoxins in all cultures that tested positive using ELISA, HPLC and TLC. Cultures that produced less than 250 ppb were typically negative by the AV method. *A. flavus* isolates positive for aflatoxins produced AFB1 and AFB2 only, whereas *A. nomius* and *A. parasiticus* produced all four aflatoxins (AFB1, AFB2, AFG1, and AFG2). Fifty-one percent of 341 soil isolates and 55% of 250 corn isolates produced aflatoxins. This study showed that YP and AV methods can be used for rapid screening but are not reliable in detecting low producers of aflatoxins.

Effect of foliar applications of neem oil and fish emulsion on bacterial spot disease and yield of tomatoes and peppers. P. A. ABBASI, D. A. Cuppels, and G. Lazarovits. Agriculture and Agri-Food, London, Ontario. Phytopathology 92:S1. Publication no. P-2002-0004-AMA.

Field trials were conducted in London, Ontario during 2000 and 2001 to test the efficacy of foliar sprays of neem oil and fish emulsion, derived from the neem seed and Menhaden fish, respectively, against bacterial spot disease (*Xanthomonas vesicatoria* or *X. axonopodis* pv. *vesicatoria*) and yield of tomatoes and peppers. Ten weekly sprays of neem oil and fish emulsion (0.5%; v/v) significantly reduced disease severity on the foliage of inoculated field-grown tomato and pepper plants in both years. The disease incidence on fruit of these plants was reduced but the effect was not always significant. However, the number of lesions on pepper fruit was significantly less with these treatments. There was a marked increase in the yield of disease-free fruit on neem oil-treated pepper plants in 2000 and fish emulsion-treated pepper plants in 2001, but none of the treatments significantly increased total tomato or pepper yield. Neem oil and fish emulsion had no observable phytotoxic effects on tomato or pepper foliage. These results suggest a possible use for neem oil or fish emulsion as foliar sprays to manage bacterial spot disease.

***Cylindrocarpum* species and other fungi isolated from bark of beech in the advancing and killing fronts of the beech bark disease epidemic in Michigan.** G. C. Adams. Michigan State University. Phytopathology 92:S1. Publication no. P-2002-0005-AMA.

Beech bark disease is a lethal disease of *Fagus grandifolia* caused by the interaction of the exotic beech scale, *Cryptococcus fagisuga*, and native and exotic species of the fungus *Neonectria*. A Michigan beech bark disease monitoring and impact analysis system was established in 2001 and 2002 consisting of long-term plots across the beech resource of Michigan. Our role was to determine the presence or absence of *Neonectria* in bark, and to identify the species. More than eighteen plots (4 subplots/plot) in 7 counties were sampled, with bark collected from 12 trees/plot. The plots were rated as having no scale, light, or heavy infestations. We developed an agar medium

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selective for isolation of *Cylindrocarpon* anamorphs of *Neonectria* from bark. Identification of *Neonectria* species was based on morphology of *Cylindrocarpon* isolates sporulating on alfalfa stems on water agar. Objectives are to delineate the killing front and to determine whether the relative abundance and distribution of native versus exotic *Neonectria* species affects the rate of disease spread.

Utilization of spiral gradient endpoints to estimate fungicide resistance potential in alternate-row-spraying of tree fruit crops. J. E. ADASKAVEG and H. Förster. Dept. Plant Pathology, Univ. of California, Riverside, CA 92521. Phytopathology 92:S2. Publication no. P-2002-0006-AMA.

Alternate-row-spraying for control of blossom diseases of deciduous tree fruit crops is a cost-saving practice used by growers in California. Criticisms of this method include increased potential for resistance to develop in pathogen populations. In field trials on sweet cherry, trees at full bloom were treated from one side with cyprodinil using an airblast sprayer at a rate of 238 g/850 L/ha. After drying, blossoms were collected for inoculation with *Monilinia fructicola* and for residue analysis from the sprayer-exposed sides and the opposite sides of the trees. Disease incidence on inoculated anthers was 61% for the non-treated, 6% for the exposed-side, and 26% for the opposite-side blossoms, corresponding to residues of 0, 16.4, and 2 mg/L cyprodinil, respectively. In spiral gradient endpoint assays, confluent growth of *M. fructicola* was affected at 0.06 mg/L. Outlier colonies developed up to 4 mg/L, well above the 2-mg/L residues of the opposite-side blossoms. This indicated that alternate-row-spraying with the single-site mode-of-action fungicide cyprodinil may allow resistance to develop in target pathogens.

Molecular mapping of *Septoria tritici* leaf blotch resistance in wheat. T. ADHIKARI, J. M. Anderson, and S. B. Goodwin. USDA-ARS, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S2. Publication no. P-2002-0007-AMA.

Septoria tritici leaf blotch (STB), caused by the ascomycete fungus *Mycosphaerella graminicola* (anamorph *Septoria tritici*), is one of the most destructive diseases of wheat worldwide. Host plant resistance is the preferred method of disease control. The identification and mapping of STB resistance genes and the development of resistant wheat cultivars can be facilitated through the use of molecular markers. A total of 126 F₁₀ recombinant inbred lines from a cross between the highly resistant synthetic hexaploid wheat, W7984, and the highly susceptible cultivar, Opata 85, was evaluated for STB reaction and molecular markers. Genetic analysis indicated that a single dominant gene controls the resistance to STB. Bulk segregant analysis based on amplified fragment length polymorphism (AFLP) was used to develop a genetic linkage map. An AFLP marker, *EcoRI*-ACG/*MseI*-CAG5, was linked in repulsion with the STB resistance gene at a distance of approximately 5.1 cM. Chromosome 7B is the putative location of this resistance gene based on its association with previously mapped microsatellite markers. Efforts to find additional linked markers are continuing.

A new experimental host for plant viruses. S. ADKINS and E. N. Roskopf. USDA-ARS-USHRL, Fort Pierce, FL 34945. Phytopathology 92:S2. Publication no. P-2002-0008-AMA.

Key West nightshade (*Solanum bahamense* L.) is a perennial solanaceous plant found in the extreme southern portion of Florida, primarily the Keys. It can be propagated by seed and cuttings and is absent from the noxious weed lists of all US states. Its susceptibility to four viruses common to Florida was evaluated by mechanical inoculation of leaves with *Tobacco mosaic virus* (TMV), *Pepper mild mottle virus* (PMMoV), *Cucumber mosaic virus* (CMV) and *Tomato spotted wilt virus* (TSWV). No symptoms were observed following inoculation with TMV or PMMoV. CMV induced necrotic local lesions on inoculated leaves. TSWV induced chlorotic rings on inoculated leaves and mosaic and malformation of uninoculated leaves. TMV, PMMoV and TSWV were shown to systemically infect *S. bahamense* through the use of indicator hosts, viral-associated double-stranded RNA analysis, RT-PCR and/or ELISA. Active growth of infected plants continued for several months after inoculation making *S. bahamense* suitable for long-term maintenance of these viruses *in planta*. We suggest that *S. bahamense* may be useful for culture collections and studies involving large numbers of virus isolates where fresh tissue is continuously required.

Analysis of candidate genes at the *Vf* apple scab resistant locus. M. AFUNIAN (1,2), P. Goodwin (1), and D. Hunter (1). (1) University of Guelph, Guelph ON Canada; (2) Plant Pests and Diseases Res. Ins. Tehran, Iran. Phytopathology 92:S2. Publication no. P-2002-0009-AMA.

Apple scab, caused by the fungus *Venturia inaequalis* is the most important disease of apple (*Malus × domestica*) worldwide. Genetic resistance to apple scab exists in several East Asiatic species including *Malus floribunda*. *Malus floribunda* 821 is the most important source of monogenic scab resistance (*Vf* gene) that was bred into nearly all commercial apple scab resistant cultivars. It was recently reported that a cluster of receptor-like genes present at the (*Vf* locus, are homologs to the (*Cf* gene of tomato. However, it was not determined which member of the cluster, either *HcrVf2* or *HcrVf4* was the *Vf* gene. Specific primers were designed for protein domain C of the *HcrVf1*, *HcrVf2* and *HcrVf4*. We have found that these primers amplified 2 bands in all *M. floribunda* and *M. × domestica* varieties tested. However, another PCR product was only present in 9 out of 10 *M. floribunda* examined. It was also present in apple scab resistant *M. × domestica* cultivars carrying the *Vf* gene but absent from scab susceptible cultivars. This indicates that the occurrence of fragment, matching *HcrVf2*, correlates closely with presence of the *Vf* gene.

Genomic fingerprinting of *Rathayibacter* species by amplified fragment length polymorphism. I. V. AGARKOVA (1), A. K. Vidaver (1), E. Postnikova (2), and N. W. Schaad (2). (1) University of Nebraska, Department of Plant Pathology, Lincoln, NE; (2) USDA ARS Foreign Disease-Weed Science Research Unit, Fort Detrick, MD 21702. Phytopathology 92:S2. Publication no. P-2002-0010-AMA.

The genus *Rathayibacter* (*Clavibacter*) includes four species of Gram-positive coryneform bacteria pathogenic to *Gramineae*: *R. tritici* and *R. iranicus*, causal agents of gumming disease of wheat; *R. rathayi*, causal agent of gumming of orchard grass; and *R. toxicus*, causal agent of ryegrass toxicity. To assess the genetic diversity within each species, AFLP's using 6 strains of *R. tritici*, 4 of *R. toxicus*, and 18 of *R. rathayi* were determined. Since only one strain of *R. iranicus* exists, it was not included. Genomic DNA was isolated by a modified Marmur method and digested with *ECORI/MSEI* restriction endonucleases and the corresponding adaptors were ligated. Selective amplification was performed with *MSEI*-C and *ECORI*+0 primers. Each strain of the three species resulted in unique fingerprints. These preliminary studies show that the AFLP analysis has a high level of discriminatory capability and should be useful for fingerprinting strains of *Rathayibacter* species.

Effect of fungicides for black pod control on soil microflora in Papua New Guinea. P. H. Aisa (1), G. Blaha (1), P. K. Hebbar (2), S. Sanogo (3), and J. H. BOWERS (4). (1) Papua New Guinea Cocoa and Coconut Research Inst., Rabaul; (2) M & M Mars, Inc.; (3) Wye Research & Education Center, Univ. of Maryland; (4) USDA, ARS, Alternate Crops & Systems Lab., Beltsville, MD. Phytopathology 92:S2. Publication no. P-2002-0011-AMA.

Populations of five groups of microorganisms were enumerated from cacao pod-baited soil and directly from soil collected from cacao plantations managed with and without fungicides for black pod control in Papua New Guinea. There were significant (P = 0.05) site × fungicide interaction effects for some microbial groups. Generally, use of fungicides reduced soil populations of total fungi, *Trichoderma* spp., and actinomycetes in both sampling schemes. However, populations of total bacteria and *Burkholderia* spp. were higher in cacao soil managed with fungicides. A high percentage of a random sample of bacterial strains isolated from soil using an infected pod as bait suppressed symptom development caused by *Phytophthora megakarya* in a leaf disk assay. Fungicide treatment of cacao plantations lowered population numbers of the main microbial groups involved in saprophytic decomposition of infected pods, but may select for bacteria important in biological control.

***Fusarium* spp. causing wheat head blight and crown rot in Australia.** O. A. Akinsanmi (1), V. Mitter (1), S. Simpfendorfer (2), D. Backhouse (3), and S. CHAKRABORTY (1). (1) CSIRO Plant Industry, CRC Trop. Pl. Prot., Univ. of Queensland 4072; (2) NSW Agriculture, Tamworth; (3) Univ. of New England, Armidale, Australia. Phytopathology 92:S2. Publication no. P-2002-0012-AMA.

In Australia head blight (FHB) incidence has recently increased with above average rainfall while crown rot (CR) continues as a chronic problem of wheat. In 2001 39 wheat fields were surveyed for CR and FHB and pure cultures obtained from samples were diagnosed using PCR assays specific to *F. pseudograminearum*, *F. graminearum*, *F. poae*, *F. avenaceum* and *F. culmorum*. Although FHB is restricted to a small area in New South Wales, *Fusarium* spp. capable of causing FHB were isolated from a wide area. CR was more widespread and often occurred with FHB in the same paddock. *F. pseudograminearum* and *F. graminearum* were the 2 most dominant spp., the former was more common in fields that had wheat in 2000. Isolates of the 2 spp. tested so far have caused both CR and FHB in infection assays, but there

is difference in aggressiveness for each disease among isolates from the same species. Given the close relationships in etiology and epidemiology between the two diseases, management approaches need to deal with the entire wheat farming system.

A new race of *Pyrenophora tritici-repentis* that produces a putative host-selective toxin. S. ALI (1), H. Ling (2), S. Meinhardt (2), and L. Franci (1). (1) Dept. Plant Pathology; (2) Dept. Biochemistry, North Dakota State Univ., Fargo, ND 58105. Phytopathology 92:S3. Publication no. P-2002-0013-AMA.

Six races of *Pyrenophora tritici-repentis*, the cause of tan spot of wheat, have been described on the wheat differentials Katepwa, Glenlea, 6B365, and Salamouni. We found an isolate of *P. tritici-repentis* from Argentina for which a conidial inoculation produced necrosis with extensive chlorosis on ND495, chlorosis on 6B365, necrosis on Katepwa and Glenlea, and a resistant reaction on Salamouni and M-3. Ptr ToxA was present in the culture filtrate. Partially purified culture filtrate contained a putative host-selective toxin that caused extensive chlorosis on ND495, which is insensitive to ToxC. The active molecule was about the same size as Ptr ToxC but had several distinguishing chemical properties. We propose that ND495 be added to the standard differential set, that this differential reaction profile be designated as race 7, and that the new toxin be designated as Ptr ToxD.

Partial nucleotide sequence and molecular detection of a putative new grapevine leafroll associated virus. R. ALKOWNI, A. Rowhani, and D. A. Golino. Department of Plant Pathology, University of California, Davis, CA 95616. Phytopathology 92:S3. Publication no. P-2002-0014-AMA.

Leafroll symptoms similar to those produced by species of *Grapevine leafroll associated virus* (GLRaV) were observed on a grapevine and confirmed when it tested positive by graft-indexing on the leafroll indicator host, *Vitis vinifera* cv. Cabernet Franc. Currently, eight different viruses, all members of *Closteroviridae*, are reported to be associated with grapevine leafroll disease. The newly recognized diseased grapevine had tested negative by RT-PCR and ELISA for the eight GLRaVs. DsRNA was prepared for a RT-PCR using degenerate oligonucleotide primers designed to non-specifically amplify a 550 to 650 nucleotide fragment from HSP70 of closteroviruses. RT-PCR products were gel isolated and sequenced. This sequence was used for comparative analysis and for developing a RT-PCR detection protocol. The sequence indicated that the virus was most similar to GLRaV-5 (79% homology). The presence of closterovirus-like HSP70 sequences implies that the newly described virus is a member of *Closterovirus* genus and the name *Grapevine leafroll associated virus-9* is proposed.

Pathogen motility and bacterial wilt disease. C. ALLEN and J. Tans-Kersten. Dept of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706. Phytopathology 92:S3. Publication no. P-2002-0015-AMA.

Ralstonia solanacearum causes bacterial wilt disease on a wide range of plants. This soil-borne pathogen invades host roots through wounds and secondary root emergence points, rapidly colonizing the xylem elements. *R. solanacearum* cells are highly motile, particularly at low cell densities, but the role of pathogen motility in disease development was not understood. We therefore constructed defined nonmotile mutants. One lacks FliC, the flagellin structural protein; a second lacks FliM, the motor switch protein; and a third lacks FliHDC, a transcriptional activator that regulates the entire flagellar biosynthetic pathway. All three nonmotile mutants were significantly reduced in virulence in a soil-soak inoculation virulence assay that required the bacteria to actively find and enter host roots from the soil. However, these same mutants remained fully virulent when they were introduced directly into the plant vascular system via cut petioles. This differential result indicates that the primary function of motility in *R. solanacearum* virulence is to facilitate the early stages of host plant invasion and colonization. A two-component regulator, PehSR, positively controls expression bacterial motility in response to cell density. We are currently exploring the interaction between *pehSR* and *fliHDC*.

Impact of *Meloidogyne incognita* on soybean varieties in Illinois. J. B. ALLEN, J. P. Bond, M. E. Schmidt, S. A. Walters, and J. S. Russin. Plant, Soil and General Agriculture, Southern Illinois University Mailcode 4415, Carbondale, IL 62901-4415. Phytopathology 92:S3. Publication no. P-2002-0016-AMA.

Meloidogyne incognita is a potential threat to crop production in Illinois. In 2000 and 2001, a survey of vegetable and soybean fields and fruit tree orchards was conducted. The nematode was detected in six soybean fields,

eight vegetable fields and five peach orchards. Fifteen parental germplasm lines were planted into 150 cc conetainers and inoculated with 10,000 eggs. After 50 days, the degree of root galling and necrosis was determined. Four soybean varieties (Pioneer 9481, Pioneer 9492, Gateway 493 and LS 94-3207) were selected and planted in infested fields. Nematode population densities were recorded at planting and every 6 weeks until harvest. At planting, the population density of *M. incognita* averaged 8 juveniles/100 cc soil. Reproduction by *M. incognita* was higher in the plots planted to P 9481. At harvest, the population densities (juveniles/100 cc soil) were 508, 41, 37 and 6 for P 9481, P 9492, GW 493 and LS 94-3207, respectively. Across the varieties, there was a linear decrease in soybean yield as the population density of *M. incognita* increased.

RAPD-PCR and rep-PCR molecular markers in *Uromyces appendiculatus* associated with *Ur-6* avirulent and virulent phenotypes in common bean. A. T. ALLEYNE (1), J. R. Steadman (1), J. G. Fenton (1) and K. M. Eskridge (2). (1) Department of Plant Pathology, University of Nebraska-Lincoln, Lincoln, NE 68583-0722; (2) Department of Biometry, University of Nebraska-Lincoln, NE 68583-0724. Phytopathology 92:S3. Publication no. P-2002-0017-AMA.

Twenty races comprising both virulent and avirulent isolates of *Uromyces appendiculatus*, on the pinto bean cultivar Olathe, which contains the *Ur-6* rust resistance gene, were screened using rep-PCR and RAPD-PCR. RAPD-PCR and rep-PCR produced eight and seven molecular markers, respectively, that correlated with avirulence. Moreover, three rep-PCR and two RAPD-PCR markers were associated with avirulent races in a larger population of rust isolates from Nebraska. Using both methods we hope to identify molecular markers associated with avirulence to the *Ur-6* gene in the *U. appendiculatus* population on the USA high plains. Phylogenetic analysis of both rep- and RAPD-PCR data also produced dendrograms consisting of a small group of virulent races and a larger group comprising mainly avirulent races. Both of these PCR-based methods will also be discussed.

***Homalodisca coagulata* (Hemiptera, Cicadellidae) transmission of *Xylella fastidiosa* to almonds.** R. P. P. ALMEIDA and A. H. Purcell. Division of Insect Biology, University of California, Berkeley, CA 94720. Phytopathology 92:S3. Publication no. P-2002-0018-AMA.

Almond leaf scorch is a disease caused by *Xylella fastidiosa*, a xylem-limited bacterium transmitted by leafhopper sharpshooters. We studied some characteristics of *Homalodisca coagulata* transmission of *X. fastidiosa* to almonds. The susceptible almond variety Peerless was used for the experiments. *H. coagulata* adults were collected on citrus in Bakersfield CA. Acquisition efficiency of *X. fastidiosa* from infected almonds was low: 3/30, 2/30 and 1/30 individually tested insects transmitted to healthy plants after acquisition access periods of 1, 2 and 4 days respectively. After a 4-day acquisition access period on infected almonds, groups of 4 *H. coagulata* were transferred to healthy plants. Efficiency of transmission per group was 16, 27 and 51%, for 1, 2 and 4 days of test plant access period respectively. Groups of 4 infective insects were also caged on 1-year-old wood or green shoots of almonds; plants inoculated in both tissues became symptomatic.

Molecular characterization of a new *Xanthomonas citri* strain isolated in Florida. A. AL-SAAD and D. W. Gabriel. Plant Molecular and Cell Biology Program, University of Florida, Gainesville. Phytopathology 92:S3. Publication no. P-2002-0019-AMA.

Asiatic (A-form) citrus canker disease is caused by *Xanthomonas citri* and affects a wide range of citrus hosts, including grapefruit and lime. New strains of *X. citri*, (A^W from Florida and A* from southwest Asia) have been identified that are primarily restricted in host range to lime. Upon inoculating grapefruit with X0053 A^W strain, we observed the appearance of a hypersensitive response (HR), indicating the presence of an avirulence (*avr*) gene. Southern hybridizations revealed that both A^W and A* carry potential *pthA* homologues. *pthA* is a member of the *avrBs3/pthA* family of avirulence/pathogenicity genes. A *pthA* homologue, *pthA^W*, was cloned from X0053. *pthA^W* fully complemented a *pthA::Tn5* knockout mutation in *X. citri*, A-form, allowing canker to develop on both lime and grapefruit. This indicates that the HR on grapefruit caused by X0053 is not due to a pleiotropic avirulence effect of *pthA^W*.

Biocontrol of *Pythium* spp. on alfalfa using native fluorescent *Pseudomonas*. N. ALTIER (1), C. Pérez (2), F. Ducamp (2), L. De La Fuente (3,4), N. Bajsa (3), L. Quagliotto (3), and A. Arias (3). (1) INIA Las Brujas, CC 33085, CP 90200, Uruguay; (2) EEMAC, UDELAR; (3) IIBCE; (4) Fac. Ciencias, UDELAR. Phytopathology 92:S3. Publication no. P-2002-0020-AMA.

Seedling diseases caused by *Pythium* spp. are one of the main constraints for alfalfa establishment. Three native strains of *Pseudomonas fluorescens* (UP61, UP143 and UP148) that produce HCN, siderophores and antibiotics, with biocontrol potential to suppress *Pythium* damping-off on *Lotus corniculatus*, were tested in field trials to evaluate their efficacy on alfalfa. Eight experiments were conducted during 2000-2001. One hundred viable seeds were sown per meter, previously treated as follows: *Sinorhizobium meliloti*, *S. meliloti* + metalaxyl (3 g a.i./kg seed), *S. meliloti* + each strain. Emergence was recorded weekly and dry matter weight of three-month-old plants was determined. When weather conditions were conducive for development of damping-off, the percentage of emergence was increased up to 44% with strain UP148 and dry matter production up to 34% with strains UP61, UP143 and UP148, as compared to the control. Experiments are being repeated to validate the implementation of this strategy for the alfalfa-*Pythium* pathosystem.

Beneficial bacteria protect microaroids from bacterial blight. A. M. ALVAREZ and C. Y. Mizumoto. Dept. Plant and Environmental Protection Science, University of Hawaii, Honolulu, HI 96822. Phytopathology 92:S4. Publication no. P-2002-0021-AMA.

Ornamental aroids are susceptible to anthurium blight caused by *Xanthomonas campestris* pv. *dieffenbachiae* (Xcd). Experiments with six anthurium cultivars (Rudolph, Tropic Fire, Pink Frost, Bubble Gum, and Cotton Candy) confirmed that tissue-cultured microplants are protected by weekly sprays with mixtures of beneficial bacteria. Bioprotection was repeated five times for cv. Rudolph, starting with microplants and continuing with plants at different ages up to two years. Inoculum levels of Xcd sufficient to obtain 100% kill of nonprotected plants under warm or cool conditions killed 15 to 25% of the protected plants under cool conditions and 36% when temperatures exceeded 30 C. Continued post-inoculation treatment of protected plants resulted in regeneration of new leaves and foliage. Plants remained symptomless, and autophotographs confirmed that Xcd no longer survived in vascular tissue or foliage. Symptomless plants eventually developed flowers and were as vigorous as noninoculated plants.

Nicobifen (BAS 510 F) - The new multi-purpose fungicide for specialty crops. E. AMMERMANN (1), G. Lorenz (1), K. Schelberger (1), M. Scherer (1), K. Eicken (2), N. Götz (2), A. Harreus (2), and H. Rang (2). BASF AG, (1) 67 114 Limburgerhof; (2) 67 056 Ludwigshafen, Germany. Phytopathology 92:S4. Publication no. P-2002-0022-AMA.

Nicobifen is the common name of the new anilide fungicide discovered and developed by BASF. Its biochemical mode of action is the interference with mitochondrial respiration and subsequent production of ATP in fungal cells by inhibiting the succinate-ubiquinone oxidoreductase (complex II) in the mitochondrial electron transport chain. It effectively controls a range of fungal pathogens including Ascomycetes (powdery mildews, *Sclerotinia* spp., *Monilinia* spp.) and Deuteromycetes (*Alternaria* spp., *Botrytis* spp.). It is a highly active fungicide for grapevines, canola, peanuts, fruits, vegetables, ornamentals and turfgrass with excellent crop safety. The expected dose rate ranges from 50 to 600 g a.i./ha for food crops and from 100 to 500 g a.i./ha for turf and ornamentals. The compound has a favourable toxicological and ecotoxicological profile. It is classified by US-EPA as a 'reduced risk candidate'. Nicobifen is being developed and registered as a solo product and with various pre-mix partners.

Nicobifen - The foundation of a new fungicide family. E. AMMERMANN (1), R. Stierl (1), W. Hanke (1), M. Scherer (1), H. Ypema (2), and T. Bardinelli (2). (1) BASF AG, Limburgerhof, Germany; (2) BASF Corp., Research Triangle Park, NC. Phytopathology 92:S4. Publication no. P-2002-0023-AMA.

Nicobifen (BAS 510 F) is a new anilide fungicide developed by BASF. It interferes with mitochondrial respiration and ATP production by inhibiting the succinate-ubiquinone oxidoreductase (complex II) in the electron transport chain. Microscopic and field studies show excellent preventative activity of nicobifen against a broad spectrum of Ascomycete and Deuteromycete pathogens. Nicobifen moves in a translaminar and acropetally systemic manner. Nicobifen is efficacious against a broad range of pathogens of legumes, canola, lettuce, peanut, potato, fruiting vegetables, and turfgrass with excellent crop safety. Nicobifen is also in development as a pre-mix fungicide (BAS 516 F) with pyraclostrobin. BAS 516 F effectively controls the majority of foliar fungal pathogens on small berries, bulb vegetables, carrots, grapes, onions, tree nuts, stonefruits, and strawberries. Nicobifen and BAS 516 F are currently under registration review by the EPA and the PMRA. Both nicobifen and BAS 516 F will make excellent foundations for disease control programs and fungicide resistance management.

The invasion of an exotic dodder in Houston, TX. D. APPEL (1), K. Camilli (2), T. Kurdyla (3), and C. A. Hientze (4). (1,3) Dept. of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843; (2) Texas Forest Service, 1101 Camino La Costa Rd., Room 215, Austin, TX 78761; (4) City of Houston, 2999 S. Wayside, Houston, TX 77023. Phytopathology 92:S4. Publication no. P-2002-0024-AMA.

In the summer of 2001, *Cuscuta japonica* Choisy, a dodder species native to east Asia, was observed in Houston, TX. This plant parasite was found at five locations in the city. The infected hosts were live oak (*Quercus virginiana* Mill.), waxleaf ligustrum (*Ligustrum japonicum*), crape myrtle (*Lagerstroemia indica*), Chinese tallow (*Sapium sebiferum*) and Chinese photinia (*Photinia serrulata*). Sites consisted of planted hosts on residential and commercial plantings. Vines were found flowering but no seeds were produced. The parasite was originally thought to be giant tree dodder (*C. exaltata*), a less aggressive native species. Asian dodder is listed on the Federal Noxious Weed list. There have only been a few previous introductions of *C. japonica* in the U.S. The source of the Houston outbreak is unknown. A program based on the use herbicides, host removals, eradicated pruning, burning of infected plant materials, and long-term monitoring is underway.

Greenhouse study of biological induction of resistance in onion against *Alternaria porri*. J. ARBOLEYA (1), A. B. da Rocha (2), I. Widders (1), B. Zandstra (1), and R. Hammerschmidt (2). (1) Dept. of Horticulture; (2) Dept. of Plant Pathology, Michigan State University, East Lansing, MI 48824. Phytopathology 92:S4. Publication no. P-2002-0025-AMA.

Induced disease resistance is one potential approach to disease control. Although induced resistance has been broadly studied, there is little information about this phenomenon in onion. The objective of this study was to determine if prior infection with necrosis-inducing pathogens would induce resistance in onion foliage against purple blotch, caused by *Alternaria porri*. Necrosis was induced on onion leaves in the greenhouse with the cucumber pathogens *Colletotrichum lagenarium* or *Didymella bryoniae*. The first non-senescent leaf of plants at the 7th leaf stage was injected at 10 sites with 10 microliters of a conidial suspension of each fungus (1×10^6 spores/ml). *D. bryoniae* more effectively caused localized necrosis and better induced resistance to *A. porri* than *C. lagenarium*. Purple blotch lesions were significantly smaller (9.8 mm) on *D. bryoniae* induced plants, as compared to control plants (14.9 mm). In addition, non-induced plants usually showed purple blotch lesions 2 days earlier and expanded faster than the induced.

Replication of Tomato spotted wilt virus ingested into the alimentary canal of adult thrips. F. M. ASSIS FILHO, C. M. Deom, and J. L. Sherwood. Dept. Plant Pathology, The University of Georgia, Athens, GA 30602. Phytopathology 92:S4. Publication no. P-2002-0026-AMA.

Previous studies using immunocytochemistry indicated there was limited TSWV replication in the midgut epithelial cells of *Frankliniella occidentalis* (western flower thrips - WFT) and *Thrips setosus* fed on TSWV-infected tissue as 1-2-day-old adults. Using immunofluorescence microscopy coupled with a monoclonal antibody against NSs protein to detect TSWV replication, we observed in *Frankliniella fusca* (tobacco thrips) and WFT that TSWV replication in epithelial cells was limited to midgut-1 in 1-2-day-old adult thrips fed on virus-infected tissue. Replication was also observed in the muscle cells surrounding the entire midgut that was similar in scope to that observed when virus is acquired by larva. No immunolabel was observed in foregut, salivary glands, and ligaments. Acquisition of TSWV diminished in older adults, with virus not being acquired by 14-15-day-old adults. This is the first report of replication of TSWV in muscle cells of adult thrips fed on virus-infected plants. The biological significance of these observations is being determined.

Protein analysis in resistant and susceptible dogwood cultivars challenged with *Microsphaera pulchra*. F. J. AVILA (1), M. T. Mmbaga (1), and L. Myles (2). (1,2) TSU Cooperative Agricultural Research Program, Nursery Crop Research Station, McMinnville, TN 37110; (2) Dept. of Biology, Nashville, TN 37209. Phytopathology 92:S4. Publication no. P-2002-0027-AMA.

Proteins that may be associated with powdery mildew resistance in *Cornus florida* were identified in resistant and susceptible cultivars. Detached leaves were inoculated with powdery mildew conidiospores and incubated at room temperature for 48 or 72 h. Non-inoculated leaves were used as controls. Total protein was extracted and analyzed using two-dimensional isoelectric focusing/SDS-polyacrylamide gel electrophoresis. Different protein patterns were detected in resistant and susceptible plants after inoculation. New proteins bands detected in the susceptible cultivars may be proteins that

could be involved in disease establishment. Among the new protein bands detected in the resistant cultivars after inoculation, seven bands showed similar isoelectric points and molecular weights to Pathogenesis Related (PR) proteins. These results suggest that the resistant mechanism in dogwood to powdery mildew may involve PR-protein synthesis.

Plectosporium blight of pumpkins, a new fungal disease in Illinois. M. Babadoost. Dept. Crop Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S5. Publication no. P-2002-0028-AMA.

Plectosporium blight (formerly called Microdochium blight) of pumpkin, caused by the fungus *Plectosporium tabacinum*, was diagnosed in Illinois for the first time in 2000. The disease was first observed in Richland County in August, then was identified in all of the pumpkin fields visited throughout the state. All parts of pumpkin plants, including leaves, vines, and fruit, were affected. Numerous spindle-shaped, white lesions were observed on vines, leaf veins, petioles, and peduncles. On leaves, the lesions were restricted to veins and no symptoms were observed on the interveinal tissue. Severely infected vines were white, dry, and brittle. The disease caused severe defoliation and death of the foliage. The fungus infected both green and orange fruit causing silver russetting on the rind. Fruit stems were covered with white lesions. The fungus produced hyaline, slightly curved, two-celled conidia on infected tissues and in culture media. Plectosporium blight caused up to 80% yield losses in pumpkin fields in 2000. In 2001, however, the disease occurred sporadically with less than 10% of fruit exhibiting symptoms.

Effects of selected fungicides and biocontrol agents on the incidence of internal discoloration of horseradish root. M. Babadoost and S. Z. ISLAM. Department of Crop Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S5. Publication no. P-2002-0029-AMA.

Internal discoloration of horseradish roots, incited by *Verticillium dahliae*, *V. longisporum*, and *Fusarium* species, causes up to 100% yield loss in Illinois. In a field trial, conducted near Collinsville, Illinois, three fungicides, mefenoxam (Apron XL LS), fludioxonil (Maxim 4FS), and thiophanate-methyl (Topsin-M 70W); nine biocontrol agents, Biovert, BioYiled, MSDS G-41, Greenex, QRD-131, QRD-132, QRD-137, SoilGard 12G, and T-22 PB; and a plant activator (Actigard 50 WG) were tested for their effectiveness on controlling the root discoloration. Horseradish 1573, a cultivar susceptible to the root discoloration, was used. Horseradish sets were treated with fungicides by soaking in fungicide suspensions at labeled rates for 5 min. To apply the biocontrol agents, sets were dipped in water, placed in a plastic bag with biocontrol agent, and shaken to coat. In addition to seed-treatment, two of the biocontrol agents were also added into the plant furrow (1 lb/40 ft) prior to planting. Actigard was sprayed onto the plants in a weekly schedule beginning second week after plants emerged from soil. The incidence of the root discoloration was 36, 42, 50, and 38% lower in plots treated with Maxim 4FS, MSDS G-41, QRD-137, and SoilGard 12G, when compared to that of untreated plots.

Two new bacterial diseases on watermelons in Illinois. M. BABADOOST and N. Pataky. Dept. Crop Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S5. Publication no. P-2002-0030-AMA.

Two bacterial diseases of watermelon, bacterial rind necrosis, caused by an *Erwinia* species, and bacterial fruit blotch, caused by *Acidovorax avenae* subsp. *citrulli*, were reported in Illinois for the first time. Bacterial rind necrosis was observed on the watermelon cultivar Summer Sweet in a commercial field in Union county in 2000. In most of the infected fruit, the entire interior of the rind exhibited brown and corky necrosis. No external symptoms or flesh infection of fruit were observed. Bacterial fruit blotch of watermelon was observed on cultivars Crimson Sweet, Royal Sweet, and Sugar Lee in six fields in Iroquois, Kankakee, and Mason counties in 2001. No symptoms of the disease were observed until fruit maturity. At maturity, the fruit developed dark green, water-soaked lesions, and the rind epidermis was cracked. The incidence of fruit infection with *A. avenae* subsp. *citrulli* in the fields ranged from 5 to 75%. The occurrence of these bacterial diseases in Illinois is important because (i) watermelon is widely grown in Illinois, (ii) *A. avenae* subsp. *citrulli* also infects other melons and pumpkins, and (iii) Illinois grows approximately 1,000 ha of melons and 8,000 ha of pumpkins.

Bell peppers resistant to Phytophthora blight. M. Babadoost and S. Z. ISLAM. Department of Crop Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S5. Publication no. P-2002-0031-AMA.

Fifty-six cultivar/lines of bell pepper were evaluated in 2001 for resistance to *Phytophthora* blight, caused by *Phytophthora capsici*. Eight-week-old

seedlings were inoculated with *P. capsici* by adding 2 ml of a zoospore suspension (105 spores/ml) at the base of each seedling grown in pots in a greenhouse. Control seedlings were treated with water. Sixteen seedlings of each line were evaluated for development of lesions on the stems, wilting, and death until 24 days after inoculation. Disease development was least on three cultivars, Emerald Isle, Paladin, and Reinger; and four lines, Abbott-1, Abbott-2, Abbott-13, and Syngenta-7326. These cultivars/lines and California Wonder, a cultivar susceptible to *Phytophthora* blight, were further evaluated in a field naturally infested with *P. capsici*. Eight-week-old seedlings were transplanted into raised beds with drip irrigation and black plastic mulch on 11 June. Treatments were organized in a randomized complete block with four replications. Each experimental unit included 10 plants. Development of the disease was evaluated throughout the season. On 2 September, 90, 93, 93, 83, 52, 75, 93, and 17% of plants of Emerald Isle, Paladin, Reinger, Abbott-1, Abbott-2, Abbott-13, Syngenta-7326, and California Wonder, respectively, were free of symptoms.

Interaction of gray leaf spot and European corn borer on stalk rot of corn. K. K. BAHK and A. P. Grybauskas. Dept. Natural Resource Sciences and Landscape Architecture, University of Maryland, College Park, MD 20742. Phytopathology 92:S5. Publication no. P-2002-0032-AMA.

Stalk rot of corn (*Zea mays*) reduces attainable yield directly by reducing grain fill and indirectly when lodging affects harvest efficiency. Infection by stalk rot agents increases with insect feeding, and any plant stress is believed to predispose plants to infection. This study examined European corn borer (*Ostrinia nubilalis*) (ECB) and gray leaf spot (*Cercospora zea-maydis*) (GLS) for their individual contribution to stalk rot and whether an interaction between the two factors synergistically worsens stalk rot. A GLS susceptible and GLS resistant hybrid and their Bt transformed near-isogenic pairs were used. There were 8 treatments: the 4 hybrids grown with chemical management for insect and foliar disease (pest reduced conditions), and grown with ECB and *C. zea-maydis* infestation. GLS development was rated and stalks were examined for ECB tunneling. Stalk rot was characterized by the number of plants lodged, stalk strength and amount of pith disintegration. Preliminary results indicate that although ECB and GLS both contribute to stalk rot, there is no synergistic interaction.

Characterization of transfer regions in the mollicute *Spiroplasma kunkelii*: Putative pathogenicity islands? X. BAI, T. Fazzolari, and S. Hogenhout. Department of Entomology, OARDC, The Ohio State University. Phytopathology 92:S5. Publication no. P-2002-0033-AMA.

In several pathogenic bacteria, transfer regions (*tra*) are typically associated with Type IV secretion and virulence, and *tra* genes are often part of pathogenicity islands. We identified four *tra* regions, each containing a putative *traE* gene, in the genome of the leafhopper-transmitted corn stunt pathogen *Spiroplasma kunkelii* strain M2. The deduced protein sequences of open reading frames surrounding the *traE* genes had significant similarities to bacterial virulence, adhesin, and/or spiroplasma virus proteins. *traE* expression was investigated by Northern blot analysis. Results showed differential expression of *traE2*, *traE3* and *traE4* in culture and in infected insects and plants, whereas *traE1* expression was not detected. Interestingly, plants exposed to leafhoppers injected with *S. kunkelii* strain CS-2B lacking *traE1* and *traE4*, and *S. kunkelii* strain PU8-17 lacking *traE1*, *traE3* and *traE4* did not show symptoms. These results suggested involvement of *tra* regions in *S. kunkelii* insect transmission and/or plant pathogenicity.

Yellow dwarf virus quantification by real-time PCR during disease development in resistant and susceptible plants. B. Balaji (1), D. B. Bucholtz (2), and J. M. ANDERSON (2). (1) Agronomy Department; (2) USDA-ARS. Phytopathology 92:S5. Publication no. P-2002-0034-AMA.

Reliable detection and quantification of yellow dwarf virus (YDV) is a critical component in managing these viral diseases in small grain cereal crops. Recently, Real-time Quantitative RT-PCR was introduced to study gene expression. We are applying this technique to detect and quantify YDV using primers specific for BYDV-PAV and CYDV-RPV coat protein genes. Given the higher sensitivity of RT-PCR and the advantage of using a real time PCR instrument, we have successfully utilized this method to detect BYDV and CYDV and examined disease development in a YDV resistant wheatgrass and susceptible oat by quantifying the level of virus. Both the wheatgrass and oats were infested with viruliferous aphids and plants collected at regular intervals for 12 days. RNA from each harvest and random hexamers was used for first strand cDNA synthesis that was used as the template for Real-time PCR. The procedure described is reproducible, sensitive, robust and has the potential to be used as a rapid diagnostic tool for BYDV and CYDV and as a means for examining mechanisms of virus resistance.

Efficacy of bacteriophage formulations for control of bacterial spot on tomato. B. BALOGH (1), J. B. Jones (1), M. T. Momol (2), S. M. Olson (2), A. Obradovic (1), P. King (2), and L. E. Jackson (3). (1) Univ. of Florida, Plant Pathology Dept, PO Box 110680, Gainesville, FL 32611; (2) Univ. of Florida, NFREC, Quincy, FL 32351; (3) Agriphi Inc., Logan, UT 84323. *Phytopathology* 92:S6. Publication no. P-2002-0035-AMA.

Bacteriophages have recently been deployed as an alternative method for controlling bacterial spot on tomato incited by *Xanthomonas campestris* pv. *vesicatoria*. However, the efficacy of phage is greatly reduced due to its short residual activity on plant foliage. The following three formulations were developed that significantly increased phage longevity on the plant surface: 1.) 0.5% pregelatinized corn flour (PCF, Lauhoff Grain Co.) + 0.5% sucrose; 2.) 0.5% Casecrete™ NH-400 (American Casein Co.) + 0.5% sucrose + 0.25% PCF; 3.) 0.75% powdered skim milk + 0.5% sucrose. The formulations and the non-formulated phage product significantly reduced disease severity in field trials on tomato compared to the standard copper-mancozeb treatment by 22, 33, 27 and 19%, respectively.

Nicobifen (BAS 510 F): A new fungicide for use on vegetables, field crops and turfgrass. T. R. BARDINELLI (1), H. L. Ypema (1), J. S. Barnes (1), H. C. Wetzel III (1), and S. Chapman (2). (1) BASF Corp. Research Triangle Park, NC 27709; (2) BASF, Morden, Manitoba R6M 1Y5, Canada. *Phytopathology* 92:S6. Publication no. P-2002-0036-AMA.

Nicobifen (BAS 510 F) is a new fungicide being developed by BASF. Nicobifen is the first active ingredient that utilizes a biochemical mode of action different from most other current fungicides against a novel range of target diseases in many important crops. These crops include legume crops, canola, fruiting vegetables, lettuce, peanut, potato, and turfgrass. Nicobifen is an excellent addition to crop management programs for both disease control and fungicide resistance management. On legume crops, nicobifen is effective against *Sclerotinia* white mold and *Ascochyta* blight. Nicobifen is very effective against white mold of canola. Lettuce pathogens controlled include lettuce drop and *Botrytis* rot. On peanut, it is effective against *Sclerotinia* blight and several foliar pathogens. On potato, nicobifen provided excellent control of white mold and early blight. On fruiting vegetables, nicobifen was very effective against early blight, *Septoria* and *Botrytis*. On turfgrass, nicobifen provides excellent control of dollar spot.

Host-response based screening of biological control agents. R. L. BARGABUS, N. K. Zidack, J. E. Sherwood, and B. J. Jacobsen. Dept. Plant Sciences/Plant Pathology, Montana State University, Bozeman, MT 59717-3150. *Phytopathology* 92:S6. Publication no. P-2002-0037-AMA.

Biological control agents (BCAs) that induce plant systemic resistance (SR) have shown disease control in several hosts. Serendipitous identification of these BCAs normally involves disease control and mode of action studies that are time and labor intensive. We have demonstrated elicitation of SR in sugar beet by treatment with *Bacillus mycoides*, a phyllosphere-colonizing, non-pathogenic BCA. SR induction by the BCA resulted in increased production of pathogenesis-related (PR) proteins (peroxidase, chitinase, beta-1,3-glucanase, PR-1) and active oxygen species (AOS) (H_2O_2 , superoxide) as determined by northern and western analysis and in-gel activity and enzymatic assays. We propose using enzymatic assays to identify SR-inducing BCAs, which directly identifies this mode of action, reducing selection time from weeks to days or hours. Preliminary results verified differentiation between SR-inducing and non-inducing Bacilli using chitinase and beta-glucanase colorimetric assays. PR-protein and AOS production in sugar beet to a range of SR-inducing and non-inducing Bacilli will be reported.

Effects of fire on annual infection levels of big bluestem by *Puccinia andropogonis*. C. W. BARNES and J. V. Groth. Dept. of Plant Pathology, University of Minnesota. *Phytopathology* 92:S6. Publication no. P-2002-0038-AMA.

Fire influences the presence and distribution of plants, and their associated plant diseases in natural ecosystems. Plots at a Long Term Ecological Research facility in Cedar Creek Minnesota, were utilized to investigate the effects of fire on the annual changes in rust infection in this natural prairie. Infection by *P. andropogonis* was measured on both its hosts, comandra (C), and big bluestem (BBS), over a four year period in two fields, one burned every fourth year, and one without burns. Fire reduced aecia on C by over 96 percent compared to years previous and following the fire, but there was no change in the overall average uredinal infection levels in BBS plots within or between fields of that year. This is likely due to the efficiency of uredinal spore dispersal. However, variation in rust levels on individual BBS plots between years was affected by fire. Simple regression of rust levels in pairs

of years gave $R^2 = 0.06$, regressing the year following the burn to the year of the burn, compared to $R^2 = 0.37$ for the non-burned field, supporting that fire can disrupt perennial disease cycles. Field differences in R^2 values hold true for all year by year comparisons.

Fungicide sensitivity differs among newly discovered fungi in the sooty blotch and flyspeck complex on apples. T. L. BARRETT, J. C. Batzer, M. L. Gleason, and P. M. Dixon. Dept. Plant Pathology, Iowa State University, Ames, IA 50011. *Phytopathology* 92:S6. Publication no. P-2002-0039-AMA.

Sensitivity to three fungicides (captan, thiophanate-methyl and ziram) among seven genetically distinct fungal groups causing sooty blotch and flyspeck was assessed *in vitro*. Plugs of two isolates per fungal group were placed on six-well tissue culture plates containing water agar amended with a range of concentrations of each fungicide. Six replicate plates per isolate were incubated at 25°C. After 3 wk, colony diameter was measured and ED₅₀ of each isolate-fungicide combination was determined. ED₅₀ differed significantly ($P < 0.05$) among the fungal groups for both captan and thiophanate-methyl. This finding provides new evidence that fungi within the sooty blotch and flyspeck complex respond differentially to fungicides commonly used to suppress the complex.

Discrete speck, a putative newly discovered fungus in the sooty blotch and flyspeck complex on apples. J. C. BATZER, M. L. Gleason, and L. H. Tiffany. Dept. Plant Pathology, Iowa State University, Ames, IA 50011. *Phytopathology* 92:S6. Publication no. P-2002-0040-AMA.

A previously undescribed fungus associated with sooty blotch disease of apple fruit (*Malus domestica*) was found in a 2000 survey of several Missouri and Iowa orchards. The discrete speck (DS) fungus was visible on apple as tiny, dark specks. DS was similar in appearance to fly speck *Zygothiala jamaicensis*. Genbank BLAST search of internal transcriber spacer region sequences of the DS fungus resulted in 98% homology (482/488 base pairs) with *Dissoconium aciculare*, reported as a hyperparasite on *Erysiphe* sp. On water agar (WA), DS produced one-celled obovoid conidia, 6.0-12.0 × 3.6-6.0 μm, and two-celled subhyaline conidia, constricted at the septum, 12-20.4 × 4.5-8.4 μm. Conidia were formed in pairs and were forcibly discharged together. Mature spermatogonia were dark, ostiolate and had thick-walled setae and radially arranged cells. When transferred from apple to WA, spermatogonia exuded pale yellow, ovoid, spermatia-like cells, 4.8 × 3.0 μm, that did not germinate. A survey of apples from 25 orchards in Iowa, Missouri, Wisconsin, and Illinois was conducted to determine the geographic prevalence of DS.

Variation in leaf wetness duration within apple tree canopies. J. C. BATZER, M. L. Gleason, K. J. Koehler, and S. E. Taylor. Dept. Plant Pathology, Iowa State University, Ames, IA 50011. *Phytopathology* 92:S6. Publication no. P-2002-0041-AMA.

Spatial heterogeneity of leaf wetness duration (LWD) within the canopy of 11-yr-old, semi-dwarf apple trees (3.7 to 4.2 m tall, north-south row orientation, cv. Golden Delicious) was quantified in an orchard near Gilbert, Iowa. Hourly averages of LWD at 12 canopy positions were taken in 3 trees from mid-July to September 2000 and in 4 trees from late May to mid-September 2001. Painted electronic wetness sensors (Model 237, Campbell Scientific, Inc.) were mounted at a 45-degree angle from horizontal, and facing north, at 4 lateral, east-west positions within the canopy at each of 3 heights (3.7 m, 2.4 m, 1.2 m.). Daily LWD (noon to 11 a.m.) was analyzed using a SAS Mixed Models Procedure. During days with >0.25 mm precipitation, LWD was 134% longer in the upper and middle than in lower canopies. During days with no precipitation, LWD varied significantly within a canopy from west to east ($P = 0.00075$) and LWD was 197% longer in upper eastern than lower western canopies. Effect of canopy LWD heterogeneity on disease management was simulated using a sooty blotch and flyspeck warning system based on LWD.

Expansion of the sooty blotch and flyspeck complex on apple using ribosomal DNA. J. C. BATZER, M. L. Gleason, T. Harrington, and W. Chen. Dept. Plant Pathology, Iowa State University, Ames, IA 50011. *Phytopathology* 92:S6. Publication no. P-2002-0042-AMA.

Sequences of the large subunit (LSU) and internal transcriber spacer (ITS) regions of rDNA were used to group 500 isolates of sooty blotch and flyspeck (SBFS) obtained from apples grown in 9 orchards in Iowa, Missouri, Wisconsin and Illinois. The original sign for each isolate was preserved on pressed apple peels. Sequences were compared with known SBFS fungi (*Zygothiala jamaicensis*, *Peltaster fruticosa*, *Leptodontium elatius*, *Geastrumia polystigmatis*, and *Stomiopeltis versicola*) from North

Carolina. Sequences were sorted into 22 clades. ITS and LSU sequences from two clades were grouped with *P. fructicola* and *Z. jamaicensis*. Twenty new clades were found. Each clade corresponded to a distinct morphology on apple and a unique colony type in culture. Prevalence of clades among the orchards was highly variable. Fungi from ten of the most prevalent clades were inoculated onto green apples and incubated in the field and in growth chambers. Signs were re-isolated and ITS sequences were identical to the original isolates.

Transformation of *Pyrenophora teres*. T. M. BECKER, C. M. Donohue, S. Neate, and J. B. Rasmussen. Department of Plant Pathology, North Dakota State University, Fargo, ND. Phytopathology 92:S7. Publication no. P-2002-0043-AMA.

Net blotch, caused by, *Pyrenophora teres*, is a significant foliar disease of barley in the northern Great Plains. The objective of this work was to develop a transformation system for the fungus for use by our group. A combination of beta-D-glucanase and driselase released over 1 billion protoplasts from mycelium of the fungus. A transformation vector developed in the lab, designated pDAN, was used for transformation. pDAN was developed by subcloning into pBluescript II a hygromycin resistance gene, originally obtained from pMOCosX, that is driven by the pcp1 promoter. Several transformants are obtained per experiment. This transformation protocol should facilitate be useful for developing tagged strains of the fungus for epidemiological studies in the greenhouse. The transformation system may ultimately be useful for identifying pathogenicity and virulence determinants in this fungus.

The *Magnaporthe pth1* mutant forms defective appressoria. J. Beckerman (1), C. Cortes (2), C. FILIPPI (3), J. Sweigard (4), B. Valent (5), and D. Ebbole (3). (1) Univ. Minnesota Ext Service, St. Paul; (2) Esc. de Quimico-Farmacobio., Morelia, Mexico; (3) Plant Pathol & Microbiol., Texas A&M Univ., College Station; (4) DuPont Ag. Biotech., Newark; (5) Plant Pathology, Kansas State Univ., Manhattan. Phytopathology 92:S7. Publication no. P-2002-0044-AMA.

M. grisea pth1 gene was identified in a screen for pathogenesis mutants and encodes an F-box LRR protein similar to yeast Grr1. Grr1 plays a key role in targeting proteins for degradation by acting as a ubiquitin-ligase. *pth1* mutants produce defective appressoria and thus are unable to penetrate host tissue. However, typical lesions were observed after wound inoculations suggesting that disruption of this gene does not affect host colonization but is specific to the penetration process. Turgor generation and carbon mobilization into appressoria was evaluated. These studies support the view that protein ubiquitination and turnover play an important role in appressorium maturation.

Polymorphism of the newly cloned chorismate mutase of the soybean cyst nematode, *Heterodera glycines*, among the virulent and avirulent resistance-breaking inbred lines. S. BEKAL, K. N. Lambert, and T. L. Niblack. Dept. Crop Science, University of Illinois, Urbana, IL 61801. Phytopathology 92:S7. Publication no. P-2002-0045-AMA.

Chorismate mutase (CM) is a protein secreted from parasitic nematode esophageal glands. The CM gene was first cloned from root knot nematode *Meloidogyne javanica* and recently reported from *Globodera rostochiensis*. The CM is believed to be involved in nematode parasitism. Using degenerate primers to the conserved regions of both known nematode CM proteins, we amplified the CM of *H. glycines*, the soybean cyst nematode (SCN), from cDNA extracted from four day old parasitic juveniles. The SCN-CM gene has been cloned and sequenced. The derived protein sequence of the SCN-CM showed about 25% identity to that of *M. javanica* and about 70% identity to that of *G. rostochiensis*. DNA gel blots of the SCN inbred lines; which differ in their ability to overcome soybean and tomato resistance showed polymorphism when hybridized to the SCN-CM cDNA probe. Our preliminary data show that the SCN-CM is a multigene family, in which some members correlate with virulence to soybean and tomato.

Genetic analysis of resistance in lettuce against *Verticillium dahliae*. R. G. BHAT (1), E. J. Ryder (2), and K. V. Subbarao (1). (1) Plant Pathology Department, University of California, Davis; (2) USDA-ARS, 1636 E. Alisal St., Salinas, CA 93905. Phytopathology 92:S7. Publication no. P-2002-0046-AMA.

Inheritance of resistance to *Verticillium dahliae* in lettuce was studied by crossing a susceptible or resistant plant of cultivar Salinas (black seeds) with two resistant plants of PI 120938 (white seeds). Parental plants, four F₁ hybrids and their F₂ progenies were inoculated in the greenhouse with an

isolate of *V. dahliae* from lettuce. In addition, F₂ progenies from four F₁ hybrids whose reactions were unknown were also inoculated. Disease reaction and seed color were recorded at the time of seed-harvest. Inoculated F₁ plants were susceptible to *V. dahliae*, and their seeds were black. In the F₂ progenies, segregation for seed color was 3 black:1 white, indicating that the crossing was successful. Based on chi-square analyses, a two-gene model with a ratio of 9:7 for susceptible:resistant plants fitted the F₂ data. This indicated complementary inheritance with both dominant alleles required for susceptibility. Observed data were in agreement with the expectation of the model when segregation of seed color was included for three-gene analyses. Minor genes might also have some effects on disease resistance.

Ecology of *Alternaria alternata* on citrus. A. BHATIA (1), T. Peever (2), K. Akimitsu (3), L. Carpenter-Boggs (2), and L. W. Timmer (1). (1) CREC, University of Florida, Lake Alfred, FL; (2) Dept. Plant Pathology, Washington State University, Pullman, WA; (3) Faculty of Agriculture, Kagawa University, Miki, Japan. Phytopathology 92:S7. Publication no. P-2002-0047-AMA.

Two distinct pathotypes of *Alternaria alternata* cause Brown Spot of fruit and leaves of Minneola tangelo and rough lemon in Florida. Several hundred isolates have been collected from healthy leaf tissue and leaf lesions of these hosts to study the ecology of both pathotypes on citrus. When inoculated on leaves, most healthy tissue isolates and a significant proportion of lesion isolates were not pathogenic. However, isolation and plating, and histological studies with SEM revealed that both types of isolates penetrate the leaf via stomata. Healthy tissue isolates sporulated less abundantly than lesion isolates in culture. Both lesion and healthy tissue isolates from rough lemon leaves caused postharvest black rot of citrus fruit. These results and data on the distribution of ACT-toxin sequences will be related to a molecular phylogeny to gain insight into the evolution of these *A. alternata* on citrus.

Molecular phylogenetic relationships among *Embellisia*, *Nimbya*, to *Alternaria* and related genera based upon analysis of ITS, mt SSU and *gpd* sequences. D. M. BIGELOW, C. E. Michoski, and B. M. Pryor. Department of Plant Pathology, University of Arizona, Tucson, AZ 85721. Phytopathology 92:S7. Publication no. P-2002-0048-AMA.

DNA sequences from rDNA and protein-coding regions were determined for two *Nimbya* and six *Embellisia* spp. and were compared to those from *Alternaria*, *Stemphylium* and other related genera. Sequences determined included rDNA from the nuclear internal transcribed spacer region (ITS1/5.8S/ITS2) and the mitochondrial small subunit (SSU), and a portion of the glyceraldehyde-3-phosphate dehydrogenase gene (*gpd*). Phylogenetic analysis was performed on each data set separately, then combined for total evidence analysis using methods of distance and maximum parsimony. Results revealed that *Embellisia* and *Nimbya* are sister groups to *Alternaria* and are more closely related to *Alternaria* than is *Stemphylium*. Five of the *Embellisia* spp. formed a monophyletic clade. However, *Embellisia indefessa* was more closely related to *Alternaria* and *Ulocladium* spp. suggesting that this group is polyphyletic. The *Nimbya* spp. generally resolved together, but the clade was poorly supported in bootstrap analysis. Potential revisions of taxonomy are discussed.

Maize secondary metabolites and *Fusarium* mycotoxin production. A. C. BILY (1,2), L. M. Reid (3), C. Lefevre (1), B. A. Blackwell (3), M. Savard (3), C. Regnault-Roger (2), J. T. Arnason (1), and B. J. R. Philogène (1). (1) Dept. of Biology, University of Ottawa, Ottawa K1N6N5, ON; (2) IBÉAS Université de Pau et des Pays de l'Adour, Pau 64000 France; (3) Eastern Cereal & Oilseed Research Centre, Agriculture & Agri-Food Canada, Ottawa K1A 0C6, ON. Phytopathology 92:S7. Publication no. P-2002-0049-AMA.

During fungal infection, cell wall bound metabolites or conjugated glycosides are released into the extracellular medium. To better understand the relationship between these compounds and *Fusarium graminearum*, we tested the effect of various common maize metabolites on *F. graminearum*'s ability to produce mycotoxins in a two-stage liquid culture that promotes high levels of mycotoxin production. Levels of mycotoxins were monitored by HPLC and LC/MS. Among the 6 metabolites tested, ferulate at 30 ppm was found to inhibit 50% of the 15-ADON appearance in the medium. Compared to the control culture, no decrease of mycelium growth and no delay in mycotoxin production were observed. Radiolabelled ¹⁴C ferulate was used to further investigate the mode of action of this compound, which is well known for its fungistatic effect and antioxidant properties.

Characterization of isolates of *Discula destructiva* in Michigan using AFLPs and RFLPs. Z. J. BLANKENHEIM and G. C. Adams. Michigan State University. Phytopathology 92:S7. Publication no. P-2002-0050-AMA.

Discula destructiva, the causal agent of Dogwood Anthracnose, is currently in several native stands of *Cornus florida* in Michigan. Infected stands occur in the municipalities of Kalamazoo, Paw Paw, Muskegon, Grand Rapids and Augusta and have been infected for approximately 12, 6, 5, 5 and 2 years, respectively. Infected stands are generally located near landscaped suburban housing and likely became infected from imported nursery stock planted nearby. Microscopic examination of fruiting bodies on dead twigs has revealed *D. destructiva* on *C. florida* and *C. kousa* being imported from several states. Spread of the disease appears limited in Michigan by the fragmented nature of the natural dogwood stands. Isolates from previously infected stands, newly infected stands, and incoming nursery stock are being characterized using molecular markers. AFLPs and RFLPs have shown differences in origin among isolates causing individual stand infection.

Evaluation of fungicide applications for management of *Cercospora leaf spot* on sugar beets. E. S. Blehm and R. M. HARVESON. University of Nebraska Panhandle Research and Extension Center, Scottsbluff, NE 69361. Phytopathology 92:S8. Publication no. P-2002-0051-AMA.

Experiments were conducted at the Panhandle Research and Extension Center, Scottsbluff, NE during 2000-2001. The objective was to compare the efficacy of systemic and protectant fungicides for managing *Cercospora* leaf spot of sugar beets. Both studies relied upon natural infection, and disease development was monitored with leaf severity ratings using a non-linear scale of 0-9. Additional data collected from both studies included root and sucrose yields, and sugar percentage. The 2000 study was furrow irrigated and the 2001 study was sprinkler irrigated. Significant disease severity differences were observed for all treatments in both years compared to controls, but yield differences were only observed in 2001. This is presumably due to more favorable conditions for disease in 2001. Both studies suggest that the timing of application is more influential in reducing disease than the type of fungicide used, and that early fungicide application is crucial during years when the environment is favorable for severe disease development.

Pythium stem canker on grain amaranth. C. C. BLOCK (1), J. W. Van Roekel (1), and L. Robertson (2). (1) USDA-ARS-NCRPIS, Iowa State University, Ames, IA 50011; (2) Dept. Plant Pathology, North Carolina State University, Raleigh, NC 27695. Phytopathology 92:S8. Publication no. P-2002-0052-AMA.

An unusual stem canker was observed on mature grain amaranth plants (Plainsman cultivar) in Nebraska and Missouri production fields during August 2000 and in Iowa plots during 2000 and 2001. Dry, tan cankers with thick black borders, similar in appearance to blackleg of crucifers, developed near the soil line. The cankers often spread 15-45 cm up the stems. In attempts to identify the causal agent, we isolated several *Phoma*-like fungi along with *Pythium* spp. Each isolate was tested for pathogenicity on greenhouse plants by drilling small holes in the stems, introducing fungal mycelium, and sealing with petrolatum. Plants were observed for 6 weeks, but only the *Pythium* isolates caused disease, usually within 4-7 days. Canker symptoms were also demonstrated on plants grown in potting mix amended with the *Pythium* cultures. All of the isolates were identified as *P. aphanidermatum*, which has been reported from amaranth, but is usually associated with a soft, basal stem rot. One *Pythium* isolate was derived from a basal stem rot, but did cause stem canker symptoms when inoculated onto Plainsman plants.

Induction of systemic resistance/susceptibility in *Pinus nigra* inoculated with *Sphaeropsis sapinea*. J. T. BLODGETT, M. Bellizzi, and P. Bonello. Dept. of Plant Pathology, Ohio State Univ., Columbus, OH 43210. Phytopathology 92:S8. Publication no. P-2002-0053-AMA.

The objective of this study was to test if inoculation of Austrian pines (*Pinus nigra*) with the fungal pathogen *Sphaeropsis sapinea* results in systemic induced resistance or susceptibility to subsequent colonization by *S. sapinea*. Six-year-old, greenhouse-grown Austrian pines were wounded at the stem base and treated with either the A or B morphotypes of *S. sapinea* (inducing inoculum); control trees were mock inoculated. At 21 days, the pines were challenge inoculated with either an A isolate or mock inoculated, on either: 1) the stem, 25 cm above the initial treatment sites; or 2) branch tips. Inoculation at the stem base with either morphotype significantly ($P < 0.001$) induced resistance in the upper stem. However, inoculation at the stem base significantly ($P < 0.001$) induced susceptibility in shoot tips, with the less aggressive B morphotype inducing inoculum stimulating greater susceptibility. This study describes a novel phenomenon in which the same pine host displays either systemic induced resistance or systemic induced susceptibility to the same pathogen, depending on the site of secondary infection.

PCR-detection of fumonisin- and trichothecene-producing *Fusarium* species. B. H. BLUHM, J. E. Flaherty, and C. P. Woloshuk. Dept. Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S8. Publication no. P-2002-0054-AMA.

Fumonisin and trichothecene mycotoxins pose serious health risks to animals and humans. A method to rapidly detect fungi that produce these mycotoxins in raw commodities is needed by the food processing industry. We are exploring a PCR-based detection strategy using three *Fusarium*-specific primer sets: one specific for the internal transcribed spacer (ITS) region of rDNA; the second specific for the *TRI6* gene involved in trichothecene biosynthesis; and the third specific for the *FUM5* gene involved in fumonisin biosynthesis. Primer specificity was tested on genomic DNA isolated from 43 fungal species representing 14 genera, including 9 *Aspergillus* spp., 9 *Fusarium* spp., and 11 *Penicillium* spp. The detection limit for the ITS primer set was 10 pg of template DNA, while the *FUM5* and *TRI6* primer sets required at least 0.1 ng and 1 ng of template DNA, respectively. To apply the PCR technique to food analysis, we have developed a simple and rapid protocol to extract fungal DNA from cornmeal.

A one-step polymerase chain reaction protocol using soybean seed for marker assisted selection of disease resistance in soybean. M. BOLTON (1), B. Nelson (1), R. Sparks (2), and A. Santoso (2). (1) Dept. Plant Pathology; (2) Biochemistry and Molecular Biology, North Dakota State University, Fargo, ND 58105. Phytopathology 92:S8. Publication no. P-2002-0055-AMA.

Host resistance is the preferred method to control important soybean pathogens such as *Heterodera glycines* and *Phytophthora sojae*. An improved marker assisted selection protocol utilizing DNA from soybean seed that implements a one-step polymerase chain reaction (PCR) process was developed to detect resistance to these pathogens. The protocol was based on a disk-based DNA purification and amplification procedure developed for soybean leaves, but the PCR denaturing and annealing times were changed to adapt the protocol to seed DNA. The DNA was amplified using micro-satellite (simple sequence repeats [SSR]) primers Satt309 and Sat_168 to detect the *rhg 1* SCN resistance gene and Satt 159 and Satt 152 to detect the *Rps 1 Phytophthora* resistance gene. This seed DNA protocol resulted in consistent visualization of SSR markers for resistance to *H. glycines* and *P. sojae*.

Storage of fungal spores on petroleum jelly. M. G. BOOSALIS, J. E. Partridge, R. Higgins, K. Powers, and J. R. Steadman. Dept. Plant Pathology, Univ. of Nebraska. Lincoln, NE 68583-0722. Phytopathology 92:S8. Publication no. P-2002-0056-AMA.

Petroleum jelly (PJ), also labeled Vaseline, is an excellent substrate for long-term storage of fungal spores. No special equipment is needed. Spores are simply collected, placed on sterile PJ in glass petri dishes stored at 15-17°C. In ongoing experiments ascospores, chlamydoconidia and conidia of *Sclerotinia sclerotiorum*; *Ustilago triticea*; and *Alternaria* sp., respectively, maintained viability up to 6 years on PJ. Viability in storage varies with species: *Fusarium solani* macroconidia (40% viable after 4 yrs), *Cercospora maydis* conidia (less than 10% viable after 4 yrs), and urediniospores of *Puccinia graminis* f. sp. *tritici* and *Puccinia recondita* (only months). We have been unable to identify a universal factor that determines amenability to this storage method. For those isolates that are amenable, this method provides a convenient and inexpensive way to maintain viable fungal cultures.

Winter fungicide applications for improved control of black spot of rose in Alabama. K. L. Bowen. Dept. of Entomology and Plant Pathology, Auburn University, AL 36849-5409. Phytopathology 92:S8. Publication no. P-2002-0057-AMA.

Control strategies for black spot based on winter fungicide applications were evaluated on hybrid tea roses in Alabama. Four winter treatments (tetraconazole, triforine, myclobutanil, and non-treated) were arranged factorially with five foliar treatments applied through the growing season. Growing season fungicide treatments included: chlorothalonil, tetraconazole, triforine, and myclobutanil applied on 14-day intervals, and chlorothalonil applied on 7-day intervals, from 1 May through 1 September. Plants were rated every two weeks for disease severity, defoliation, vigor and flower production and averaged over the season. Disease levels in May of each year indicated that winter applications of tetraconazole reduced disease severity compared to other winter treatments, and this disease reduction persisted through the 2000 growing season. Average plant vigor did not differ among winter treatments. There were no significant interactions on disease, defoliation, vigor or flowers for the winter X growing season treatments.

Evaluation of the multiple pathogen strategy for biological control of green foxtail. S. M. BOYETCHKO (1), G. Peng (1), K. Sawchyn (1), K. Byer (1), and R. Charudattan (2). (1) Agriculture and Agri-Food Canada, Saskatoon, Saskatchewan, Canada S7N 0X2; (2) Dept. Plant Pathology, University of Florida, Gainesville, FL 32611. Phytopathology 92:S9. Publication no. P-2002-0058-AMA.

Green foxtail (*Setaria viridis*) is one of the most abundant annual grass weeds in the Canadian prairies. Bioherbicidal control using three fungi isolated from Florida (*Drechslera gigantea* and two *Exserohilum* spp.) was evaluated under different dew period durations (4, 6, and 18 h). The pathogens were foliar-applied to green foxtail plants at the 2-3 leaf stage as a mixture (1:1:1 by volume) at a rate of 10^6 spores per ml in either water or Metamucil®. Appropriate controls were included. Disease severity (Horsfall-Barratt scale) and biomass were recorded after 1 wk. Little or no disease occurred on plants treated with pathogens in water or Metamucil® with 4 and 6 h dew, but significant levels of disease occurred when 18 h dew was provided. Almost complete mortality occurred when plants were treated with the pathogens in Metamucil® at 18 h dew. There were corresponding reductions in biomass with increasing disease severity. These pathogens show promise as bioherbicides for green foxtail in Canada.

Root disease severity and root-infecting pathogens associated with dryland wheat and barley in southeastern Idaho. C. A. BRADLEY, C. A. Strausbaugh, and R. L. Forster. University of Idaho, Kimberly Research and Extension Center, Kimberly, ID 83341. Phytopathology 92:S9. Publication no. P-2002-0059-AMA.

Root diseases of dryland wheat and barley in southeastern Idaho have not been extensively studied, and no systematic survey has been conducted to document the identity or severity of root-infecting pathogens in that region. A survey was conducted in June 2001 to assess root disease severity and identify soil-borne pathogens present in 46 wheat and 23 barley fields in dryland production areas of 12 counties in southeastern Idaho. Nematode extraction assays from soil revealed that 100% of the fields surveyed had lesion nematodes (*Pratylenchus neglectus* and/or *P. thornei*), 83% had stunt nematodes (*Tylenchorhynchus* sp.), 10% had pin nematodes (*Paratylenchus* sp.), and 6% had dagger nematodes (*Xiphinema americanum*). Pathogenic fungi isolated from symptomatic roots included *Fusarium* spp., *Bipolaris sorokiniana*, *Rhizoctonia solani*, and *Gaeumannomyces graminis* var. *tritici*. Root disease severity index values ranged from 0 to 36 for *Rhizoctonia* root rot, 0 to 6 for take-all, and 0 to 56 for *Fusarium*/common root rot.

Survey of North Georgia wine grapes for Pierce's disease as related to elevation. P. M. BRANNEN (1) and C. J. Chang (2). (1) Dept. Plant Path., Univ. of Georgia, Athens, GA 30602; (2) Dept. Plant Path., Georgia Expt. Station, Griffin, GA 30223. Phytopathology 92:S9. Publication no. P-2002-0060-AMA.

Pierce's disease (PD), caused by *Xylella fastidiosa*, has limited wine grape production in the Southeast. Lower temps. negatively impact the bacterium. Increasing elev. results in lower av. temps., and wine grape producers in GA have taken this into account for site selection. A two-year vineyard survey was conducted to determine a relationship between PD and elev. in GA. Samples consisted of new shoots with at least six leaves. When no symptoms were present, 20 random samples were collected. When symptoms were present, 20 symptomatic shoots and 10 asymptomatic shoots were collected for comparison. Elev. was determined through use of GPS units. Samples were processed through ELISA; confirmation was established by plating bacteria on PW media. Epidemic PD levels were observed at 236-242 m (25-75% incidence). At 500-542 m elev., PD incidence was low (<1%), but PD was consistently observed in both survey years. No PD infections were observed at >542 m elev. The highest elev. sampled was 692 m. Climatic variation may shift the safe zone, but >400 m elev. sites may allow for wine grape production in GA.

Green manure crops and soil solarization effects on *Aphanomyces cochlioides*. J. R. BRANTNER (1), C. E. Windels (1), and A. T. Dyer (2). (1) Univ. Minn., NW Res. & Outreach Center, Crookston 56716; (2) Univ. Minn., St. Paul 55108. Phytopathology 92:S9. Publication no. P-2002-0061-AMA.

Oospores of *A. cochlioides* survive in soil for years so options are needed to reduce inoculum densities in producers' fields. An experiment was conducted in an *Aphanomyces* nursery to determine the effects of buckwheat, oilseed radish, sorghum sudan grass and a fallow control (all with and without solarization) on root rot of sugar beet and survival of *A. cochlioides* oospores in soil. Green manure crops were grown 8 wk, then incorporated into soil; soil samples were collected and indexed for root rot

(0-100 scale); and oospores were buried at 8, 15 and 23 cm depths. Half of each plot was covered with a clear polyethylene tarp for 9 wk. Buckwheat, oilseed radish and sorghum sudan grass manure crops resulted in significantly lower soil index values (63, 75, 78, respectively) than the control (97), but did not affect oospore survival. Solarization did not affect soil index values, but at the 8-cm depth, oospore survival was 16% compared to non-solarized soil at 59%. Thus, green manure crops suppress *Aphanomyces* root rot on sugar beet through mechanisms that may not be related to a reduction in oospore inoculum.

Growth of *Gaeumannomyces graminis* var. *tritici* is inhibited by *Brassica* spp. Mulches. T. S. BREEDEN (1), B. H. Ownley (1), D. R. West (2), and C. E. Sams (2). (1) Dept. Entomol. & Plant Pathol.; (2) Dept. Plant Sci. & Landscape Systems, Univ. of Tennessee, Knoxville, TN 37996. Phytopathology 92:S9. Publication no. P-2002-0062-AMA.

Gaeumannomyces graminis var. *tritici* (*Ggt*) causes take-all disease of wheat and barley. Genetic resistance is not available in commercial wheat cultivars and chemical seed treatments are not cost-effective. Mulches of *Brassica* spp. release glucosinolate compounds during decomposition and are under investigation for control of soilborne pathogens in various crops. Our objective was to determine if *Brassica* mulches inhibit mycelial growth of *Ggt*. The experiment was designed as a factorial in a RCB with 3 isolates of *Ggt* (211.1, WX, A2) and 3 mulch treatments (*Brassica juncea* cv Indian Mustard, *Brassica napus* cv Dwarf Essex Rape, no mulch control) with 3 replicates. Fresh plant tissue was harvested, macerated, and placed into glass jars, which were covered by inverted 100-mm Petri dishes containing potato dextrose agar with a plug of *Ggt*. Jars were sealed to prevent loss of volatile compounds. Colony diameters were measured for 6 days. Mycelial growth of *Ggt* isolates was inhibited significantly by *B. napus*, while no growth was recorded for *Ggt* exposed to *B. juncea*.

Cotton leaf crumple virus is the first member of a previously undiscovered phylogenetic group of New World begomoviruses. J. K. BROWN and A. M. Idris. University of Arizona. Phytopathology 92:S9. Publication no. P-2002-0063-AMA.

Cotton leaf crumple virus (CLCrV) is a bipartite, whitefly-transmitted geminivirus from the southwestern US and Sonora, Mexico that has been known to infect cotton since the 1950's. The CLCrV DNA A and DNA B components for isolates from Arizona and Sonora were cloned and the nucleotide sequences were determined. Sequence comparisons indicated that the DNA A component (GB Accession AF480940) shared the highest nucleotide sequence identities with members of the SLCV group, while the closest relatives for the CLCrV DNA-B component (GB Accession AF480941) were begomoviruses from the Caribbean, Central America, and Mexico. Parsimony and maximum likelihood analyses indicated that CLCrV is the first member of a previously undiscovered begomovirus group from the New World. The Rep binding element within the common region of CLCrV, GGAGT-CT-GGAGT, is 100% conserved for both DNA-A and DNA-B components, indicating they are cognate for the same virus. Lack of phylogenetic congruence between CLCrV DNA A and DNA B indicate that they have evolved along different paths, and/or that recombination may have occurred.

The effect of low dose ultraviolet light-C seed treatment on induced resistance in cabbage to black rot *Xanthomonas campestris* pv. *campestris*. J. E. Brown (1), T. Y. Lu (1), C. STEVENS (2), V. A. Khan (2), J. Y. Lu (2), C. L. Wilson (3), D. J. Collins (4), M. A. Wilson (5), E. C. K. Igwegbe (2), E. Chalutz (6), and S. Drobny (6). (1) Dept. Hort Auburn Univ, AL; (2) Dept. Agri Sci Tuskegee Univ, Tuskegee, AL; (3) USDA ARS NAA AFRS, Kearneysville, WV; (4) Dept. Plant Path Auburn Univ, AL; (5) Dept. Agri SE Missouri State Univ, Cape Girardeau; (6) ARO, Volcani Center, Bet Dagan Israel. Phytopathology 92:S9. Publication no. P-2002-0064-AMA.

Low doses of horsetick (*n. hormesis*) ultraviolet light-C (UV-C) seed treatments were used to elicit host resistance to black rot, and improve the quality and growth response of cabbages in greenhouse studies. Different UV-C doses (1.3 to 7.5 kJm⁻²) were tested to determine their ability to induce resistance to black rot. The optimum UV-C dose of 3.6 kJm⁻² was effective in reducing black rot and the population density of *Xanthomonas campestris* pv. *campestris* in infected cabbage leaves. Seeds treated with UV-C at 3.6 kJm⁻² produced plants with the most desirable color, highest weight, largest head diameter and delayed maturity. The effect of storage time at room temperature on disease incidence of black rot of cabbage from seeds treated with a low horsetick UV-C dose of 3.6 kJm⁻², was 90%, 40%, 60% and 60% reduction of black rot in plants from UV-C treated seeds stored for 2 days, 1, 5, and 8 months, respectively, 8 weeks after transplanting cabbage plants.

Survey of isolates of *Sclerotinia homoeocarpa* in Indiana for sensitivity to three fungicides. L. E. BROWNBACK and R. Latin. Dept. Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S10. Publication no. P-2002-0065-AMA.

Repeated fungicide applications from May through September are often required to manage dollar spot, caused by *Sclerotinia homoeocarpa*, on creeping bentgrass fairways in the Midwest. A noticeable decline in performance of systemic and local systemic fungicides has occurred at some golf courses. Marginal performance may be due to various factors, including the development of fungicide-insensitive strains of the pathogen. This research involves a survey of Indiana golf courses to determine the levels of sensitivity to three fungicides in populations of *S. homoeocarpa*. Isolates were transferred to PDA amended with seven dilutions each of iprodione, propiconazole, and thiophanate methyl. Colony diameters were measured 4 days after transfer. Sensitivity to fungicide was expressed in terms of 50% effective concentration (EC₅₀) values. Significant differences in EC₅₀ values were observed among isolates for each fungicide. Isolates that were insensitive to fungicides in the assay corresponded to those golf courses where fungicide performance appeared to decline.

Chinese wingnut rootstocks for English walnut: Resistance to *Phytophthora* and graft compatibility. G. T. BROWNE (1), J. A. Grant (2), and H. E. Becherer (1). (1) USDA-ARS, Dept. of Plant Pathology, Univ. of Calif., Davis, CA 95616; (2) UCCE, Stockton, CA 95205. Phytopathology 92:S10. Publication no. P-2002-0066-AMA.

Seedlings from seven open-pollinated selections of Chinese wingnut (CW) (*Pterocarya stenoptera*) were tested for resistance to *Phytophthora cinnamomi* (*Pcin*) and *P. citricola* (*Pcit*) and graft compatibility with English walnut (*Juglans regia*). Seedlings of Northern California black walnut (NB) (*J. hindsii*) and/or Paradox hybrid (PH) (*J. hindsii* × *regia*) served as graft-compatible rootstock standards. All CW selections were resistant to *Pcin* and *Pcit* in the greenhouse (mean root rot 0-36%), but NB and PH were susceptible to *Pcin* (89-100%) and varied with *Pcit* (24-100%). Two years after planting in an orchard infested with *Pcin*, 75-95% of English trees on CW rootstocks had survived, but only 15% of those on PH were alive. During the first 3 yr of graft compatibility trials, English scions of Hartley, Tulare, and Vina grew well on CW, PH, and NB, but Chandler and Serr grew well only on PH and NB. CW holds promise as a *Phytophthora*-resistant rootstock for some English cultivars, but long-term compatibility and yield evaluations are needed to fully determine its commercial value.

***Xanthomonas* spp. affecting golf course putting greens in the Northeast.** M. Browning, N. A. MITKOWSKI, and N. Jackson. Dept. of Plant Sciences, University of Rhode Island, Kingston, RI 02881. Phytopathology 92:S10. Publication no. P-2002-0067-AMA.

During the past few growing seasons in the Northeast and Canada, sporadic disease symptoms associated with bacterial colonization of turfgrass vascular tissues has been observed, primarily on *Poa annua*. Symptoms included yellowing of leaf blades followed by collapse of tissues, occurring in 2 cm patches. Diagnosis was determined by examination of recently yellowed central shoots for bacterial streaming from xylem vessels. In Summer 2001, isolations were made from affected *P. annua* and *Agrostis palustris* samples received in the URI Turfgrass Disease Diagnostic Laboratory. Pathogenicity tests were carried out on *A. tenuis*, *Festuca rubra*, *P. pratensis*, *Lolium perenne* and *P. annua*. Pathogenicity testing was undertaken on mature grass plants grown at high relative humidity. It was determined that at least one isolate (M-1) was highly pathogenic on *P. annua*. Temperature growth parameters were defined through the use of a thermal gradient plate. The M-1 isolate was determined to be a xanthomonad, based on fatty acid analysis. ITS regions were also sequenced to determine taxonomic status.

Association of a *Xylella fastidiosa* protein with chlorosis induction in *Chenopodium quinoa* leaves. G. BRUENING (1,2), E. B. Re (2), E. L. Civerolo (1,4), Y. M. Lee (3), P. A. Feldstein (2), and J. M. Buzayan (1). (1) Plant Pathology; (2) Center for Engineering Plants for Resistance against Pathogens; (3) Molecular Structure Facility, University of California Davis; (4) USDA-ARS, SJVASC, Parlier, CA. Phytopathology 92:S10. Publication no. P-2002-0068-AMA.

Xf cells were held at room temperature, or at 100°C for 20 min and cooled, and infiltrated into Cq leaves. Chlorosis, confined to the infiltrated area, developed within 48 hr. High-speed centrifugation collected the elicitor activity in the precipitate. Suspended precipitate, incubated with any of three proteases, lost elicitor activity. Sodium dodecyl sulfate (SDS) extraction of the precipitate retained elicitor activity and simplified the SDS-gel electrophoresis pattern of the precipitate. Mass spectrometry of a trypsin-treated gel

band corresponding to protein with apparent molecular weight about 41,000 identified amino acid sequences corresponding to about 40% of the Xf outer membrane protein mopB. Material extracted from the same band position in a parallel, unstained gel lane induced chlorosis when infiltrated into Cq leaves. Our results suggest that Xf mopB is a heat stable elicitor of chlorosis in Cq leaves.

The effects of fungicides on the yeast populations of creeping bentgrass. J. W. BUCK and L. L. Burpee. Dept. Plant Pathology, University of Georgia, Griffin, GA 30223. Phytopathology 92:S10. Publication no. P-2002-0069-AMA.

The effects of fungicides on the yeast flora of bentgrass was investigated. In spring 2001, azoxystrobin, chlorothalonil, flutolanil, and propiconazole were applied separately over six weeks to bentgrass. Total and fungicide-resistant yeast populations were assessed by dilution plating onto PDA or fungicide amended PDA. Total yeast populations in the fungicide treated plots were significantly lower than the check plots on three of four sample dates. In the fall, azoxystrobin or propiconazole were applied twice to the bentgrass over three weeks. Significantly larger total yeast populations were observed compared to resistant populations for each treatment on every sample date. Total yeast populations were significantly higher in the check plots compared to either the propiconazole or azoxystrobin treated plots on the first three of five sample dates. Yeasts not exposed to fungicides were more sensitive in vitro to chlorothalonil, propiconazole, flutolanil, and iprodione than isolates from fungicide treated bentgrass. These results suggest fungicide resistance among phylloplane yeasts is widespread and could be an important bio-control phenotype.

Specificity of grape and peach replant disorders. L. R. BULLUCK, III (1), G. T. Browne (1), S. M. Schneider (2), and T. J. Trout (2). (1) USDA ARS, Dept. Plant Pathology, Univ. of Calif., Davis, CA 95616; (2) USDA ARS WMRL, Parlier, CA 93648. Phytopathology 92:S10. Publication no. P-2002-0070-AMA.

New peach and grape plantings can suffer from replant disorder (RD), i.e., poor vigor and delayed production on sites previously devoted to the crops. Preplant fumigation can prevent RD, but some causal aspects remain unknown. We investigated specificity between grape and peach RD. Soil was sampled from RD-affected vineyards and orchards (3 each) near Parlier, CA, fumigated (F) (67:33 methyl bromide:chloropicrin, 0.37g/l soil) or nonfumigated (NF), and planted with peach or grape in a greenhouse. Responses were assessed 2 mo after planting. For peach, less root cortex necrosis and greater root weight resulted in F orchard and vineyard soils and NF vineyard soil than in NF orchard soil (fumigation × crop history effect [F × C] $P < 0.003$). For grape, less root necrosis occurred in F vineyard and orchard soils than in NF vineyard soil, but amount of root necrosis in NF orchard soil was intermediate (F × C $P = 0.014$); root weight was affected little by fumigation or crop history ($P > 0.08$). Peach RD may be more severe after peach than after grape on NF sites. Crop history and fumigation effects are less clear for grape.

Relationship between charcoal rot, the stay-green trait, and irrigation in grain sorghum. M. G. BURGESS (1), C. M. Rush (1), G. Piccinni (2), K. Steddom (1), and F. Workneh (1). (1) Texas Agricultural Experiment Station, Bushland, TX; (2) Texas A&M Research and Extension Center, Uvalde, TX. Phytopathology 92:S10. Publication no. P-2002-0071-AMA.

Charcoal rot of grain sorghum, caused by *Macrophomina phaseolina*, is impacted by drought stress. The stay-green gene confers tolerance to drought stress in many commercial sorghum cultivars. A field study was conducted in 2001 in Uvalde and Bushland, Texas, by planting five stay-green cultivars and one non stay-green. Irrigation treatments included 100% Potential Evapotranspiration (PET), 75% PET, 50% PET, and a limited treatment in which 50% PET was applied until anthesis and 100% PET during grain fill. Plants were inoculated at heading using infested toothpicks. Stalks were split, and lesion length and head weight were measured. Inoculated plants exhibited lesions that were 54% longer than in control plants. Lesions in the stay-green lines were 27% shorter than in the non stay-green line and there were significant differences between cultivars. Results suggest that charcoal rot severity can be affected by irrigation and presence of the stay-green gene.

A new locus in soybean for resistance to *Phytophthora sojae*. K. D. BURNHAM (1), A. E. Dorrance (2), D. M. Francis (1), R. J. Fioritto (1), and S. K. St. Martin (1). (1) The Ohio State University/OARDC Dep. of Hort. and Crop Science; (2) The Ohio State University/OARDC Dep. of Plant Pathology. Phytopathology 92:S10. Publication no. P-2002-0072-AMA.

Soybean diseases can result in the loss of thousands of metric tons each year. *Phytophthora* root and stem rot which is caused by *Phytophthora sojae* is the second leading cause of yield loss in the United States. Currently, seven loci for resistance with thirteen genes or alleles have been identified in soybean which are effective against different pathotypes of the pathogen. However, populations of *P. sojae* exist in many soybean production regions that cause disease on plants with many if not all of these genes. Consequently, the need for new resistance loci is great. A number of plant introductions (PIs) from South Korea were identified in an earlier study that may contain a novel resistance locus. The objective of this project was to determine if a new locus for resistance to *Phytophthora sojae* was present and to map its location. One of the South Korean PIs from this previous study, PI399073, was crossed with Williams and S 19-90. Using traditional crossing populations and disease assays combined with molecular markers, SSR (single sequence repeat), RFLP (restriction fragment length polymorphism), and isozyme markers a new allele was identified. The locus, *Rps8*, has been mapped to MLG (major linkage group) A2 in two different crosses with PI399073. This is the first locus for *Phytophthora* resistance that has been identified on this MLG group.

Quantitative trait loci for partial resistance to *Phytophthora sojae* in soybeans. K. BURNHAM (1), A. E. Dorrance (2), T. VanToai (3), and S. K. St. Martin (4). (1) Dept. of Horticulture and Crop Science, Ohio State University, Wooster, 44691; (2) Dept. of Plant Pathology, Ohio State University, Wooster, 44691; (3) USDA-ARS, Soil Drainage Research Unit; and (4) Dept. of Horticulture and Crop Science, Ohio State Univ. Phytopathology 92:S11. Publication no. P-2002-0073-AMA.

Partial resistance to *P. sojae* in soybeans is expressed as a reduced level of root rot and is effective against all populations of the pathogen. Three soybean populations, Conrad × Sloan, Conrad × Harosoy, and Conrad × Williams, were evaluated for lesion growth rate and with SSR markers to identify putative quantitative trait loci. The three populations segregated for lesion growth rate as measured by root inoculations. Family mean heritability estimates for the three populations at the F4:6 generation were 0.62, 0.87 and 0.57. Approximately 70, 45, and 60 SSR markers were polymorphic in each of the three populations, respectively. Preliminary analysis indicate that allele(s) in Conrad, at a locus on soybean linkage group F, confer partial resistance to *P. sojae*.

Characterization of *Phytophthora* species from an irrigation water recycling system at a container nursery in southwestern Virginia. E. A. BUSH (1), C. Hong (1,2), and E. L. Stromberg (1). (1) Dept. PPWS, VPI & SU, Blacksburg, VA 24061; (2) VPI & SU, Hampton Roads AREC, Virginia Beach, VA 23455. Phytopathology 92:S11. Publication no. P-2002-0074-AMA.

Containing and recycling nursery effluent is a relatively simple method to avoid discharge of pollutants associated with fertilizer and pesticide applications; however, plant pathogens may be spread through irrigation water. *Phytophthora* in recycled irrigation and effluent water were characterized at a container nursery in southwestern Virginia, using filtering and baiting with two selective media (PARP and PARPH). *Pythium* spp. were recovered more frequently and in greater numbers than *Phytophthora* spp. *P. capsici*, *P. citricola*, *P. citrophthora*, *P. cryptogea*, *P. drechsleri*, and *P. nicotianae* were recovered in filtering assays. *P. cryptogea* and *P. drechsleri* were the only *Phytophthora* spp. recovered from baits placed on the surface of the irrigation reservoir, whereas *P. cactorum*, *P. capsici*, *P. citrophthora*, and *P. citricola* were recovered from baits placed at depths. Hymexazol-amended medium was found to have limitations in recovery of *Phytophthora* spp.

Light-dependent bleaching of detached barley leaf tissue by deoxynivalenol. W. R. BUSHNELL, T. M. Seeland, and D. E. Krueger. USDA-ARS Cereal Disease Laboratory and Dept. Plant Pathology, University of Minnesota, St. Paul, MN 55108. Phytopathology 92:S11. Publication no. P-2002-0075-AMA.

The mycotoxin, deoxynivalenol (DON) accumulates in wheat and barley heads infected with *Fusarium graminearum*. To assess the effects of DON on green plant tissues, we partially stripped the abaxial epidermis from detached Robust barley leaf segments (1 cm long) and floated them with stripped mesophyll in contact with aqueous DON solutions. In light (150-450 micro mol m²sec⁻¹), DON (30-200 ppm) caused complete loss of pigmentation in stripped mesophyll within 2-4 days. The greater the light intensity or the DON concentration, the more rapid was the bleaching effect. The bleached tissue was neither water-soaked nor collapsed, but chloroplasts and other cytoplasmic organelles became disorganized as observed by transmission electron microscopy. In darkness, DON at 30-100 ppm had an opposite

effect, preserving chloroplasts, causing leaf segments to remain dark green for 6-7 days and preventing the yellowing that occurred in segments floated on water. Because of the opposing effects of DON in light and darkness, assays of plants for sensitivity to DON should include trials under both conditions.

Insensitivity to mefenoxam in *Phytophthora capsici* isolates on pepper and squash in North Carolina. A. C. CAFÉ-FILHO and J. B. Ristaino. North Carolina State University. Phytopathology 92:S11. Publication no. P-2002-0076-AMA.

Mefenoxam is a popular fungicide in NC, and in 2001, incidence of *Phytophthora* blight on pepper (*Capsicum annuum*) and squash (*Cucurbita pepo*) was high. A 1997 study showed 59% of *Phytophthora capsici* isolates were resistant to mefenoxam, and 10% were intermediate in sensitivity. To assess the mefenoxam sensitivity of *P. capsici* populations now prevailing in NC, 75 isolates were collected in the 2001 season from 5 pepper and one squash field. The frequency of resistant isolates in 2001 was 63% and comparable to 1997 levels. Percentage growth of resistant isolates on amended media was >80% and >100% of the non-amended control at the 100 ppm and at the 5 ppm levels, respectively. Sensitive isolates had no growth at 5 ppm and no intermediate isolates were found. All isolates from 3 fields, including the squash field, were resistant. Other fields had either mixes of sensitive and resistant isolates or only sensitive isolates. Prevalence of resistance to mefenoxam in NC has remained stable since 1997. This is the first report of a mefenoxam resistant population of *P. capsici* on squash in NC.

Use of nitrate nonutilizing mutants to examine vegetative compatibility groups in *Cercospora kikuchii*. G. CAI and R. W. Schneider. Dept. Plant Pathology and Crop Physiology, LSU Agricultural Center, Baton Rouge, LA 70803. Phytopathology 92:S11. Publication no. P-2002-0077-AMA.

Potato dextrose agar with 1.5, 3.0, and 5.0 percent potassium chlorate (KPS medium) was used to generate nitrate nonutilizing (*nit*) mutants from *Cercospora kikuchii*. *C. kikuchii* colonies were olive green with concentric rings and sparse aerial mycelium on KPS medium. *Nit* mutants appeared in about 3 weeks as sectors with dense aerial mycelium that was occasionally reddish brown. In general, KPS medium with 3.0 percent potassium chlorate produced most *nit* mutants. One *nit1* and one *NitM* were identified from each of the six *C. kikuchii* strains and paired with each other in all possible combinations. Five strains were self-compatible, but they were not compatible with each other. We are currently using this method to examine vegetative compatibility groups in *C. kikuchii* from a large worldwide collection.

Characterization of a nitric oxide synthase protein in the *Streptomyces turgidiscabies* pathogenicity island. K. D. CAMERON (1), B. R. Crane (2), and R. Loria (1). (1) Dept. Plant Pathology; (2) Dept. Chemistry, Cornell University, Ithaca, NY 14853. Phytopathology 92:S11. Publication no. P-2002-0078-AMA.

Genes coding for nitric oxide synthases (NOS) have been identified throughout the animal kingdom. Eukaryotic NOSs catalyze the oxidation of L-arginine to L-citrulline and nitric oxide, with *N*-hydroxy-L-arginine formed as an enzyme-bound intermediate. In mammalian systems, nitric oxide is involved in normal cell function and defense mechanisms. Although NOS proteins have been identified in Gram-positive bacteria, only one, (deiNOS from *Deinococcus radiodurans*) has been characterized. Here we identify a NOS gene from *S. turgidiscabies* that codes for strNOS, a protein with homology to a NOS from *Bacillus halodurans* and to iNOS_{OX}, an inducible murine NOS. Unlike NOS-like enzymes from other Gram-positive bacteria, the strNOS protein may contain the N-terminal beta-hairpin hook region which is important in the mammalian enzymes for dimerization, H₄B binding and catalytic activity. The gene is conserved between at least three pathogenic *Streptomyces* species suggesting its importance in pathogenicity in agricultural systems.

Reactions of *Rp1-D*-resistant and non-*Rp* sweet corn to *Rp1-D*-virulent *Puccinia sorghi*. M. A. CAMPANA, J. K. Pataky, and P. M. Michener. Dept. of Crop Sciences, Univ. of Illinois, Urbana, IL. Phytopathology 92:S11. Publication no. P-2002-0079-AMA.

Reactions of *Rp1-D*-resistant and non-*Rp* sweet corn hybrids to *Rp1-D*-virulent and avirulent *P. sorghi* were compared in three experiments. Rust severity for three replicates of single rows of 176 non-*Rp* hybrids inoculated with *Rp1-D*-virulent isolates was similar to rust severity on these hybrids inoculated with avirulent *P. sorghi* in a separate trial (r = 0.90). When 137

Rp1-D-resistant and 176 non-*Rp* hybrids were inoculated with *Rp1-D*-virulent *P. sorghi*, distributions were similar for groups of *Rp1-D*-resistant and non-*Rp* hybrids. Severity ranged from 0 to 60% and averaged 33% for both groups. When five replicates of 40 pairs of *Rp1-D*-resistant and non-*Rp* versions of the same hybrid were inoculated with *Rp1-D*-virulent *P. sorghi*, rust severity ranged from 15% to 46% among hybrid pairs, but severity did not differ between *Rp1-D* and non-*Rp* versions of hybrids. The slope of the linear regression of rust severity for *Rp1-D*-resistant vs. non-*Rp* hybrids was 0.95 ($r^2 = 0.96$). Thus, *Rp1-D*-virulent and avirulent isolates caused the same amount of rust on non-*Rp* hybrids, and the *Rp1-D* gene provided no resistance against *Rp1-D*-virulent isolates.

In-furrow treatments of Abound 2SC fail to enhance disease control or increase yield of peanut. H. L. CAMPBELL, A. K. Hagan, and K. L. Bowen. Dept. of Entomology and Plant Pathology, Auburn University. Phytopathology 92:S12. Publication no. P-2002-0080-AMA.

In 2000 and 2001, Abound 2SC was evaluated in field trials for control of early and late leaf spot, caused by *Cercospora arachidicola* and *Cercosporidium personatum*, respectively, and for southern stem rot (SSR) caused by *Sclerotium rolfsii*. Abound 2SC was applied in-furrow at planting at rates ranging from 0.85 g to 1.28 g ai/1000 row m followed by foliar applications at 224 or 336 g ai/ha of the same fungicide at either 30, 60, or 90 days after planting. Chlorothalonil was applied for leaf spot control at 1262 g ai/ha or was tank-mixed with propiconazole at 841 and 63 g ai/ha, respectively. Efficacy of Abound 2SC against SSR was equal to or greater than Folicur 3.6F and significantly greater than chlorothalonil alone. However, in-furrow applications of Abound 2SC failed to enhance control of leaf spot diseases or SSR. Yield gains with foliar applications of Abound 2SC ranged from 10-22% in 2000 to 13-30% in 2001 when compared to full season chlorothalonil alone. However, Abound 2SC applied in-furrow did not significantly increase yields in plots treated post plant with the same fungicide or chlorothalonil.

Control of leaf spot diseases and southern stem rot in Alabama using Moncut 70DF. H. L. CAMPBELL, A. K. Hagan, and K. L. Bowen. Dept. of Entomology and Plant Pathology, Auburn University. Phytopathology 92:S12. Publication no. P-2002-0081-AMA.

In 2000 and 2001, Moncut 70DF, a new formulation of flutolanil, was compared in field trials with fungicides currently registered for the control of southern stem rot (SSR) caused by *Sclerotium rolfsii* on 'Georgia Green' peanut. In 2000, peanuts treated with Moncut 70DF at 840 g ai/ha suffered less SSR damage than those treated with chlorothalonil alone. Yield gains were better than those obtained with Folicur 3.6F at 226 g ai/ha and comparable to those provided by Abound 2SC at 336 g ai/ha. In 2001, Moncut 70DF was tested at 421 g ai/ha and 840 g ai/ha for control of SSR. At 421 g ai/ha and 840 g ai/ha rates of Moncut 70DF, SSR damage was lower than that observed in plots treated with chlorothalonil alone. Control of SSR with Moncut 70DF at 840 g ai/ha was equal to or superior to that observed with Abound 2SC and Folicur 3.6F, respectively. Yield gains were 18% and 23% higher at 421 and 840 g ai/ha, respectively, than those obtained with chlorothalonil alone and were comparable to those noted for the Folicur 3.6F and Abound 2SC-treated peanuts.

Use of *Melaleuca alternifolia* oil for plant disease control. J. M. CAOLO-TANSKI (1), L. E. Hanson (2), A. L. Hill (2), and J. P. Hill (1). (1) Dept. BSPM, Colorado State Univ., Ft. Collins, CO 80523; (2) USDA-ARS, Ft. Collins, CO 80526. Phytopathology 92:S12. Publication no. P-2002-0082-AMA.

The essential oil from *Melaleuca alternifolia* has been used for pharmaceutical and household products and as an antiseptic treatment for human ailments. Growing public concern over the use of synthetic pesticides emphasizes the need for alternative treatments. Our work tests the effectiveness of melaleuca oil to control several plant pathogens. The oil's anti-microbial activity was tested *in vitro* against 7 fungal and 2 oomycetous plant pathogens. Greenhouse studies were conducted to determine the efficacy of melaleuca oil as a seed treatment in the control of *Rhizoctonia solani* (AG-4) on cotton, and *Cochliobolus sativum* and *Fusarium graminearum* on wheat. Experiments in the field evaluated the oil's potential to reduce disease incidence and severity from foliar pathogens such as *Alternaria solani* on potato and *Cercospora beticola* on sugarbeets, and seed pathogens such as *C. sativum* and *F. graminearum* on wheat. Growth *in vitro* of all organisms was significantly decreased, but disease control in the field was variable. There may be potential to use *M. alternifolia* oil for plant pathogen control.

Molecular variability and pathogenesis of *Phoma medicaginis* var. *medicaginis* isolates from Minnesota. C. CASTELL (1), D. Samac (1,2),

and L. J. Szabo (1,3). (1) Plant Pathology Dept. University of Minnesota, St Paul, MN 55108; (1,2) USDA-ARS-Plant Science Research; (1,3) USDA-ARS Cereal Disease Lab. Phytopathology 92:S12. Publication no. P-2002-0083-AMA.

Phoma medicaginis var. *medicaginis*, the causal agent of spring black stem and leaf spot on alfalfa, causes serious losses in yield and quality worldwide. Alfalfa cultivars usually exhibit low to moderate levels of resistance. This study was undertaken to determine the molecular and pathogenic variability of *P. m.* var. *medicaginis* isolates from different environments and plant organs. A collection of 71 single spore isolates from northern and southern areas of Minnesota was used. Amplified Fragment Length Polymorphism (AFLP) analysis allowed separation of the majority of the isolates into two groups, which correlated with the northern and southern locations. Currently, additional isolates and primer-pair combinations are being evaluated in order to detect if there is organ specialization. In addition, isolates from different locations and organs are being tested to determine if there are differences in pathogenicity. This research will provide information on the population structure *Phoma medicaginis* var. *medicaginis* on alfalfa.

Phenotypic recurrent selection for improving partial resistance to crown rust (*Puccinia coronata* Corda) in oat (*Avena sativa* L.) populations. C. CASTELL (1) and D. Stuthman (2). (1) Plant Pathology Dept. Univ. of MN, St Paul, MN 55108; (2) Agronomy and Plant Genetics. Phytopathology 92:S12. Publication no. P-2002-0084-AMA.

Crown rust is the most devastating disease in oat. Partial resistance (PR), an incomplete type of resistance associated with slow-rusting mechanisms has been proposed as an alternative to major resistant genes. The objectives of this research were to conduct phenotypic recurrent selection (RS) to increase the level of PR to crown rust, test the efficiency of the RS procedure to accumulate genes for PR, and detect slow-rusting mechanisms related with that type of resistance. Visual estimations of disease severity were done on the flag leaf and flag leaf¹. One plant was selected within each of the 21 of the 63 F2 populations that had on average a lower level of disease severity. These 21 plants became the parents for the cycle two of RS. C2r (Cycle two rust) parents had, on average, the smallest diseased leaf area, and the lowest number of pustules compared to the original RS parents, and the cycle zero parents for PR. Infection frequency may be the slow-rusting mechanism involved. Seedling tests indicated that partial resistance is acting in the adult plant stage.

***Phomopsis vaccinii* as a probable causal agent of upright dieback of cranberry.** N. J. CATLIN and F. L. Caruso. University of Massachusetts, Cranberry Station, E. Wareham, MA 02538. Phytopathology 92:S12. Publication no. P-2002-0085-AMA.

Upright dieback of cranberry (*Vaccinium macrocarpon*) can be a persistent problem in cranberry beds nationwide. Though seldom devastating, 25% of uprights are affected in severe cases. Several fungi are routinely cultured from symptomatic uprights, but the primary causal agent(s) remain uncertain. Koch's Postulates have not yet been completed for *Phomopsis vaccinii*, but this fungus has been assumed to be involved because it is frequently recovered from diseased uprights and it has a proven role in twig blight and canker diseases of blueberry (*Vaccinium corymbosum*). Previously, various phenological stages of the host were inoculated using several techniques without success. To eliminate problems associated with infested plant stock, possible latent infections, and cross contamination, tissue cultured plants were used for inoculation trials. Agar plugs of mycelia of two *P. vaccinii* isolates or a sterile agar control were placed on leaves of either wounded or non-wounded plants. Wounds were produced by either cutting the stem or piercing a leaf. Dieback symptoms were observed 6-8 days after inoculation. Symptom development will be discussed.

How clonal are populations of *Xanthomonas axonopodis* pv. *citri* in Brazil? P. C. CERESINI (1), Z. R. Campos (1), J. Belasque, Jr. (2), J. A. Ferro (3), and E. G. M. Lemos (3). (1) Dept. Fitossanidade, UNESP - Universidade Estadual Paulista, Ilha Solteira, SP 15385-000; (2) Fundecitrus, Araraquara, SP 14807-040; (3) Dept. Tecnologia, UNESP, Jaboticabal, SP 14884-900, Brazil. Phytopathology 92:S12. Publication no. P-2002-0086-AMA.

Xanthomonas axonopodis pv. *citri* (*Xac*) is a major pathogen infecting citrus in Brazil and worldwide, causing the citrus canker disease. The pathogen was reportedly introduced in Brazil in the 50's. Since then, the ecology, pathology and genetic diversity of *Xac* on citrus have been extensively studied. Despite these studies, the extent and magnitude of recombination is still unclear in Brazilian populations of *Xac*. The role of recombination would be potentially an important aspect of *Xac* life history contributing to

the genetic diversity and structure of field populations. In this study we tested the null hypotheses that the *Xac* population has a non-randomly mating (clonal) structure in Brazil. To address this hypothesis, we used a strategy for detection of individual genotypes of *Xac* by multilocus sequencing typing. Preliminary data supported clonality as all fifty-five *Xac* isolates from São Paulo, Santa Catarina and Rio Grande do Sul were of a single haplotype.

Efficacy of disinfectants in control of pathogens of greenhouse vegetables. R. F. CERKAUSKAS (1), L. W. Stobbs (2), R. Brown (1), and L. Van Driel (2). (1) Agriculture & Agri-Food Canada, Harrow, N0R 1G0; (2) Vineland Station, L0R 2E0, Ontario. Phytopathology 92:S13. Publication no. P-2002-0087-AMA.

The efficacy of several disinfectants in control of bacterial, fungal and viral pathogens of greenhouse vegetables was determined. Chemproicide (CP, Pace Chemicals Ltd., didecylidimethyl ammonium chloride, 7.5% w/w), Virkon (VK, Vetoquinol Canada Inc., potassium monopersulfate 21.4% w/w), and Virucidal Extra (VE, DiversyLever Canada, potassium hydrogen peroxy-monosulphate, 40-70% w/w) were effective at 0.4%, 0.5%, and 0.25%, respectively, in in-vitro tests using isolates of *Agrobacterium*, *Clavibacter*, *Erwinia*, *Pseudomonas*, and *Xanthomonas*. VK controlled cucumber necrosis (CNV), cucumber mosaic (CMV), tomato mosaic (TomMV), and turnip mosaic (TuMV) at 1%, and potato virus Y (PVY) and tobacco mosaic (TMV) but not alfalfa mosaic (AMV) at 2%. CP failed to control all these viruses at 3% except CMV and PepMV. VE controlled CNV, CMV, TomMV, TuMV, and PVY at 2%. CP, VK and VE were effective at 0.4%, 0.5% and 0.5%, respectively, in control of *Botrytis cinerea*, *Colletotrichum orbiculare*, *Didymella bryoniae*, *Fusarium solani*, and other sp. of *Fusarium*. Other results will be presented.

Genetic variability of dollar spot resistance in clones, cultivars, and species of bentgrass using six VCG isolates. N. CHAKRABORTY (1), J. Curley (1), J. Powell (2), and G. Jung (1). (1) Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706; (2) Dept. of Plant Pathology, University of Minnesota, St. Paul, MN 55108. Phytopathology 92:S13. Publication no. P-2002-0088-AMA.

Dollar spot caused by *Sclerotinia homeocarpa* is the most prevalent and economically important turf disease in North America. Due to fungicide resistance and environmental concerns, host resistance has become important. Previous reports indicate differences among bentgrass cultivars in their susceptibility to dollar spot. Two inoculation experiments were performed in the greenhouse, to detect genetic variation at the species, cultivar, and clone level, and to identify relatively tolerant clones. These results can be used to breed broadly resistant cultivars by pyramiding resistance genes from different sources via comparative QTL mapping. Seventy-nine clones of ten cultivars of the creeping, colonial, dryland, and velvet bentgrass species were grown from single seed in the greenhouse. The dollar spot isolate used for inoculation was MNI. Disease was scored as a percentage of diseased tissue area. Significant variation of disease response among clones within cultivars, cultivars within species and between species was noted in both experiments. In general, dryland, colonial, and velvet were more resistant than creeping. The disease response noted using the MNI isolate will be tested with seven other isolates belonging to the six VCGs identified by Dr. Jon Powell. These results will strengthen the findings that there is a significant variation among bentgrass genotypes in their resistance to dollar spot, and will detect any race specific interactions.

Polycyclic infection by *Colletotrichum gloeosporioides* at high CO₂ selects for increased aggressiveness. S. CHAKRABORTY (1) and S. Datta (2). (1) CSIRO Plant Industry, University of Queensland, Australia 4071; (2) Dept. Statistics, University of Georgia, Athens, GA 30602. Phytopathology 92:S13. Publication no. P-2002-0089-AMA.

Atmospheric CO₂ concentration has increased by 31% since pre-industrial times and models project concentrations of 540 to 970 ppm by 2100. Doubling of CO₂ increases biomass, yield and canopy size of crops by 30%, but this estimate is from studies that ignore potential impacts of pest, disease and weeds. At elevated CO₂ many necrotrophic fungal pathogens are slow to invade host tissue but produce increased amounts of spores due to increased fecundity. To study pathogen evolution on a resistant and a susceptible host, we have monitored aggressiveness of 2 *C. gloeosporioides* isolates on 2 *Stylosanthes scabra* varieties over 25 sequential infection cycles in a controlled environment. Aggressiveness increases at 350ppm CO₂ over all infection cycles, while at 700 ppm it initially declines and then increases to levels higher than at ambient CO₂. Spore production increases with each cycle at elevated but not at ambient CO₂. Increased fecundity and aggressiveness in enlarged host canopy provide opportunities for rapid pathogen evolution.

Minimum inhibitory concentrations of terpene on growth of *Xylella fastidiosa* strains. C. J. CHANG (1) and L. Franklin (2). (1) Dept. Plant Pathology, Univ. of Georgia, Griffin, GA 30223; (2) Alpha Gamma Research Inc., 5170 Chemin De Vie, Atlanta, GA 30342. Phytopathology 92:S13. Publication no. P-2002-0090-AMA.

MICs of terpene AC-2 on growth of 11 strains of *X. fastidiosa* representing 5 grape strains, 2 sycamore strains, and 1 strain each of peach, plum, pecan, and oleander were studied. Concentrations of 500, 250, and 125 ppm of terpene AC-2 were prepared in sterile water. *X. fastidiosa* was grown in PW agar at 30 C. An aliquot of 0.5 mL of each strain was added to 0.5 mL of each terpene solution or sterile water in a sterile test tube. The final concentrations of terpene AC-2 were 250, 125, and 62.5 ppm, respectively. This treated cell suspension was incubated for 24 hrs at 30°C before the color-changing units were determined by a 10-fold serial dilution in fresh PW to 10⁻⁹. All culture tubes were incubated for 20 days before final readings were taken. The MICs, defined as the lowest concentrations in which no cells survived the treatment, were 125 ppm for 4 grape strains, 2 sycamore strains and 1 peach strain, and 62.5 ppm for strains from grape, plum, pecan, and oleander. Terpene AC-2 may be used as a control agent for diseases induced by *X. fastidiosa*.

Screening for sycamores that may be tolerant to leaf scorch disease caused by *Xylella fastidiosa*. C. J. CHANG (1), T. D. Leininger (2), and K. O. Britton (3). (1) Dept. Plant Pathology, University of Georgia, Griffin, GA 30223; (2) USDA Forest Service, Stoneville, MS 38776; (3) USDA Forest Service, Athens, GA 30602. Phytopathology 92:S13. Publication no. P-2002-0091-AMA.

A total of 3264 two-year-old seedlings representing nine families was injected with a cell suspension of *X. fastidiosa* (1840) or with PW broth (1424) in May 2000. Two hundred samples were collected in October 2000 for the detection of *X. fastidiosa* using the ELISA Kit manufactured by Agdia, Inc. In 2001, the same tests were performed for two hundred and six samples collected monthly from the same marked trees from June to October. Results from Oct 2000 tests showed infection rates in percent of 27, 32, 16, 18, 22, 16, 22, 29, and 30 for family 295522, 27012, Filler, 296511, 295582, 295201, 295032, WV398, and 11006 respectively, as compared to 35, 60, 57, 29, 37, 26, 37, 50, and 50 for 2001 results. Data suggest various degrees of tolerance among sycamore families. Families 296511 and 295201 were most tolerant whereas 27012 and Filler most susceptible to the infection of *X. fastidiosa*. The monthly infection rates were 10, 15, 23, 42, and 43% for June, July, August, September, and October, respectively.

Efficacy of fungicides on the control of snow molds in turfgrass. T. H. CHANG, J. Gregos, and G. Jung. Dept. of Plant Pathology, Univ. of Wisconsin, Madison, WI 53706. Phytopathology 92:S13. Publication no. P-2002-0092-AMA.

Snow mold, caused by two Typhula species, *T. ishikariensis* and *T. incarnata*, is one of the major turfgrass diseases in Wisconsin. Three varieties of *T. ishikariensis* species were found in Wisconsin. The most effective method for controlling snow molds is chemical treatment. To evaluate the inhibitory effect of four fungicides, the mycelium growth of Typhula isolates was measured on potato dextrose agar (PDA) amended with the fungicides at three concentrations. Propiconizol was the most effective in inhibiting the radial growth of the tested isolates at 1.0 ug a.i./ml of PDA. Ranges of percentage in the growth reduction among the isolates for individual fungicide is as follows: PCNB (0-19%), Propiconizol (73-86%), Azoxystrobin (20-69%), and Chlorothalonil (13-69%). In addition, eight fungicides were applied to two experimental field plots located in northern and central regions of Wisconsin during late fall in 1999 and 2000. The plots were evaluated for disease symptoms after snowmelt in the spring of the following year. The percentages of disease severity averaged for two years in untreated plots were 55.3% and 63.8% in the central and northern regions, respectively. The average percentage range of control value for all fungicides were 41.8 to 78.8% in both regions.

Quantitative and rapid on-farm testing of compost stability. C. M. CHANGA, P. Wang, F. C. Michel, Jr., M. E. Watson, and H. A. J. Hoitink. Dept. of Plant Pathology, Ohio Agricultural Research and Development Center, The Ohio State University, Wooster, OH 44691. Phytopathology 92:S13. Publication no. P-2002-0093-AMA.

Compost stability critically affects disease suppression. The standard test for assessing stability by CO₂ respirometry requires three days. A rapid, 4-hr test kit (Solvita kit) was evaluated for reliability of measuring CO₂ evolution. The kit utilizes a scale of 1 to 8 (1 = fresh; 8 = highly stable compost). Changes in stability during composting of dairy manure with wheat straw,

dairy manure with sawdust and swine manure with sawdust were monitored. The three composts attained acceptable mean (n=6) stability levels of 0.63, 0.76 and 0.31 mg CO₂-C g⁻¹ VS day⁻¹, respectively. Their corresponding Solvita values were 6.8, 6.5 and 7.0. Regression analysis revealed highly significant (P < 0.0001) negative linear relationships between the two methods. Coefficients of determination for the three composts were 0.79, 0.69 and 0.82, respectively. We conclude that the test kit is suitable for on-farm determination of compost stability.

Maize chlorotic dwarf virus genome sequence and polyprotein cleavage. R. CHAOUCH (1,2), K. Willie (2), M. G. Redinbaugh (2), and S. A. Hogenhout (1). (1) Department of Entomology, Ohio State University; (2) USDA-ARS Department of Plant Pathology, OARDC, Wooster, OH 44691. Phytopathology 92:S14. Publication no. P-2002-0094-AMA.

The genomic sequence of the severe Ohio Maize chlorotic dwarf virus isolate (MCDV-S), was determined from overlapping cDNA clones. The ~400 kDa polyprotein encoded by the viral genome is post-translationally cleaved into several functional proteins. Whereas the three MCDV capsid proteins, a cysteine protease and RNA-dependent RNA polymerase were identified on the polyprotein sequence, functional proteins from three separate regions of the virus polyprotein have not been identified and characterized. cDNAs for these polyprotein regions were cloned into the pTrcHis expression vector. His-tagged fusion proteins of 75.5 kDa (MCDV1), 34.6 kDa (MCDV2) and 66.4 kDa (MCDV3) proteins were produced in *E. coli*. Antisera raised to MCDV1, 2, and 3 were used to determine the sizes of functional proteins by Western blot analysis and to immunoprecipitate the functional proteins for subsequent N-terminal sequencing. Preliminary data show that MCDV1 antisera detects 50 and 35 kDa proteins in both infected plants and virion, but not in the healthy plants.

Heterobasidion annosum associated with mortality of Christmas trees in the Pacific Northwest. G. A. CHASTAGNER (1), I. M. Thomsen (2), J. Hudak (1), and K. L. Riley (1). (1) Washington State University, Puyallup, WA 98371; (2) Danish Forest and Landscape Res. Institute, Hoersholm, DK. Phytopathology 92:S14. Publication no. P-2002-0095-AMA.

Historically, Annosus root rot (*Heterobasidion annosum*) has seldom been a problem in Pacific Northwest Christmas tree plantations. During the past three years, the prevalence of this disease has increased significantly in 2nd and 3rd rotation noble and Fraser fir plantings. During 2001, 19 field plots were established to obtain a better understanding of the association of this disease with tree mortality. A total of 23,337 trees were examined for above ground symptoms such as branch flagging, wilting, and death. The percentage of symptomatic trees in these plots ranged from 2.5 to 13.6%. If missing and replanted trees were included, the maximum percentage reached 29.2%. *H. annosum* was associated with 78% of the dead and dying trees. Annosus root rot was detected on noble, Fraser, and grand fir, as well as Douglas-fir. Noble and Fraser fir appears to be very susceptible to this disease. These field plots will be monitored during the next several of years to determine the level of mortality prior to harvest.

Root diseases associated with dead and dying noble fir Christmas trees in the Pacific Northwest. G. A. CHASTAGNER and J. Hudak. Washington State University, Puyallup, WA 98371. Phytopathology 92:S14. Publication no. P-2002-0096-AMA.

As noble fir Christmas tree production has increased in the Pacific Northwest, the incidence of root related problems in 2nd and 3rd rotation plantations has significantly increased. A survey of 67 noble fir plantations was conducted during the 2000 growing season to determine the causes of root problems and to better understand the importance of certain root diseases in western Oregon and Washington Christmas tree plantations. Plantations were selected based on responses by growers to a written survey. Sampling and laboratory examination indicated that three root diseases accounted for about 83% of the dead and dying trees in these plantations. Phytophthora root rot and stem canker was the most common disease, occurring in 37.7% of the sites and 44.7% of the sampled trees. Annosus root rot (*Heterobasidion annosum*) was found at 23.9% of the sites and on 23.7% of the trees. Armillaria was found at 18% of the sites and on 14.7% of the trees. Root disease incidence at the different sites ranged from <1 to 30%. The cause of the problems on the remaining trees appeared to be related to improper planting and environmental stress.

An evolutionary perspective of xylella diseases in grapevine, citrus, and mulberry. J. CHEN (1), J. S. Hartung (2), C. J. Chang (3), and A. K. Vidaver (4). (1) Div. Agriculture, Florida A&M University, Tallahassee, FL 32307; (2) Fruit Lab, USDA-ARS, Beltsville, MD 20705; (3) Dept. Plant Pathology,

University of Georgia, Griffin, GA 30223; (4) Dept. Plant Pathology, University of Nebraska, Lincoln, NE 68583. Phytopathology 92:S14. Publication no. P-2002-0097-AMA.

Xylella fastidiosa causes diseases on many economically important plants. An understanding of how xylella diseases originated and evolved is important for disease prevention and management. We evaluated the phylogenetic relationships of *X. fastidiosa* strains from citrus, grapevine, and mulberry through analyses of RAPDs and 16S rDNA genes. RAPD analysis detected three clonal groups of strains that cause Pierce's disease (PD) of grapevine, citrus variegated chlorosis (CVC), and mulberry leaf scorch (MLS). Analysis of 16S rDNA sequences also identified the PD and CVC groups, but with a less stable evolutionary tree, and included MLS strains in the PD group. The Asiatic origins of the major commercial grape and citrus cultivars suggest the recent evolution of both PD and CVC disease in North and South America, respectively.

Detection of double stranded RNA in California isolates of Botryosphaeria dothidea from pistachio. W.-Q. CHEN (1,2) and T. J. Michailides (1). (1) Kearney Agricultural Center, University of California, Davis, Parlier, CA 93648; (2) Dept. Plant Protection, Northwest Sci-Tech University of Agriculture and Forestry, Yangling, Shaanxi, China 712100. Phytopathology 92:S14. Publication no. P-2002-0098-AMA.

To determine if double-stranded RNA was present in *Botryosphaeria dothidea*, causal agent of panicle and shoot blight of pistachio in California, 33 isolates were analyzed and also tested for relative virulence, based on dormant stems of pistachio and apple inoculation. A dsRNA extraction procedure was applied using mycelia of 4 day-old cultures grown on potato dextrose agar covered by a cellophane membrane. Isolation of dsRNA was by means of DNazol and phenol-chloroform-isoamyl alcohol extraction. Three of 33 isolates (9.09%) contained dsRNA. Each isolate contained one fragment, of 3.7, 2.5, and 2.5 kilobase pairs, respectively. While significant variation in virulence exists among isolates, no correlation between dsRNA presence and virulence was found. These studies indicate that the dsRNA segments did not confer hypovirulence to the pathogen, and therefore hypovirulence seems to have little potential for biological control of panicle and shoot blight on pistachio.

Development of RGAP markers for stripe rust resistance gene Yr15 and use of the markers to detect the gene in breeding lines. X. M. CHEN (1,2) and G. P. Yan (2). (1) USDA-ARS; (2) WSU, Pullman, WA 99164-6430. Phytopathology 92:S14. Publication no. P-2002-0099-AMA.

To develop molecular markers for *Yr15*, a wheat gene conferring resistance to stripe rust caused by *Puccinia striiformis* f. sp. *tritici* (PST), BC₇:F₄ lines were developed by backcrossing wheat line 'V763-251' to 'Avocet Susceptible' (AVS). Seedlings of the parents and BC₇:F₄ progeny were evaluated for resistance to three PST races under controlled greenhouse conditions. The resistance gene analog polymorphism (RGAP) technique was used to identify molecular markers for *Yr15*. Eighty-six of 343 primer pairs produced polymorphic bands in bulk segregant analyses. Of 11 RGAP markers produced by eight selected primer pairs and confirmed by co-segregation analysis with 196 BC₇:F₄ lines, one marker completely co-segregated with *Yr15* and the others were linked to *Yr15* with a genetic distance ranging from 0.8 to 13.8 cM. Five of the RGAP markers were used to determine if *Yr15* was present in 10 wheat breeding lines that were developed from crosses with the *Yr15* donor. The co-segregating RGAP marker clearly detected *Yr15* in one of the 10 lines, but not in others, a result that was supported by the tests of these lines with six PST races.

Epidemics and races of Puccinia striiformis in North America in 2001. X. M. CHEN (1,2) and M. K. Moore (2). (1) USDA-ARS; (2) WSU, Pullman, WA 99164-6430. Phytopathology 92:S14. Publication no. P-2002-0100-AMA.

Wheat stripe rust caused by *Puccinia striiformis* f. sp. *tritici* (PST) was widespread throughout North America in 2001. Yield losses were estimated to be more than 39 million bushels in the US. Collections of wheat stripe rust from the US and Canada were analyzed on 20 wheat genotypes that are used to differentiate PST races. Twenty-four previously identified races and 10 new races were identified. The most predominant races in the US and Canada were PST-78 (virulent on Lemhi, Heines VII, Lee, Fielder, Express, *Yr8*, *Yr9*, Clement, and Compar) and PST-80 (with all virulences of PST-78 plus virulence on Produra), which were first detected in 2000. Stripe rust of barley, caused by *P. striiformis* f. sp. *hordei* (PSH) occurred mainly in Texas, California, and the US Pacific Northwest. The disease caused only localized damage. Virulence patterns of barley stripe rust collections were determined on 12 barley genotypes that are used to differentiate PSH races. Of 19 races

detected in 2001, eight were new. Six of the new races were virulent on Bancroft, a recently released barley cultivar with stripe rust resistance. The Bancroft-virulent races were widely distributed throughout the US.

Characterization of an endosperm protein expressed at higher levels in maize genotypes resistant to *Aspergillus flavus* infection/aflatoxin production. Z.-Y. CHEN (1), R. L. Brown (2), K. E. Damann (1), and T. E. Cleveland (2). (1) Dept. Plant Pathology and Crop Physiology, Louisiana State Univ. Ag. Center, Baton Rouge, LA 70803; (2) SRRC, USDA-ARS, New Orleans, LA 70179. *Phytopathology* 92:S15. Publication no. P-2002-0101-AMA.

Aflatoxins are carcinogens produced by *Aspergillus flavus* and *A. parasiticus* during infection of susceptible crops such as maize. Though resistant maize genotypes have been identified, the incorporation of resistance into commercial lines has been slow due to the lack of selectable markers. Through comparisons of kernel proteins between resistant and susceptible genotypes separated using 2-D PAGE gels, several protein spots were found unique or upregulated in resistant genotypes, and were sequenced using ESI-MS/MS. One of them showed high homology to a PR-10 protein from sorghum. However, the biological function of this class of proteins remains unknown, although it has been proposed that they possess a RNase activity. To further investigate this protein, its cDNA has been cloned and it encodes a protein of 160 aa. This gene has been transferred into both *E. coli* and tobacco for antifungal studies. Its possible role in host resistance will be discussed.

Characterization of internal transcribed spacers from the black knot pathogen *Dibotryon morbosum* and gall related fungi on *Prunus* species. C. Chen, W. G. D. FERNANDO, and W. R. Remphrey. Dept of Plant Science, University of Manitoba, Winnipeg R3T 2N2, Canada. *Phytopathology* 92:S15. Publication no. P-2002-0102-AMA.

Dibotryon morbosum, causal agent of black knot, occurs on stone fruit trees (*Prunus* spp.) throughout Canada. For this study, total genome extraction was performed on pathogen strains CWC2, SABIA, SABIB, PFC1, ATCC15085, as well as other gall associated fungal strains *Fusarium* sp. KNR1, *Phoma* sp. JN1, *Trichothecium roseum* BRD1 and plant leaf tissue. A ribosomal region of the ITS-I, ITS-II and 5.8S region on the genome was amplified with PCR using ITS universal primers. A region of 534 bp was sequenced from both directions. The results indicated that similarity of the ITS region is more than 98% between the type culture of strain ATCC15085 and other *D. morbosum* strains, or *T. roseum* BRD1 isolated from cherry and plum trees in Canada. The ITS sequence of ATCC15085 partially matched to *Fusarium* sp. KNR1 and *Phoma* sp. JN1. However, there was no match between any fungal strains and host plant tissues in this region. In future studies, ITS fragment could be used as cDNA probe to identify *D. morbosum* genome extracted from diseased tissue using southern blot.

Introduction of the Tomato Spotted Wilt Virus nucleocapsid gene into a runner-type peanut. K. Chenault. USDA-ARS, Plant Science and Water Conservation Laboratory. *Phytopathology* 92:S15. Publication no. P-2002-0103-AMA.

Tomato Spotted Wilt Virus (TSWV) infects over 400 species of plants, including peanut. TSWV is a widespread peanut pathogen in the Southeastern United States causing significant losses to producers in that region. Although not yet a problem to peanut producers in the Southwestern United States, TSWV has been found recently in areas of northern Texas and southern Oklahoma. Traditional breeding practices have produced only few TSWV resistant peanut cultivars that are currently available for production in the Southwest. Coat protein-mediated resistance introduced into crops by genetic engineering offers an alternative method of control of infection by plant viruses. In this study, the nucleocapsid gene of TSWV was introduced into the runner-type peanut Okrun by microprojectile bombardment. Results from analysis of both T₀ and T₁ generations indicate stable incorporation and expression of the TSWV N gene into the cultivar Okrun has been achieved.

Induction of systemic acquired resistance by acibenzolar-S-methyl to plant-parasitic nematodes in pineapple. B. CHINNASRI, B. S. Sipes, and D. P. Schmitt. Dept. of Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, HI 96822. *Phytopathology* 92:S15. Publication no. P-2002-0104-AMA.

The effect of acibenzolar-S-methyl, a systemic acquired resistance inducer in plants, on *Meloidogyne javanica* and *Rotylenchulus reniformis* in pineapple was determined in the greenhouse. Foliar application of acibenzolar-S-

methyl at 50 mg/L at planting, 1, and 3 months after planting did not decrease the number of eggs of *M. javanica* and *R. reniformis* at the twelfth month after planting as compared to the untreated control. However, foliar treatment at 100 and 200 mg/L resulted in 40% and 55% fewer eggs of *M. javanica* and *R. reniformis*, respectively. Average fresh shoot and dry root weights of pineapple treated with 100 mg/L acibenzolar-S-methyl were 549 g and 29 g as compared to 579 g and 32 g in untreated plants. Another experiment was established to determine the phytotoxicity of acibenzolar-S-methyl to pineapple. At 12 months after planting, average fresh shoot and dry root weights of pineapple treated with 50 and 100 mg/L acibenzolar-S-methyl were not different from the untreated pineapples ($P > 0.05$). However, acibenzolar-S-methyl at 200 and 400 mg/L was phytotoxic. Foliar application of acibenzolar-S-methyl at 100 mg/L may activate intrinsic resistance of pineapple to *M. javanica* and *R. reniformis*.

Involvement of hormones in symptomatology of postbloom fruit drop (PFD) of citrus caused by *Colletotrichum acutatum*. K. R. CHUNG, R. Yuan, J. K. Burns, and L. W. Timmer. University of Florida. *Phytopathology* 92:S15. Publication no. P-2002-0105-AMA.

PFD is caused by *C. acutatum*. The fungus infects flower petals causing brownish lesions that result in fruit drop, production of persistent calyces, and leaf distortion. This suggests that hormones may be involved in symptom development. After infection by *C. acutatum*, ethylene and IAA increased significantly in citrus flowers. The ethylene concentration in infected flower tissues compared to water controls increased by 3-fold 5 days post inoculation. IAA accumulation in the infected petals was as much as 140 times that of the water control. ABA showed no significant response. Genes encoding proteins required for ethylene and jasmonic acid (JA) biosynthesis were highly expressed in infected flowers. Genes encoding IAA glucose transferase and auxin-responsive GH3-like protein were also highly expressed, whereas expression of the gene encoding IAA amino acid hydrolase was not different. These results combined with the biochemical data indicate that imbalance of IAA, ethylene and JA in *C. acutatum*-infected flowers may be involved in young fruit drop and other symptoms.

Genetics of root-knot nematode resistance in *Arachis* interspecific hybrids. G. T. CHURCH (1), J. L. Starr (1), and C. E. Simpson (2). (1) Dept. of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843; (2) TAES, Stephenville, TX 76401. *Phytopathology* 92:S15. Publication no. P-2002-0106-AMA.

A single dominant gene for resistance to *Meloidogyne arenaria* has been introgressed into tetraploid, cultivated peanut from the synthetic triple species hybrid, TxAG-6, where each species was a diploid wild *Arachis* spp. Evidence suggests that TxAG-6 has additional genes for resistance. The objective of this study was to determine the number of resistance genes present in TxAG-6 and BC₃F₂ breeding populations. TxAG-6 was crossed with the susceptible *A. hypogaea* cv. Florunner. F₂ plants were evaluated for resistance based on nematode reproduction under greenhouse conditions. Segregation of the resistance phenotype fit a one dominant gene and one recessive gene model, with 92 susceptible and 15 resistant individuals. Segregation in three BC₃F₂ populations, derived from TxAG-6 × *A. hypogaea* without selection, also fit a one dominant gene and one recessive gene model. These results suggest TxAG-6 possesses a second, recessive, gene for resistance to *M. arenaria*, in addition to the dominant gene identified previously.

Predicting forest ecosystems at risk to invasion of exotic forest pathogens in the USA. S. D. Cohen. USDA-APHIS-PPD, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108. *Phytopathology* 92:S15. Publication no. P-2002-0107-AMA.

Exotic forest pathogens pose a potential invasive threat to forest ecosystems of the USA. Some forest ecosystems may be more susceptible to invasion and establishment of exotic pathogens. One approach to predicting ecosystems at risk is to estimate the probability of pathogen establishment based on climate matching criteria, pathogen distribution records, and host availability. Estimating the probability of establishment entailed using a climate modeling software (CLIMEX) followed by logistic regression in a statistical program (SAS) and creation of isoclines in a GIS software (ARC-View, ver. 3.2a). This approach was evaluated with data from known outbreaks in the USA of the exotic fungal pathogen, *Melampsora laurici-populina*, Eurasian Poplar Leaf Rust. CLIMEX includes a number of weather stations for each continent. Increasing the number of weather stations in USA considered in the model improved the ecosystem predictions. Six climate factors, were initially included in the analysis and of the six factors; rainfall was the most significant predictor of pathogen establishment.

Protection of citrus rootstocks against *Phytophthora* species by prior inoculation with a hypovirulent *P. nicotianae* isolate. G. C. COLBURN, K. R. Chung, and J. H. Graham. University of Florida. Phytopathology 92:S16. Publication no. P-2002-0108-AMA.

Phytophthora root rot of citrus in Florida is caused by *P. nicotianae* and *P. palmivora*. Management of Phytophthora root rot includes the use of tolerant rootstocks and fungicides, but biological controls are not available. A naturally occurring mutant of *P. nicotianae* was identified as hypovirulent on citrus and reduced root rot caused by virulent isolates. Three-month-old sweet orange, sour orange, and Swingle citrumelo seedlings were planted individually in 250-ml containers. All rootstocks with prior exposure to the hypovirulent *P. nicotianae* isolate for either two or four weeks had a lower disease severity rating. Co-inoculation of the hypovirulent *P. nicotianae* isolate with either virulent isolate did not reduce disease severity. This demonstrates that the hypovirulent *P. nicotianae* isolate must pre-colonize the host roots to provide protection from virulent isolates. The mechanisms by which the hypovirulent isolate provides protection against virulent *Phytophthora* species are being investigated.

AFLP markers associated with virulence of *Heterodera glycines* on resistant soybean. A. L. COLGROVE (1) and T. L. Niblack (2). (1) Dept. of Plant Microbiology and Pathology, University of Missouri, Columbia, MO 65211; (2) Dept. of Crop Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S16. Publication no. P-2002-0109-AMA.

Rapidly changing field populations of *Heterodera glycines*, the soybean cyst nematode, can overcome resistance in soybean and complicate soybean cyst nematode management. Molecular markers associated with female development on resistant soybeans could help to identify virulent populations and track changes in field populations. AFLP analysis has been used to compare inbred lines of *H. glycines* with different virulence profiles on resistant soybean cultivars. The inbred lines compared are those with high female indices (FI) on PI88788 or PI437654 versus those with FI of zero on the same resistant soybeans. A 700-bp fragment has been found to be associated with development on PI88788; it does not occur in populations that do not develop on PI88788. A 1-kb marker has been found only in populations able to develop on PI437654, and absent in all other populations tested. This fragment has been cloned and sequenced and is under further investigation.

Characterization of bacterial communities in soil during the transition to organic agriculture. A. A. COLLINS, D. C. Fargo, and F. J. Louws. Dept. Plant Pathology, NC State University, Raleigh, NC 27695. Phytopathology 92:S16. Publication no. P-2002-0110-AMA.

Bacterial communities constitute a key component of the biological dynamics in sustainable agricultural systems and can be characterized based on their functional roles in soil ecology and plant health. A microtiter plate assay utilizing selective media and a most probable number method was adapted to quantify populations of *Burkholderia* and fluorescent pseudomonads as influenced by management strategies. Six treatments representing strategies for transition from traditional to organic crop production were implemented. Representative strains from each treatment site were characterized using genomic fingerprinting protocols (rep-PCR), and 16S rDNA sequencing. Bacterial populations examined were differentially impacted by both season and transition strategy. Genomic characterization of isolates provided an assessment of population diversity and validation of cultural methods. These results verify the accuracy and scope of our cultural methods for monitoring populations and demonstrate the disparate effects of seasonal and treatment influences upon key bacterial communities in agroecosystems.

An assessment of the bioactivity of endophytic actinomycetes. J. T. COOMBS (1,2) and C. M. M. Franco (2). (1) Plant Pathology, Cornell University, Ithaca, NY 14850; (2) Biotechnology, Flinders University, Adelaide, Australia. Phytopathology 92:S16. Publication no. P-2002-0111-AMA.

Wheat is colonised by endophytes from a number of actinomycete genera. Isolates were screened for the ability to influence plant growth and control phytopathogenic fungi *in vitro* and *in planta*. The data indicated that plant growth promotion (PGP) was most prevalent among a group of isolates related to *Streptomyces setonii*. 55% of all the isolates were able to strongly inhibit at least one of the six pathogens tested *in vitro*, while several actinomycete isolates inhibited all of the pathogens tested. *In planta* control of *Gaeumannomyces graminis* var. *tritici* (Ggt) was exhibited by 24% of the isolates. As with PGP activity, biocontrol of Ggt was commonly observed in the *S. setonii*-like isolates, with 80% significantly reducing disease symptoms on the host. Several endophyte strains also reduced symptoms of Ggt and *Rhizoctonia solani* on wheat in field trials.

Colonisation of *Arabidopsis thaliana* by an endophytic *Streptomyces* sp. J. T. COOMBS (1,2), C. M. M. Franco (2), and R. Loria (1). (1) Plant Pathology, Cornell University, Ithaca, NY 14850; (2) Biotechnology, Flinders University, Adelaide, Australia. Phytopathology 92:S16. Publication no. P-2002-0112-AMA.

Streptomyces sp. EN27 is a wheat endophyte; colonisation is associated with disease suppression and plant growth promotion. Our objective is to dissect the molecular plant-microbe interactions of the endophytic phenotype. Therefore, we assessed the ability of strain EN27 to colonize *Arabidopsis thaliana*. Seed were inoculated with this streptomycete and roots and shoots of seedlings were examined after 7 - 10 days with light and fluorescence microscopy and scanning electron microscopy. Growth of inoculated and control plants was evaluated. This strain readily colonized the interior and exterior of *A. thaliana* and increased plant growth. *A. thaliana* will be useful as a model host species for characterization of endophytic growth and the genetics of host response to endophytic colonization.

Complete sequence of pEN2701, a 12kb cryptic plasmid from an endophytic *Streptomyces* sp. J. T. COOMBS (1,2), C. M. M. Franco (2), and R. Loria (1). (1) Plant Pathology, Cornell University, Ithaca, NY 14850; (2) Biotechnology, Flinders University, Adelaide, Australia. Phytopathology 92:S16. Publication no. P-2002-0113-AMA.

Streptomyces sp. EN27 was isolated from surface sterilised wheat roots, where it was found to increase plant growth and antagonise the fungal root pathogen *Gaeumannomyces graminis* var. *tritici*. This isolate was examined for the presence of plasmids using pulsed field gel electrophoresis (PFGE). PFGE indicated that this streptomycete carries both a large and a small plasmid. The smaller plasmid was designated pEN2701. Standard agarose gel electrophoresis identified the smaller plasmid as circular, with a size of approximately 12kb when linearised with *Pst*I. We describe the isolation of the plasmid, its complete DNA sequence and analysis of the putative coding sequences.

Molecular mapping and quantitative trait loci (QTL) analysis of late blight resistance in potato. S. COSTANZO (1), B. J. Christ (1), and K. G. Haynes (2). (1) Dept. Plant Pathology, Penn State University, University Park, PA 16802; (2) USDA/ARS, Vegetable Lab, Beltsville, MD 20705. Phytopathology 92:S16. Publication no. P-2002-0114-AMA.

Phytophthora infestans, the causal agent of late blight, is responsible for severe losses in potato production worldwide. Since this oomycete can rapidly overcome major race-specific resistance genes, identifying the basis for quantitative resistance has become a high priority. A population of 240 F₁ progenies derived from a cross between two diploid hybrid *Solanum phureja* × *S. stenotomum* clones were evaluated in replicated trials for late blight resistance. A sub-population of 132 clones was genotyped with 140 restriction fragment length polymorphism (RFLP) markers covering 1082 cM of the potato genome with an average distance of 8.4 cM between markers. Based on this map, interval mapping analysis was performed which identified various QTLs affecting resistance to late blight. A major QTL was detected on chromosome 9 which appears unique to this hybrid population since it has not been previously reported in other late blight resistance studies. We plan to obtain more comprehensive molecular marker coverage of this region for future analysis.

A multilocus gene genealogy of *Pyricularia oryzae* to identify the origin of rice infecting populations. B. C. COUCH and L. M. Kohn. Department of Botany, University of Toronto, Mississauga Ontario L5L 1C6. Phytopathology 92:S16. Publication no. P-2002-0115-AMA.

Magnaporthe oryzae Couch¹ is the newly described teleomorph of the anamorph, *Pyricularia oryzae*. Prior studies suggest that *M. oryzae* consists of genetically distinct, host-specific populations. Populations from rice sharing a common origin should also share pathogenicity factors specific for rice. A multilocus phylogenetic analysis of *M. oryzae* is presented based on two samples. The first sample reflects the geographic and host diversity of *M. oryzae*. This sample was used to infer the relationships among *M. oryzae* isolates from different hosts. The second sample consists of parallel population samples of *M. oryzae* from rice and weeds from the Philippines and Vietnam to test for significant host associations within *M. oryzae*. In order to identify genetic changes that may be associated with pathogenicity towards rice, we have mapped AVR gene alleles and species specific pathogenicity genes on this phylogeny. ¹Couch, BC and Kohn, LM (2002) A multilocus gene genealogy concordant with host preference indicates segregation of a new species, *Magnaporthe oryzae*, from *M. grisea*. *Mycologia*. 94(3): In press.

Identification of *Alternaria brassicicola* genes differentially expressed during pathogenesis on *Arabidopsis thaliana* using suppression subtractive hybridization. R. A. CRAMER and C. B. Lawrence. Department of Bioagricultural Sciences and Pest Management, Plant-Fungal Genomics Laboratory, Colorado State University. *Phytopathology* 92:S17. Publication no. P-2002-0116-AMA.

Necrotrophic fungal pathogens are responsible for some of the world's most devastating plant diseases. *Alternaria brassicicola* (Schwein.) Wiltshire is a necrotrophic fungus that causes black spot disease on a wide range of cruciferous hosts including the model plant *Arabidopsis*. The objective of this study was to identify genes up-regulated during the early stages of *A. brassicicola* infection on *Arabidopsis*. Suppression subtractive hybridization (SSH) was employed to create a cDNA library enriched for such genes. Fungal spores were germinated either in sterile water or on leaves of the susceptible *Arabidopsis* ecotype *Landsberg erecta* (Ler). After a 24 hour incubation period at 24°C, RNA was extracted from these two fungal spore samples and used to create cDNA populations for use in SSH. Subtraction was performed between these cDNA populations to create a library enriched for genes unique to the spores germinated on the plant leaf surface. Up-regulation of clones corresponding to individual genes was confirmed using a dot blot technique coupled with virtual northern analysis. Fifty up-regulated clones were selected and sequenced. Database homology searches using blastn and blastx revealed sequences with homology to a putative arsenite ATPase translocase (ABC transporter), translation initiation factor, various glycoproteins, *Alternaria* allergen precursor, cyanide hydratase, and formate dehydrogenase that may be involved in pathogenesis. Based on the results obtained, SSH is an effective technique to identify fungal genes that may be important in the early stage of infection. Future research will involve functional characterization of these putatively important pathogenesis-related genes.

1,3-D and oxamyl for nematode management in cucumbers. A. S. CSINOS (1), K. W. Seebold (1), and R. F. Davis (2). (1) University of Georgia; (2) USDA. *Phytopathology* 92:S17. Publication no. P-2002-0117-AMA.

Field plots were established 8 Aug 2001 on a Tifton loamy sand naturally infested with *Meloidogyne incognita*. The experiment was a randomized complete block design with polyethylene film mulch beds 7.6 m long and 0.76 m wide replicated 5 times. Blue dye was injected into the drip tape to estimate movement of oxamyl in the soil. Treatments were methyl bromide (MB) 224 kg/ha; 1,3-D; 1,3-D plus oxamyl 0, 2 and 4 wks post plant (pp); 1,3-D plus oxamyl 2, 4 and 6 wks pp; oxamyl 0, 2 and 4 wks pp; oxamyl 2, 4 and 6 wks pp; 1,3-D plus oxamyl 4, 6, 8 wks pp; oxamyl 0, 2 and 4 wks pp; and control. All 1,3-D was applied at 76 l/ha by chisel injection 46 cm deep. All oxamyl was applied at 4.67 l/ha per application via drip tape. Cucumber weight was increased by the MB and 1,3-D + oxamyl 2, 4 and 6 weeks pp treatments. Numbers of cucumbers were increased by all treatments except those which received only oxamyl. Numbers of *M. incognita* juveniles and the root gall index observed at harvest were decreased by all treatments except oxamyl alone. MB and 1,3-D treatments reduced colony forming units of *Pythium* spp. at planting and at harvest.

Does the viable but non-culturable state have implications for eradication of citrus canker bacterium? J. CUBERO and J. H. Graham. Citrus Research and Education Center, University of Florida, Lake Alfred, FL 33850. *Phytopathology* 92:S17. Publication no. P-2002-0118-AMA.

Establishment of *Xanthomonas axonopodis* pv. citri (*Xac*), the cause of citrus bacterial canker (CBC), threatens Florida citriculture. Hence, a massive effort is underway to eradicate CBC. Effective eradication is based on the presumption that the bacterium has limited survival outside of lesions and, therefore, elimination of symptomatic tissues removes all the extant inoculum sources. A viable but non-culturable (VBNC) condition is defined as a physiological state wherein bacterial cells remain viable but cannot be recovered on culture media. VBNC may be induced by adverse conditions that result in a starvation phase from which bacteria may be resuscitated. *Xanthomonas axonopodis* pv. citrumelo, was used as a model to explore for the existence of a VBNC state. Viability is evaluated with a double fluorescence assay (LIVE/DEAD, Molecular Probes) based on different permeability of the intact vs. damaged membranes. In a low nutrient solution with copper, a proportion of the population appeared viable by LIVE/DEAD kit, but unculturable for several days.

Genetic variability and QTL mapping of resistance to gray leaf spot in a *Lolium pseudo-testcross* population. J. CURLEY (1), S. Warnke (2), R. Barker (2), S. Leong (1), and G. Jung (1). (1) Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706; (2) USDA-ARS National

Forage Seed Production Research Center, Oregon State University, Corvallis, OR 97331. *Phytopathology* 92:S17. Publication no. P-2002-0119-AMA.

Gray leaf spot (GLS) is a serious fungal disease on the important turf and forage species, perennial ryegrass (*Lolium perenne*) caused by the rice blast fungus *Magnaporthe grisea*. Early reports suggest little resistance is present in perennial ryegrass cultivars. However, greenhouse inoculations in our lab using several rice and ryegrass isolates suggests some resistance is present, both in sixteen F1 clones derived from crosses among individual clones of cultivars, and in a subsample of progenies from an annual × perennial ryegrass (MFA × MFB) F2 pseudo-testcross population. These two groups of genotypes varied widely in resistance, as measured by lesion type and severity. The results were generally consistent, and the clones MFA and MFB were generally more tolerant than others. Also, the progeny subsample, when scored by the most severe lesion present of the three replicates, appears to segregate in a 1:2:1 ratio, though this needs confirmation. Thus, both the MFA × MFB population and the F1 genotypes show potential for use in quantitative trait locus (QTL) mapping and resistance breeding through recurrent selection. This study will examine variability in resistance, select parents for a new F2 pseudo-testcross mapping population, and make crosses. Also, genetic linkage map construction will begin using AFLP and RAPD markers, and RFLP synteny anchor probes from cereal crops. A partial map has already been constructed for the existing MFA × MFB population, and the phenotypic segregation seen in the subsample will enable QTL analysis in this population. With this information, locations and effects of QTL for GLS resistance in both rice and ryegrass can be compared via comparative QTL mapping in two different populations, which can then lead to map-based cloning of novel resistance genes in both species.

Antibiotic, genetic, and nutrient utilization characteristics among a collection of Streptomycetes. A. L. DAVELOS, D. Johnson, and L. L. Kinkel. Dept. Plant Pathology, University of Minnesota, St. Paul, MN 55108. *Phytopathology* 92:S17. Publication no. P-2002-0120-AMA.

Antibiotic inhibition is believed to play a significant role in competitive interactions in soil. However, the dynamics of antibiotic inhibition and resistance among soil microbes have been poorly characterized. To rapidly screen antibiotic capabilities among soilborne microbes, a collection of standard test isolates was developed. Ten Streptomycete isolates from a variety of agricultural soils and having diverse inhibitory activities were characterized based upon antibiotic inhibitory, resistance and induction capabilities in all possible pair-wise combinations. Nutrient utilization profiles and genetic relatedness using rep-PCR were also evaluated. Isolates that were good at inhibiting others were resistant to more isolates than were poor inhibitors. Inhibition was more frequent among isolates having similar nutrient use patterns than among isolates having distinct nutrient use profiles. These genetically distinct isolates present a diverse range of antibiotic inhibition, resistance, and induction capabilities against which field isolates may be characterized.

Laboratory evaluation of the effects of temperature and soil matric potential on infection of wheat by *Polymyxa graminis* using air pressure cells. L. E. DAVIDSON (1), R. R. Schindelbeck (2), H. M. van Es (2), and G. C. Bergstrom (1). (1) Dept. Plant Pathology; (2) Dept. Crop and Soil Sciences, Cornell University, Ithaca, NY 14853. *Phytopathology* 92:S17. Publication no. P-2002-0121-AMA.

Infection by *Polymyxa graminis* and the dozen viruses it is presumed to vector is affected by a number of environmental parameters, primarily temperature and soil matric potential. In this study, air pressure cells were used to establish soil matric potential treatments on wheat growing in *P. graminis*-infested soil cores. Following removal from the air pressure cell apparatus, matric potentials were maintained by daily hand watering and infection was detected by ELISA. Soil matric potentials of -20 kPa and wetter were significantly more conducive for *P. graminis* infection and virus transmission than -40 kPa. Using this knowledge, the duration of conducive moisture required for infection at ten days after planting was tested at 6.5, 10, 15, and 20°C. Significant infection occurred within 48 hours at 10, 15, and 20°C, but not at 6.5°C. Infection occurred within 24 hours at 15°C. We are validating these results in the field. Air pressure cells are a novel tool for the epidemiology of soil-borne pathogens.

Another canker-causing *Phytophthora* from California and Oregon forest trees. J. M. Davidson (1), M. Garbelotto (2), E. M. Hansen (3), P. REESER (3), and D. M. Rizzo (1). (1) Dept. Plant Pathology, U.C. Davis, Davis, CA 95616; (2) Dept. Environmental Science, Policy, and Management, U. C. Berkeley, Berkeley, CA 94720; (3) Dept. Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331. *Phytopathology* 92:S17. Publication no. P-2002-0122-AMA.

Phytophthora ramorum causes sudden oak death on tanoak and coast live oak trees, and foliar and dieback symptoms on other tree species. A second, apparently undescribed *Phytophthora* species is occasionally isolated from lethal cankers on tanoak and coast live oak, and from foliar lesions on tanoak and myrtlewood, in areas where *P. ramorum* is also active. ITS DNA sequence indicates close relationship to *P. ilicis* (a foliar pathogen of holly) and *P. psychrophila* (newly described from European oak forest soils). It is homothallic with amphigynous antheridia, and has deciduous sporangia. It grows more slowly, with a lower temperature optimum, than *P. ramorum*. In log inoculation tests it is nearly as pathogenic to tanoak as *P. ramorum*. It does not infect holly leaves in leaf inoculation tests. In the forest it is usually associated with single killed trees, in contrast to the expanding patches of mortality caused by *P. ramorum*.

Development of transgenic ringspot virus-resistant papaya for Florida. M. J. DAVIS and Z. Ying. Tropical Research and Education Center, University of Florida, Homestead, FL 33031. Phytopathology 92:S18. Publication no. P-2002-0123-AMA.

To develop transgenic papaya with PRV resistance for Florida, somatic embryos were subjected to *Agrobacterium*-mediated transformation with constructs containing sense or anti-sense orientations of a PRV coat protein gene from a Florida isolate. The sense orientation of the gene was either not modified or modified by the insertion of either frame-shift or stop-codon mutations. Following regeneration, 360 putatively transgenic plants were inoculated with PRV. Twenty of the most resistant lines having single transgene copies were selected and crossed with five elite papaya genotypes. However, only lines with the frame-shift or stop-codon mutations were highly fertile. In the field, 293 (23.3%) of 1258 transgenic progeny from 54 crosses became naturally infected by PRV within one year of planting. In comparison, 29 (96.7%) of 30 non-transgenic plants in the same planting became infected. Infection varied from 4.6% to 38.9% among the five elite parents, and from 10.4% to 87.9% among the 17 transgenic lines. Selections were made among plants of the most resistant lines for further variety development.

The effect of root tip exudates on nematode behavior. J. DECKER, X. Zhao, M. Schmitt, M. A. McClure, and M. Hawes. University of Arizona. Phytopathology 92:S18. Publication no. P-2002-0124-AMA.

A non-polar, heat-stable product from root exudates of legumes (Factor N) is known to trigger a transient, reversible state of quiescence in certain plant-parasitic and free-living nematodes. The purpose of this study was to characterize the activity of the exudate and determine its effect on the mobility, and infectivity of *Meloidogyne incognita*. Alfalfa, cucumber, and other plants, produced root tip exudates that induced quiescence in *M. incognita*, *Ditylenchus dipsaci*, *Caenorhabditis elegans* and *Steinernema glaseri*. Lettuce and cotton did not. Infection of alfalfa seedlings by *M. incognita* more than doubled when infective juveniles were incubated up to 12 hours in pea root exudates. Several mutants of *C. elegans* with defects in known metabolic pathways were screened for sensitivity to pea and cucumber root tip exudates. None differed from the wild-type individuals in their response. Treatment of wild-type individuals of *C. elegans* with ethyl methane sulfonate resulted in a mutant population that resisted quiescence. We conclude that behavioral response of nematodes to root tip exudates may be genetically determined.

Detection of anthracnose race 73 on dry beans in Manitoba, Canada. L. E. DEL RIO (1), R. S. Lamppa (1), P. L. Gross (1), J. Prischmann (2), and B. Brolley (3). (1) Dept. Plant Pathology, North Dakota State Univ. Fargo, ND 58105; (2) North Dakota State Seed Dept. Fargo, ND 58105; (3) Manitoba Agriculture and Food, Carman, Mb R0G 0J0. Phytopathology 92:S18. Publication no. P-2002-0125-AMA.

Five *Colletotrichum lindemuthianum* (Sacc. & Magnus) Lams.-Scrib. isolates from Manitoba were inoculated on a set of 12 standard differentials for race identification purposes. Two isolates were retrieved from seed samples processed by the North Dakota State Seed Dept.; the rest were retrieved from infected pods. Spores produced on Mathur's medium were suspended in water and Tween 80 (0.1% v/v) and adjusted to 106 spores/ml. Thirty two-week old seedlings of each differential were inoculated with each isolate on their primary leaves using a Paasche airbrush. Inoculated plants were incubated in moist chambers for five days at 20°C under 14-hr fluorescent light and then moved back to the greenhouse. Disease reaction was assessed three days later. All isolates produced sporulating lesions only on differentials 'Michelite', 'Cornell 49242', and 'Mexico 222', reaction typical of race 73. Although anthracnose has been present in Manitoba for long time, no races had been identified before.

Effect of foliar applications of calcium on control of white mold on dry beans. L. E. DEL RIO, P. L. Gross, R. S. Lamppa, and J. Venette. Dept. Plant Pathology, North Dakota State University, Fargo, ND 58105. Phytopathology 92:S18. Publication no. P-2002-0126-AMA.

Calcium has been proposed as an inexpensive alternative to control white mold (*Sclerotinia sclerotiorum*) of dry beans. Foliar applications of 2,000 ppm of calcium sulfate and/or calcium chloride, alone or in combination with 590 g a.i. per ha of thiophanate methyl or benomyl (one half of the recommended rates), were evaluated between 1997 and 2001 in North Dakota. Experiments had four replications and were conducted at two locations every year. Treatments were delivered at flowering time at a rate of 300 l per ha. Experiments were individually analyzed using orthogonal contrasts, and when statistically acceptable, data were combined over locations by year. White mold incidence averaged 60% in untreated (water) controls for the 10 environments. Calcium alone did not significantly reduce white mold incidence or severity and did not increase yields in any environment compared to untreated controls. Calcium had no effect on disease or yield when mixed with reduced fungicide rates. Fungicides applied at full rate (1,180 g a.i. per ha) offered significantly better protection than the mix of calcium and reduced fungicide in all environments.

Rice germplasm reaction to a venezuelan isolate of the sheath blight pathogen. N. J. DELGADO, H. A. Rodriguez, and M. C. Ramon. Instituto Nacional de Investigaciones Agricolas, Venezuela. Phytopathology 92:S18. Publication no. P-2002-0127-AMA.

Sheath blight (ShB) is an important worldwide rice disease caused by *Rhizoctonia solani* Khun. In Venezuela, its incidence has increased in recent years in intensive rice production systems. Complete resistance to this pathogen has not been found in rice, but differences in reaction have been observed among varieties. The objective of this study was to assess the reaction of 40 rice accessions from the small grain collection at Aberdeen, ID, to the rice sheath blight pathogen. The experiment was in a complete randomized design with two repetitions. Seeds were sterilized and germinated in autoclaved paper towels, and transplanted to sterilized soil in a greenhouse concrete tank. Sixty days old plants were inoculated placing a sclerotium to the inner side of the second sheath, at the collar level. Lesion size and lesion number were recorded on inoculated and subsequent leaves, position of the highest infected sheath and number of diseased tillers were counted. The results show that most of the evaluated accessions were susceptible to the ShB isolate used but two of them were moderately resistant and will be used for breeding purposes.

Refugia of *Colletotrichum acutatum* in dormant highbush blueberry. A. DeMARSAY and P. V. Oudemans. Department of Plant Biology and Pathology, Rutgers University Blueberry and Cranberry Research and Extension Center, Chatsworth, NJ 08019. Phytopathology 92:S18. Publication no. P-2002-0128-AMA.

Identification of the overwintering sites, or refugia, of *Colletotrichum acutatum*, the causal agent of anthracnose fruit rot of highbush blueberry, is crucial to risk assessment and effective disease management. Current literature suggests that *C. acutatum* overwinters only in dead tissues of the host. Studies in 2001 provided evidence that the fungus can survive in or on both dead and live plant parts. Dormant canes (cv. 'Bluecrop') from a commercial field were incubated in moist chambers and emerging spore masses tallied to determine the number and proportion of infections arising from various tissue types. Blighted tips of canes harbored the pathogen in significantly higher proportions than other types of tissue. Because there were many more flower and vegetative buds than dead tips on each cane, the actual number of infections arising from live tissue equaled or exceeded that from dead tissue. A similar analysis of dormant canes from three cultivars with varying susceptibilities to anthracnose ('Bluecrop', 'Duke', and 'Elliott'), collected from unsprayed fields, is underway.

Effects of burning and nutrient additions on *Puccinia dioicae* infecting *Erigeron strigosus* in tallgrass prairie. S. P. DENDY, K. A. Garrett, and H. U. Ahmed. Dept. of Plant Pathology, Kansas State University, Manhattan, KS. Phytopathology 92:S18. Publication no. P-2002-0129-AMA.

Puccinia dioicae causes a leaf rust on daisy fleabane, *Erigeron strigosus*, a common annual forb in tallgrass prairie. Unburned or annually burned plots were divided into subplots with added N, P, N+P or no amendment. In mid-June, we measured leaf rust severity (percent infected leaves per plant), inflorescence size and plant height. Mean leaf rust severity was higher in 2000 (mean 28%) than in 2001 (mean 4%), probably because of the unusually hot and dry summer after sampling in 2000. Leaf rust severity was significantly higher in unburned (mean 42%) than in burned (mean 8%) plots

in 2000. Rust severity was not affected by P amendment, but inflorescence size was greater in +P than in -P plots in 2000. Rust severity and inflorescence size were higher in unburned than in burned plots, and higher in +P than in -P plots in 2001. Rust severity and inflorescence size were greater in +N than in -N unburned plots in 2001. Height was greater in +P than in -P plots in both years. There was some evidence for correlation between the abundance of the primary host, *Carex grvida*, and rust severity on alternate host *E. strigosus*.

Hypovirulence-associated dsRNA from *Sclerotinia homoeocarpa* is conspecific with *Ophiostoma mitovirus 3a-OnuLd*. F. DENG (1), R. Xu (2), M. S. Melzer (1), and G. J. Boland (1). (1) Dept. of Environmental Biology; (2) Laboratory Service Division, University of Guelph. Phytopathology 92:S19. Publication no. P-2002-0130-AMA.

The nucleotide sequence of the hypovirulence-associated double-stranded RNA (dsRNA) in *Sclerotinia homoeocarpa*, the causal agent of dollar spot of turfgrass, was obtained. This dsRNA is 2632 bp long and one strand contains an opening reading frame with the potential to encode a protein of 720 amino acids. The amino acid sequence contains all the conserved motifs of RNA-dependent RNA polymerases (RdRps). Sequence analysis of nucleotide and RdRp-like protein revealed that this dsRNA is homologous with previously characterized mitochondrial viruses and dsRNAs, and shares 91.1% nucleotide and 95.0% amino acid sequence identities with the *Ophiostoma mitovirus 3a-OnuLd* from *Ophiostoma novo-ulmi*, the causal agent of Dutch elm disease. The results indicate that these two dsRNA viruses are conspecific. This is the first report that a dsRNA virus naturally occurs in two taxonomically distinct fungi, and indicates that horizontal transmission of this virus may have occurred between these fungi.

Mapping chromosome 7 specific ESTs of *Magnaporthe grisea*. J. DENG (1), H. Zhu (2), W. Choi (1), and R. A. Dean (1). (1) Fungal Genomics Laboratory, North Carolina State University, Campus Box 7251, Raleigh, NC 27695-7251; (2) Department of Molecular, Cellular, and Developmental Biology, Yale University, New Haven, CT 06520. Phytopathology 92:S19. Publication no. P-2002-0131-AMA.

Magnaporthe grisea is a destructive fungal plant pathogen and the causal agent of rice blast disease. To clarify gene organization and distribution and to help refine the physical map, we anchored appressorium stage ESTs to chromosome 7. We first identified the unique set of ESTs previously anchored to chromosome 7 and used them in a two dimensional pooling strategy to probe against a BAC library consisting of 9216 clones arrayed on a nylon filter (25X genome coverage). Contigs containing the positively hybridizing BACs were then identified and the contig's chromosomal location was determined using the existing physical map on our federated database Magnaporthe db. ESTs hybridizing to unanchored BACs or to BACs assigned to a different chromosome were probe against a CHEF gel blot to determine if they reside on chromosome 7. As the result, we were able to confirm that 86% of the initial 181 ESTs reside on chromosome 7. Furthermore, we assigned five previously unanchored contigs to chromosome 7.

Insight into *Ralstonia solanacearum* pathogenesis using strains expressing fluorescent proteins. T. P. DENNY and H. Liu. Dept. Plant Pathology, Univ. of Georgia, Athens, GA 30602. Phytopathology 92:S19. Publication no. P-2002-0132-AMA.

Stable derivatives of wild-type strain AW1 were made that constitutively express green fluorescent proteins (GFP-UV, ECFP-S72A, GFP-mut2, GFP-mut3) or the improved red fluorescent protein DsRed2, and permitted single cells to be observed microscopically. Following soil-drench inoculation with a GFP-mut2+ strain, tomato plants were observed at frequent intervals while bacteria invaded unwounded roots and systemically colonized tap roots and stems. Major conclusions were: (i) epidermal cells of lateral roots could be internally colonized within one day; (ii) the vascular cylinder of some lateral roots was extensively colonized within 2 days; (iii) one or more xylem vessels were colonized throughout the plant by 4 days, which is before the first wilt symptoms appear; and (iv) bacteria migrated into both the stem pith and cortex by 4 days, and later virtually all plant tissues were invaded. An AW1 GFP-mut2+ mutant lacking type III (*hrp*) protein secretion invaded roots as well as the wild type. Inactivation of type II (*out*) protein secretion almost abolished invasion and colonization of unwounded roots.

Biological diversity of *Gibberella zeae* from Nepal: genotypes, virulence, and toxins. A. E. DESJARDINS (1), R. D. Plattner (1), and A. M. Jarosz (2). (1) USDA-ARS National Center for Agricultural Utilization Research, Peoria, IL; (2) Michigan State University, East Lansing, MI. Phytopathology 92:S19. Publication no. P-2002-0133-AMA.

Gibberella zeae collected from small farms in Nepal displayed unusually high biological diversity. 600 strains from various hosts were classified using SCAR and AFLP markers, ability to cause wheat head blight and produce nivalenol (NIV) or deoxynivalenol (DON). Strains from SCARs 3 and 5 were more virulent (70% blight severity), and strains from SCAR 2 were less virulence (32%), suggesting that strains in this SCAR are not adapted to infect wheat. One-third of the strains tested produced only DON while 2/3 produced NIV. In SCARs 1, 2, and 5, >95% of the strains in each SCAR produced the same toxin, either DON or NIV, but in SCAR 3 equal numbers of DON-producers and NIV-producers were present. Generally, NIV-producing strains were less virulent (38%) on wheat than DON producers (57%). Preliminary AFLP results suggest that SCARs 3 and 5 are very similar to each other, while SCARs 1 and 2 differ from each other and the 3/5 complex. Our data suggest that the Nepal population of *G. zeae* is more diverse and more structured than the population in the United States.

MARYBLYT: Can there be improvements? M. M. DEWDNEY (1), A. R. Biggs (2), G. Lightner (2), and W. W. Turechek (1). (1) Cornell University, Geneva, NY; (2) West Virginia University, Kearneysville, WV. Phytopathology 92:S19. Publication no. P-2002-0134-AMA.

MARYBLYT is a forecaster used to identify infection periods for fire blight of apple and pear. In the blossom blight submodel of MARYBLYT, the minimum conditions (i.e., thresholds) necessary for blossom infection are: 1) flowers open with stigmas and petals intact; 2) accumulation of at least 110 cumulative degree hours greater than 18.3 C from the start of bloom; 3) a wetting event of at least 0.25 mm of rain or heavy dew or a rain of 2.5 mm or more the previous day; and 4) an average daily temperature of 15.6 C. MARYBLYT characterizes risk as either low, moderate, high or infection depending on whether one, two, three, or four of the risk factors have exceeded their minimum values. The thresholds used in the MARYBLYT model were selected based on empirical assessment of meteorological data in combination with the appearance of fire blight symptoms. ROC (Receiver Operator Curve) analysis, however, is a statistical procedure used to optimize thresholds for decision making. We used ROC analysis to validate MARYBLYT parameters and to identify where key improvements need to be made using historical data sets.

Suppression of *Rhizoctonia* pre-emergence damping-off of impatiens by enhanced microbial activity in potting mix amended with composted swine waste. H. G. DIAB EL-ARAB, S. Hu, and D. M. Benson. North Carolina State University, Raleigh. Phytopathology 92:S19. Publication no. P-2002-0135-AMA.

A peat moss based potting mix was amended with microbially stable compost at rates 4 to 20% to evaluate suppression of *Rhizoctonia* damping-off in impatiens. Plug trays filled with compost amended potting mix were seeded with impatiens then covered with a reserved portion of the treatment mix infested with *R. solani*. Stand counts were made until noninfested controls reached complete emergence. Samples were removed from duplicate trays for analysis of microbial respiration, biomass carbon and nitrogen. Pre-emergence damping-off was significantly lower for impatiens grown in mix amended with 20% compost than in the nonamended mix. A multiphasic increase in microbial respiration from compost-amended samples at seeding suggested that bacterial utilization of carbon occurred within 24 hr. A second increase occurred around 7 days when fungi would be expected to predominate. Microbial biomass carbon and nitrogen were greatest in mix amended with 20% compost. Our results suggest that enhanced microbial activity in stable swine compost is responsible for suppression of *R. solani*.

Temperature and wetness duration requirements for almond anthracnose development on leaves and blossoms. J. DIEGUEZ-URIBEONDO, H. Förster, and J. Adaskaveg. Dept. Plant Pathology, University of California, Riverside, CA 92521. Phytopathology 92:S19. Publication no. P-2002-0136-AMA.

Anthracnose, caused by *Colletotrichum acutatum*, is an important disease of almonds in California. Because infections can occur throughout the spring and early summer, a model for disease prediction would improve anthracnose management by more effective fungicide application timings. Microclimate parameters conducive for disease development (temperature, wetness duration - WD) were evaluated in growth chamber inoculation studies on cv. NePlus Ultra, Wood Colony, Carmel, and Nonpareil. For both leaves and blossoms, 10°C represented a critical low threshold temperature for disease development. Longer WD was needed for leaves than for blossoms indicating the higher susceptibility of blossoms. At 15°C, the positive slope of the regression of disease intensity on WD between 3 and 72 h was significantly greater for leaves than for blossoms. Field data indicated that fruit responded similar to leaf tissue. Among the cultivars evaluated,

Nonpareil was least and NePlus Ultra was most susceptible. Our data show that different disease prediction models are required for blossom and leaf/fruit tissues.

Digital image analysis of the internal light spots in appressoria of *Colletotrichum acutatum*. J. DIEGUEZ-URIBEONDO, H. Förster, J. E. Adaskaveg. Dept. Plant Pathology, University of California, Riverside, CA 92521. Phytopathology 92:S20. Publication no. P-2002-0137-AMA.

The initial penetration process of almond leaves by *Colletotrichum acutatum* was studied using the image-analysis software Syncrosopy Automontage. This software was used to analyze light micrographs of appressoria to generate a single, completely focused montage image with a continuous depth of field for a series of sequential, partially focused digital images. It is generally accepted that the internal light spot (ILS) of appressoria is the penetration pore. In studies on the development of the ILS, we observed that 50% and 95% of the appressoria formed an ILS after inoculated leaves were incubated for 12 and 24 h at 20 C, respectively. We also found that appressoria on glass surfaces never produced an ILS. Comparative image analysis of appressoria with and without ILS using color depth mapping and line profile software options showed that ILS had a depth relief that was below that of the leaf surface. This study represents the first direct evidence that the ILS is the developing infection peg. Thus, this new technique adds another dimension to light microscopy of plant pathogen interactions.

Effect of cereal residue burning on incidence and the stratified distribution of *Fusarium graminearum* and *Cochliobolus sativus* in wheat and barley plants. R. Dill-Macky and B. SALAS. Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108-0010. Phytopathology 92:S20. Publication no. P-2002-0138-AMA.

The stratified incidence of *Fusarium graminearum*, *Cochliobolus sativus* and other pathogens was studied in barley and wheat planted in three field plots in Minnesota in 2001 after cereal residue was burned using a flame thrower. Isolations made on half-strength PDA (pH=5.5) from subcrown internodes, crowns, nodes and kernels showed that regardless of the host, *F. graminearum* was mostly associated with kernels, whereas *C. sativus* was mostly associated with crowns and node-1. In contrast, *Pyrenophora teres* in barley was mostly associated with node-3. Incidence of *F. graminearum* was less ($P = 0.05$) in wheat plants collected from burned plots (3.3%) in comparison with those collected from the non-burned plots (5.3%). The effect of residue burning on the incidence of *C. sativus* and *P. teres* was not significant. Our data shows that *F. graminearum*, *C. sativus* and *P. teres* preferentially colonize certain plant parts and that residue burning may provide another option in the management of cereal diseases such as Fusarium head blight.

Carbohydrates for rapid indefinite axenic growth of the fusiform rust fungus. A. M. Diner. U.S.D.A. Forest Service, Southern Research Station, SFR, University of Florida, Gainesville, FL 32611. Phytopathology 92:S20. Publication no. P-2002-0139-AMA.

Rust fungi show slow and limited (staled) growth on axenic media. Several carbon sources were examined in attempts to promote rapid growth and preclude staling of *Cronartium quercuum* f. sp. *fusiforme*. Glucose is commonly used in axenic media for rust fungi. Substituted for glucose were arabinose, cellulose (soluble), dextrin type 4, erythritol, lactose, mannitol, sorbitol, sucrose and xylose, individually added (1% w/v) to agar-solidified Harvey and Grasham's medium. Media were inoculated at 30 sites each with 10ul of a mycelial homogenate. Plates were dark-incubated @ 22C for 8 weeks. No growth was supported by arabinose, lactose or xylose. By 8 weeks, established colonies on glucose, sucrose, sorbitol, dextrin and mannitol were 5, 10, 10, 12, and 15 mm diameter, respectively. Erythritol supported only slight sub-surface growth. Growth on cellulose was negligible. Growth of established colonies was progressive through at least 12 weeks. Colonies on dextrin medium reached 22 mm in 6 months, with growth becoming exclusively sub-surface after 8 weeks, and viability retained throughout.

Silver nitrate completely decontaminates slash pine seed. A. M. Diner. U.S.D.A. Forest Service, Southern Research Station, SFR, University of Florida, Gainesville, FL 32611. Phytopathology 92:S20. Publication no. P-2002-0140-AMA.

Silver nitrate was used to decontaminate unstratified, dry-stored vs. 18h-imbibed slash pine (*Pinus elliotii* Engelm.) seed for subsequent *in vitro* research. Seed, agitated in aqueous 0.25% (w/v) silver nitrate for 0, 1, 3, 5, 10, 30, 60 min and 6 h at 22C were rinsed 3 x 30 min in sterile, distilled

water. Seed (150) from each treatment were applied to water agar in Petri dishes, then Parafilm-sealed and incubated 22C under a 16-h, 40uE-day. Germination occurred between 4-10 days in all but the 6h treatments (5-14 days). Untreated seed showed pervasive contaminant overgrowth by 4 days; all seed eventuated colonies, and no data were taken therefrom. Germination frequencies for the treated, dry seed were 63, 68, 74, 65, 59 and 55%, respectively, and 65, 75, 58, 58, 69 and 61% respectively, for the treated, imbibed seed. Numbers of contaminated seed were 8, 6, 4, 3, 1, and 0, respectively for treated, dry seed, and 3, 3, 3, 1, 0 and 0 for treated, imbibed seed. Conventional, multi-step treatment using ethanol, bleach and peroxide showed 26% contamination.

Managing *Phytophthora sojae* in the midst of population shifts. A. E. DORRANCE (1), D. Mills (1), and P. E. Lipps (1). Dept. of Plant Pathology, The Ohio State University, Wooster, OH 44691. Phytopathology 92:S20. Publication no. P-2002-0141-AMA.

Recent soil surveys in Ohio have identified a greater number of fields in which *P. sojae* was isolated as well as an increase in pathotype diversity among isolates compared to previous surveys. From a survey of 86 fields, 82 were positive for *P. sojae* and 72 races were identified among 429 isolates. Many of the populations that now exist in this region can cause disease on soybean cultivars with commonly deployed *Rps* resistance genes. Additional disease management practices are needed to minimize the economic impact of these changing populations. Yields of soybean cultivars with a single *Rps* gene combined with partial resistance consistently ranked higher than cultivars with no partial resistance across 7 environments. The pathogen populations ranged from simple (susceptible interaction with <3 *Rps* genes) to complex in these environments. Selection of soybean cultivars with single *Rps* genes combined with high levels of partial resistance; increasing soil drainage through tiling or tillage; and the use of seed treatments to provide protection prior to emergence are tools that can mitigate some of the adverse effects on yield.

Effect of buried drip irrigation on reducing fungal fruit decay in fig orchards. M. A. DOSTER (1), T. J. Michailides (1), and D. A. Goldhamer (2). (1) Dept. Plant Pathology; (2) Dept. Land, Air, and Water Resources, University of California, Davis/Kearney Agricultural Center, Parlier, CA 93648. Phytopathology 92:S20. Publication no. P-2002-0142-AMA.

Buried drip irrigation was compared to the common practice of surface drip irrigation in two commercial fig orchards (cv. Conadria and cv. Black Mission) in California in 1998 and 1999. Surface drip irrigation caused the soil surface surrounding the water emitters to become wet, resulting in figs sitting on the wet soil before harvest. In contrast, where the drip lines were buried, the soil surface remained dry. The levels of decay fungi (*Alternaria Ulocladium*, *Aspergillus* sections *Nigri* and *Flavi*, *Fusarium*, and *Penicillium*) were quantified in the soil, on leaves, and in the figs. Although in the soil and on the leaves the densities of decay fungi did not differ significantly between the two irrigation treatments, fruit from the wet areas under the trees irrigated with surface drip tended to have a higher incidence of decay, especially on the external fruit surface, than fruit from dry areas. In addition to increasing irrigation efficiency, the use of buried drip irrigation should result in fewer culls and less fruit decay.

Identifying peptides that bind to and inhibit secreted cellulase of soybean cyst nematode using a phage display peptide library. W. DU (1), R. S. Hussey (3), T. J. Baum (2), and E. L. Davis (1). (1) Department of Plant Pathology, Campus Box 7616, North Carolina State University, Raleigh, NC 27695-7616; (2) Department of Plant Pathology, Iowa State University, Ames, IA 50011; (3) Department of Plant Pathology, University of Georgia, Athens, GA 30602. Phytopathology 92:S20. Publication no. P-2002-0143-AMA.

The feasibility of identifying inhibitors of the products of nematode parasitism genes is being investigated using the secreted cellulases of *Heterodera glycines* as a model target. The HG-eng-2 endoglucanase gene of *H. glycines* was expressed in a *Pichia pastoris* host. Active recombinant HG-ENG-2 with a C-terminal polyhistidine tag was affinity-purified. Purified rHG-ENG-2 was used as a binding target in biopanning experiments with a commercial phage-display combinatorial random 7-mer peptide library. Two stringent rounds of biopanning produced over 1,000 plaques with peptides that bound to rHG-ENG-2. Individual plaques are now being investigated to determine the number of unique peptides that bind to rHG-ENG-2 and to identify those peptides that inhibit the ability of rHG-ENG-2 to degrade carboxymethyl-cellulose. One promising 7-mer peptide that binds to and reduces the cellulolytic activity of rHG-ENG-2 has been identified to date.

Leaf spot of spinach seed crops in Washington state. L. J. DU TOIT and M. L. Derie. Dept. Plant Pathology, Washington State University, Mount Vernon R.E.U., Mount Vernon, WA 98273. Phytopathology 92:S21. Publication no. P-2002-0144-AMA.

Cladosporium variabile was thought to be the primary pathogen causing leaf spot of spinach seed crops in northwestern Washington. In 2001, isolates of *Stemphylium botryosum* from spinach in this region were shown to be pathogenic to spinach. In the greenhouse, *S. botryosum* caused more severe foliar symptoms on spinach than *C. variabile*. To examine the influence of pollen on leaf spot, spinach plants were inoculated in the greenhouse with 10^5 spores/ml of *C. variabile* or *S. botryosum*, with or without 10^6 spinach pollen grains/ml. Severity of leaf spot was greater in the presence of pollen, and enhancement of disease in the presence of pollen was greater for *S. botryosum* than *C. variabile*. Results illustrate the need to initiate protective fungicide applications prior to pollen shed. Chlorothalonil and mancozeb (registered on spinach seed crops in Washington) were evaluated in the greenhouse for control of *C. variabile*, *S. botryosum*, or *C. variabile* + *S. botryosum*. Both fungicides provided better control of *C. variabile* than *S. botryosum*. Therefore, fungicides with improved efficacy are needed for control of the leaf spot complex on spinach seed crops.

Fatty acid production by species of *Phytophthora*: Potential for identification. C.-H. Duan, M. B. Riley, and S. N. JEFFERS. Dept. Plant Pathology & Physiology, Clemson University, Clemson, SC 29634. Phytopathology 92:S21. Publication no. P-2002-0145-AMA.

To determine the potential of fatty acid methyl ester (FAME) analysis for identification of *Phytophthora* spp., fatty acids (FAs) produced by five isolates each of *P. cinnamomi*, *P. nicotianae*, *P. citrophthora*, *P. cactorum*, and *P. cryptogea* were examined. Effects of temperature (10, 15, 20, 25, 30, 35°C), mycelium age (4, 8, 12, 16 days), and medium (5% clarified V8 broth, 10% lima bean broth, 20% carrot broth, 0.25x potato dextrose broth, 0.25x Sabouraud dextrose broth) on FA production were investigated. In these species, 20 to 25 FAs were found, 15 FAs were common, and 5 FAs were dominant (14:0, 16:0, 18:2, 18:1, 20:5). Species produced different amounts of individual FAs, and culture parameters affected the amounts produced. However, the effects of culture parameters on FA production were not consistent among species. Isolates then were grown under standard conditions (0.25x PDB at 20°C for 4 days) and FAME profiles were analyzed. Isolates of the same species clustered except for *P. cryptogea*, which appears to represent a complex of species. Species clusters revealed by FAME were consistent with those generated after AFLP analysis.

Detection of *Pasteuria penetrans* in soil with *Pasteuria* specific PCR. Y. P. Duan (1), W. Hammer (2), Y. Huang (2), T. E. Hewlett (3), M. L. SMITHER-KOPPERL (3), J. H. White (3), and A. Ogram (1). (1) Soil and Water Science Dept.; Univ. of Florida, Gainesville, FL 32611; (2) Epcot Science, The Land, Walt Disney World, Lake Buena Vista, FL 32830; and (3) Entomos, LLC, Gainesville, FL 32608. Phytopathology 92:S21. Publication no. P-2002-0146-AMA.

Pasteuria spp. are obligate parasites of nematodes and have been demonstrated as effective biological control agents of plant parasitic nematodes. Methods of estimating *Pasteuria* endospore soil densities include a biological assay using juveniles in predried soil, and a method that extracts and detects surface coat proteins of endospores. These methods are time consuming and unreliable at spore densities of less than 10,000 endospores/g, and do not differentiate between species of *Pasteuria*. An approach using PCR with *Pasteuria*-specific primers was evaluated in sandy soil previously infested with *P. penetrans* at The Land Greenhouses, Epcot, Walt Disney World. DNA extracted directly from the sandy soil of experimental agronomic plots was subjected to PCR using *Pasteuria*-specific primers. Results from the PCR detection correlated well with historical applications of *P. penetrans* and microscopic observation of endospore-infested nematodes.

Weather variables related to *Gibberella zeae* perithecia development. N. S. DUFAULT (1), E. D. De Wolf (1), P. E. Lipps (2), L. V. Madden (2). (1) Dept. Plant Pathology, Pennsylvania State University, University Park, PA 16801; (2) Dept. Plant Pathology, Ohio State University/OARDC, Wooster, OH 44691. Phytopathology 92:S21. Publication no. P-2002-0147-AMA.

Information regarding weather variables related to *Gibberella zeae* perithecia development could be useful in developing wheat head scab forecasting systems. Perithecia development on colonized corn stalks and weather variables were monitored within wheat field environments near Wooster, OH in 2000, and State College, PA in 2001. The duration of stalk water content greater than 10 percent (DSW) was also recorded. Weather variables were paired with fungal development observations to identify variables associated

with perithecia development. Rate of perithecia production and number of perithecia per cm² of stalk tissue was greater at the OH compared to the PA location. At both locations, an increase in perithecia development was associated with extended periods of DSW, but the OH location had 10 additional days with average temperatures greater than 15°C and received 73 mm more rainfall. Predicting perithecia development of *G. zeae* using temperature and moisture variables may improve the accuracy of forecasting systems.

Suppression of early leaf spot through peanut-corn intercropping. L. E. DUFFIE (1), B. B. Shew (1), and M. A. Boudreau (2). (1) Dept. Plant Pathology, NC State Univ., Raleigh, NC 27695; (2) Dept. Biology, Warren Wilson College, Asheville, NC 28815. Phytopathology 92:S21. Publication no. P-2002-0148-AMA.

Intercropping peanut (p) and corn (c) was evaluated for its potential to suppress early leaf spot (ELS) of peanut, caused by *Cercospora arachidicola*. In 2000, treatments included nonsprayed peanut (*Arachis hypogaea*), sprayed peanut, intimate intercrop (c, p, c, p), and strip intercrop (2c, 4p, 4c, 4p, 2c). A new strip (4c, 4p, 4c, 4p) was added in 2001. Focal epidemics were initiated centrally in each plot. ELS incidence was determined weekly and disease gradients were examined in four directions. In both years, AUDPC's were greatest in nonsprayed monocrop, least in sprayed monocrop, and intermediate in strip intercrops. AUDPC's for intimate intercrop were less than those for nonsprayed monocrop in 2001, but not in 2000. The old strip treatment AUDPC was equivalent to that of sprayed monocrop in 2001. A treatment by distance interaction ($P < 0.05$) found during the last four sampling days of each year was attributed to low disease levels in the sprayed monocrop. A distance by direction interaction ($P < 0.05$) on day 49 in 2000 and day 46 in 2001 corresponded with rapid disease increase.

Human pathogen safety issues with compost extracts. B. DUFFY, S. Ravva, and L. Stanker. Foodborne Contaminants Research Unit, USDA-ARS, Albany, CA 94710. Phytopathology 92:S21. Publication no. P-2002-0149-AMA.

Compost watery extracts, also called compost teas, are gaining popularity among organic growers for their disease suppressive activity when applied to foliage. Production methods often call for organic amendments, such as molasses, to stimulate microbial populations. We have found that certain amendments also favor regrowth of human pathogenic bacteria, raising concerns about potential contamination of treated food crops. Using GFP and antibiotic-resistance marked outbreak strains, we found that *Escherichia coli* O157:H7 and *Salmonella enterica* serovar Thompson colonized strawberry after foliar application of contaminated compost extracts. Regrowth of these pathogens from 1 CFU/ml to populations over 4 log greater was dependent on the type of amendment and starter compost material used. Risk of pathogen regrowth was minimized by avoiding amendments altogether, or by adding anti-microbial plant compounds such as thymol, azadirachtin, clove and cinnamon oil.

Compromised plant health increases risk of opportunistic colonization by human pathogenic *Salmonella* and *E. coli* O157:H7. B. DUFFY (1) and J. M. Raaijmakers (2). (1) USDA-ARS, Albany, CA; (2) Plant Sciences, Wageningen University, The Netherlands. Phytopathology 92:S21. Publication no. P-2002-0150-AMA.

Human disease outbreaks traced to foodborne *Salmonella* and *E. coli* O157:H7 are increasing worldwide. These bacteria opportunistically colonize a variety of plants, with some degree of host specificity. Here we demonstrate that host health influences colonization. *S. enteritidis*, *S. typhimurium* and *E. coli* O157:H7 populations established at low levels on healthy wheat roots (10^3 cfu/g root) and strawberries (10^2 cfu/fruit). Human pathogen colonization was significantly increased on hosts infected with a fungal pathogen. Populations averaged 10^4 - 10^5 cfu/g root or fruit on wheat infected by *Rhizoctonia solani* or *Gauemannomyces graminis* var. *tritici* infected wheat, and on strawberry infected by *Botrytis cinerea*, depending on the bacterium-disease combination. None of the human pathogens affected fungal growth or plant disease development. Colonization of *Botrytis* infected tomato foliage will be discussed. Our results indicate that the increased risk of contamination of diseased crops should be considered in designing intervention strategies to reduce outbreaks.

Incidence and competitive interactions of *Botrytis cinerea* and other filamentous fungi quiescent in grape berries and dormant buds in central Washington state. F. M. DUGAN (1), S. L. Lupien (1), and G. G. Grove (2). (1) USDA-ARS Western Regional Plant Introduction Station, Pullman, WA 99164-6402; (2) Dept. Plant Pathology, Washington State University IAREC, Prosser, WA 99350-9687. Phytopathology 92:S21. Publication no. P-2002-0151-AMA.

Recovery of quiescent fungi from surface-disinfested, non-symptomatic grape berries and dormant buds demonstrated dominance of *Alternaria*, *Aureobasidium*, *Cladosporium*, and *Ulocladium*. Up to 78% of berries were colonized prior to harvest. *Botrytis cinerea* was recovered from 0.2-0.5% of berries just after fruit set, and 1.6-4.8% of over-wintered dormant buds. In laboratory inoculations of mature berries with *Alternaria alternata*, *A. infectoria*, *Aureobasidium pullulans*, *Cladosporium herbarum*, *C. clado-sporioides*, *Ulocladium atrum* and *B. cinerea*, only *Botrytis* was aggressive in rotting berries. Inoculations with *B. cinerea* alone, and in combination with the other species, demonstrated that prior occupation of wounds by the other species resulted in reduced lesion size compared to inoculation with *Botrytis* alone. We hypothesize that common, naturally occurring fungi constrain establishment of bunch rot via niche exclusion.

Control of *Phytophthora infestans* infection in potato with Gavel 75 DF™. A. E. DUTTLE, A. McFadden, R. L. Smith, G. Kemmitt, and B. Olson. Dow AgroSciences, 9330 Zionsville Rd., Indianapolis, IN 46268. Phytopathology 92:S22. Publication no. P-2002-0152-AMA.

Gavel 75 DF is a new fungicide introduced for use in North America for disease control in Potatoes during 2001. Gavel contains a new active ingredient, zoxamide plus mancozeb in an 1:8 blend. Zoxamide belongs to the Benzamide class of fungicides which have a unique mode-of-action that has demonstrated a high degree of efficacy for controlling pathogens in the Oomycete class of fungi. Research with Gavel 75 DF application timing, method, number of seasonal applications and product rotations on control of *Phytophthora infestans*. Effects on sporulation, incidence, severity and yield were evaluated. Results of greenhouse and laboratory trials indicate Gavel significantly reduced sporulation when applied to active late blight lesions. Gavel applied alone or in rotation with other fungicides reduced incidence and severity of infection equal to or greater than standard materials.

Comparison of pink snow mold incidence within a mixed stand of annual bluegrass and creeping bentgrass. P. J. DWYER (1), A. N. Kravchenko (2), and J. M. Vargas (1). Depts. (1) Plant Pathology and (2) Crop and Soil Science, Michigan State University. Phytopathology 92:S22. Publication no. P-2002-0153-AMA.

It has been suggested that annual bluegrass is more susceptible than creeping bentgrass to *Microdochium nivale*, although this has not been tested experimentally. Our research sought to identify differences in the amount of disease between the two grasses when growing in a mixed stand. Acetate grids were overlaid on an infected putting green, and disease patterns were traced. The selective herbicide isoxaflutole was applied to identify the two grasses; creeping bentgrass was temporarily discolored while the annual bluegrass was unaffected. The grids were again overlaid for tracing of the different grass areas within each plot. The grids were digitized and measured for the percent disease within each grass. Our measurements and analysis confirmed that annual bluegrass had significantly more disease than creeping bentgrass. Knowing which grass species is more likely to become infected may help turf managers monitor and treat areas more efficiently based on host susceptibility to *Microdochium nivale*.

Modeling survival of oospores of *Aphanomyces cochlioides*. A. T. DYER and C. E. Windels. Univ. MN, NWROC Crookston, MN 56716. Phytopathology 92:S22. Publication no. P-2002-0154-AMA.

The Weibull model ($S(t) = \exp(-rt^s)$; t - time; r, s - fitted parameters) was evaluated for predicting the cumulative, continuous temperature-time effects on survival of oospores of *A. cochlioides*, causal agent of root rot of sugar beets. Oospores were exposed to 35, 40, 45 and 50°C in water and then examined microscopically for viability. There was a strong relationship between the $\ln(r)$ and temperature ($R^2 = 0.98, P < 0.001$), and no relationship between "s" and temperature ($R^2 = 0.08, P = 0.48$). To determine if oospores displayed self-repair or enhanced susceptibility when exposed to high/low temperature fluctuations, they were examined for viability after each of 4, 24-hour cycles of 45°C for 4 h and 21°C for 20 h. Short exposures to high temperatures had a cumulative effect on viability. Survival of five isolates was examined to define model modifications for a variable population. A single fitted Weibull model was comparable to a composite of five individual models for defining population survival. In conclusion, parameter modifications for the Weibull model were defined, which predicted the cumulative effect of lethal temperatures on oospore survival for isolates and populations of *A. cochlioides*.

Soluble amino acid changes in potatoes in response to salt treatment and *Fusarium sambucinum* infection. S. I. DZENGELESKI, A. B. da Rocha, W. W. Kirk, and R. Hammerschmidt. Dept. of Plant Pathology, Michigan State

University, East Lansing, MI 48824. Phytopathology 92:S22. Publication no. P-2002-0155-AMA.

Discoloration has been observed in tubers from fields with high soil salt content and infestation with *F. sambucinum*. The objective of this study was to determine if discoloration is associated with changes in amino acids related to osmotic stress (proline, PRO) and involved in phenolic compounds production (tyrosine, TYR; phenylalanine, PHE). A field experiment was carried out in East Lansing, MI where half of the potatoes (cv. Atlantic) were inoculated with *F. sambucinum* prior to planting. At 46 days after planting, NaCl was added to the soil weekly for 5 weeks in concentrations ranging from 0-85g/L. Leaf samples from NaCl treated plants had on average a two-fold increase in PRO and PHE, compared to control plants. Samples from inoculated and NaCl treated plants had the same amino acid changes as NaCl treated, plus an increase in TYR. Tuber tissue had more PRO in NaCl treated plants, while treatment with both NaCl and *Fusarium* led to a two-fold increase in TYR and PHE content.

Plants, Pathogens, and People: A web site designed to introduce students to important plant diseases. D. M. EASTBURN and C. J. D'Arcy. Dept. Crop Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S22. Publication no. P-2002-0156-AMA.

The internet can be used to deliver educational materials in a manner that fosters student participation and inquiry. The Plants, Pathogens, and People web site helps students learn about important plant diseases through active learning and inquiry based instruction. The site presently includes information on three diseases, each chosen to highlight an important concept or issue in plant pathology. Late blight of potato (*Phytophthora infestans*) is used to demonstrate components of the plant disease triangle. Dutch elm disease (*Ophiostoma ulmi*) illustrates the advantages and disadvantages of monocultures. Crown gall (*Agrobacterium tumefaciens*) introduces the benefits and risks of genetic engineering. Virtual laboratory activities are available that allow students to apply the scientific process and explore methods of experimentation. Students can record their methods, results, and conclusions in a lab notebook that can be emailed to an instructor. These experiences introduce students to scientific and societal issues related to plant pathology, and increase their understanding of science and the agricultural system.

Loblolly pine decline, *Leptographium* spp. and root -feeding insects. L. ECKHARDT (1), J. Jones (1), N. Hess (2) and E. Carter (3). (1) Department of Plant Pathology and Crop Physiology, Louisiana Agricultural Experiment Station, Louisiana State University Agricultural Center, Baton Rouge, LA; (2) USFS, FHP, Pineville, LA; (3) USFS, SRS, Auburn, AL. Phytopathology 92:S22. Publication no. P-2002-0157-AMA.

Our data show a correlation of *Leptographium* spp. with symptoms of loblolly decline, insect and vegetation densities, and deteriorated roots. *Leptographium* species are associated with root-feeding insects that attack pine and are commonly present during pine decline and mortality in the United States. The relative importance of the beetles and fungi in the decline is still unclear. In the southeastern US, loblolly decline stands are more likely to contain *Leptographium* species in their root systems and be more vulnerable to southern pine beetle than symptomless stands. Decline plots in Alabama have *Leptographium* species associated with both roots and soil as well as root-feeding insects. Soil analysis shows that bulk density and total porosity are not growth limiting. Inoculation experiments indicate that *Leptographium* is capable of killing seedlings.

Evaluation of mulch type and placement for management of crown rot of hosta. B. A. EDMUNDS, M. L. Gleason, and S. N. Wegulo. Dept. Plant Pathology, Iowa State University, Ames, IA 50010. Phytopathology 92:S22. Publication no. P-2002-0158-AMA.

Three types of organic mulch were tested for ability to reduce spread of *Sclerotium rolfsii* var. *delphinii* on hosta at an Iowa field site. Seventy 1.2-m-diameter plots were delimited by aluminum flashing. On 1 Jun 2001 two hosta plants (cv. Golden Tiara) were transplanted, 30 cm apart, into each plot. Experimental design was a randomized complete block with 10 replications. Mulch (medium pine chunk bark, hardwood mulch, or pine needles) was added to the plots in two placement treatments, either completely mulched (both plants mulched) or partially mulched (one plant mulched, the other not mulched). Completely non-mulched plots were included as controls. One plant in each plot was inoculated on 5 Jul with pathogen-infested carrot disks. Disease severity on non-inoculated plants was assessed thirteen times in each plot between 9 Jul and 18 Aug. Plants in completely mulched pine bark and hardwood plots showed more ($P < 0.05$) disease than plants in either partially mulched plots of these materials or non-mulched plots. Plants in pine needle mulch remained symptomless regardless of mulch placement.

Primary germ tube formation by conidia of *Blumeria graminis hordei* on leaf epidermal cells of *Hordeum vulgare* and *Arabidopsis*. H. H. Edwards. Dept. Biol. Sci., Western Illinois University. Phytopathology 92:S23. Publication no. P-2002-0159-AMA.

Within 1 h post inoculation a primary germ tube (PGT) emerges from the conidium quickly making contact with the leaf surface (cuticle). As revealed by the TEM an appressorial-like swelling develops where contact is made. Beneath this swelling a peg [designated the cuticular peg (CP)] forms that breaches the cuticle but not the host cell wall. The CP undergoes the following development that is completed by 10 h. The outer portion of the PGT wall becomes electron dense where the CP will form. This outer part of the cell wall expands into the cuticle as it is digested away, forming the CP. The CP becomes separated into a loose fibrillar structure whereas the inner portion of the cell wall retains an appearance similar to the rest of the PGT inner wall. There is no further development of the CP. PGT development is similar in barley (host) and *Arabidopsis* (nonhost). In the barley epidermal cell beneath the PGT, cytoplasmic aggregation occurs leading to the formation of an electron-dense papilla detectable as early as 3 h. In *Arabidopsis* there is less cytoplasmic aggregation resulting in a very thin, nonelectron-dense papilla.

Identification of citrus genes responding to bacterial canker infection. B. El YACOUBI, T. E. Kornahk, J. M. Davis, and D. W. Gabriel. Plant Molecular and Cellular Biology Program, University of Florida, Gainesville, FL 32611-0680. Phytopathology 92:S23. Publication no. P-2002-0160-AMA.

Bacterial canker is among the most serious diseases of citrus worldwide; symptoms include hyperplastic cell divisions, host cell death and premature fruit drop. We used differential display to identify citrus genes differentially regulated during early stages of infection by *Xanthomonas citri* strain B69. Gene expression profiles of leaves inoculated with B69 were compared with those of leaves inoculated with an isogenic, non-pathogenic *pthB* knockout mutation. Twenty-seven partial cDNA clones were identified that appeared to be differentially regulated in response to B69 infection. Some of these citrus canker responsive (ccr) clones exhibited high sequence similarity to pathogenicity related (PR) genes, signal transduction pathway components, and other defense related genes. Northern blot analyses are being performed to confirm that these clones are differentially expressed in canker-infected leaves.

Spatial and temporal development of *Fusarium* Head blight epidemics on winter wheat in Ohio. S. M. EL-ALLAF, L. V. Madden, and P. E. Lipps. Dept. of Plant Pathology, The Ohio State University/OARDC, Wooster, OH 44691. Phytopathology 92:S23. Publication no. P-2002-0161-AMA.

The spatial and temporal development of *Fusarium* head blight (scab) caused by *Fusarium graminearum* (*Gibberella zeae*) was studied in two fields at the Ohio Agricultural Research and Development Center in Wooster during the 2001 crop season. In each field, three transects were established, with 15 sample points per transect, spaced at 0.75 -m intervals. Disease assessments were made twice a week from 11 June to 26 June. At each sample point, scab incidence and severity were recorded in a 0.3-m sub-transect across the plant rows. The Gompertz model was appropriate for describing incidence and severity data sets. The beta-binomial distribution fitted 75% of the data sets ($P > 0.05$), based on a likelihood ratio test. Disease heterogeneity in individual data sets was measured using the index of dispersion (variance ratio), C(alpha) test, and estimated theta parameter of the beta-binomial. Using these indices, overdispersion was detected in more than 90% of the data sets, which indicated that diseased heads were aggregated to highly aggregated within wheat fields.

Effect of biocontrol agents and fungicides on *Fusarium* head blight development on winter wheat in Ohio. S. M. EL-ALLAF, P. E. Lipps, and L. V. Madden. Dept. of Plant Pathology, The Ohio State University/OARDC, Wooster, OH 44691. Phytopathology 92:S23. Publication no. P-2002-0162-AMA.

Two biocontrol agents (TrigoCor 1448 and USDA/OH 182.9) and three foliar fungicides (Folicur, AMS21619, and BAS 505) were tested to determine their effect on the development of *Fusarium* head blight (scab) on the susceptible wheat cultivar Elkhart during the 2001 crop season. Tests were conducted in replicated, inoculated, and mist irrigated plots. The biological agents and foliar fungicides were applied at flowering. *Fusarium* head blight incidence and severity were assessed twice a week from 11 June to 26 June at 15 locations per plot. The treatments exhibited differences in epidemic development. The epidemic rate (r), area under disease curve (AUDPC), and maximum disease (Y_{max}) for both incidence and severity in

plots treated with fungicides AMS21619 and BAS 505 were low and significantly ($P = 0.05$) different from untreated plots. On the other hand, r , AUDPC, and Y_{max} in plots treated with the biological agents were high and not significantly different from control plots. The results indicate the AMS21619 and BAS 505 fungicides have greater potential for management of scab than the other treatments tested.

Ultrastructure studies of potato virus Y (PVY) in Egypt. A. A. EL-AMRETY (1), M. I. El-Refari (2), M. I. El Khalily (2), and E. F. Mohamed (2). (1) Dept. Virology, Plant pathology Res. Inst., ARC, Giza, Egypt; (2) Dept. Agriculture Botany Fac. Agric, Cairo Univ., Fayoum, Egypt. Phytopathology 92:S23. Publication no. P-2002-0163-AMA.

An examination of ultrathin section of leaves of potato virus Y (PVY)-infected *Nicotiana tabacum* CV. White burly revealed some alterations in the ultrastructure of chloroplasts, mitochondria, and nuclei. The mesophyll cells of young leaves showed several ultrastructural changes due to virus infection. PVY induced cytopathological inclusion bodies in infected tobacco plants. Examination of epidermal stripes stained with specific stains, through light microscope, showed that, cytoplasmic inclusion bodies induced by PVY are of proteinacious nature. Virus aggregates were detected in cytoplasm. Chloroplasts became distorted and misshapen. In addition degeneration of chloroplasts was observed. Mitochondria were affected due to virus infection, which caused the formation of vacuole-like vesicles in irregular shape in mitochondria, and disarranged. In addition to the mitochondria was malformaed and its outer membrane was ruptured. Nucleus also affected with virus infection, which caused rupture of its membrane and degeneration of the nucleus. Key Word: Potato, PVY, cytopathological studies.

Evaluation of inoculation methods to determine differences in resistance to *Fusarium graminearum* in wheat genotypes. J. S. ENGLE, P. E. Lipps, and L. V. Madden. Dept. Plant Pathology, Ohio State University, Wooster, OH 44691. Phytopathology 92:S23. Publication no. P-2002-0164-AMA.

Greenhouse assays that detect type I and type II resistance in wheat to *F. graminearum* are needed. Disease severity using a central floret inoculation in the greenhouse was moderately correlated to field disease severity in previous studies. Genotypes with known reactions in the field were tested in the greenhouse using four inoculation techniques to evaluate resistance type. Disease incidence and severity assessments were recorded and used to calculate AUDPC and rate of disease increase. Disease assessments for severity or incidence in the greenhouse identified different genotypes as having a resistant response. Standard greenhouse inoculations, a central floret injection to screen for type I resistance and atomizing the surface of the spike to screen for type II resistance, did not detect a resistant response in certain genotypes known to have type I or II resistance in the field. These results indicate a disparity between greenhouse disease assessment method or inoculation technique and putative resistance type from field screening.

Effect of wheat floral structure extracts and endogenous compounds on the growth of *Fusarium graminearum*. J. S. ENGLE, P. E. Lipps, T. L. Graham, and M. J. Boehm. Dept. Plant Pathology, Ohio State University, Wooster, OH 44691. Phytopathology 92:S23. Publication no. P-2002-0165-AMA.

Fusarium head blight is more severe when infection occurs during anthesis, indicating anthers may be an important route for infection. Choline and betaine have been extracted from wheat floral parts and enhanced *F. graminearum* infection *in vivo* in previous work. Growth of *F. graminearum* was examined on agar amended with floral part extracts of genotypes with various resistance levels. Results indicated no significant effect of anther, palea, or lemma extracts on radial growth compared to controls. The effect of choline, betaine, and an equal molar mixture at 10 to 1000 micro molar concentrations on spore germination and hyphal growth *in vitro* was examined. Spore germination was not significantly affected when compared to controls. Hyphal growth was also not significantly affected by choline or betaine when compared to controls. Equal molar mixtures showed significant inhibition of growth when compared to controls at high concentrations of betaine. Results indicate that endogenous compounds in floral parts as evaluated in this study may not be correlated with wheat resistance to *F. graminearum*.

Field validation studies on a disease predictive model for *Phomopsis* cane and leaf spot of grape. O. Erincik, L. V. Madden, L. L. WILSON, and M. A. Ellis. Department of Plant Pathology, OARDC, The Ohio State University, Wooster, OH 44691. Phytopathology 92:S23. Publication no. P-2002-0166-AMA.

A generalized Beta model for predicting grape leaf and cane infections by *Phomopsis viticola* was validated by inoculating 'Seyval' and 'Catawba' vines during natural rain events in the field during 2000 and 2001. Inoculations were initiated during the first rain event after vine-growth reached Eichhorn-Lorenz growth stage 09 and were repeated during rain events throughout the growing season. Average temperature and hours of wetness for each event and inoculation were recorded and used in the model equation to predict disease severity on leaves and internodes. Correlation coefficients between observed disease severities following field inoculations and predicted disease severities for both cultivars were high (between 0.71 and 0.81). These results indicate that the model reliably predicted leaf and cane infection on both cultivars over a wide range of wetness durations and temperatures. The model may be useful in developing a disease forecasting systems for *Phomopsis* cane and leaf spot on grapes.

First report of the pycnidial state of *Phaeoconiella chlamydospora*, a causal agent of black measles (esca) and Petri disease in California vineyards. A. ESKALEN, S. N. Rooney, and W. D. Gubler. Dept. of Plant Pathology, University of California, Davis, CA 95616. Phytopathology 92:S24. Publication no. P-2002-0167-AMA.

Phaeoconiella chlamydospora is considered to be the primary pathogen causing esca and Petri disease. Previous studies have shown that *Pa. Chlamydospora* has the ability to produce pycnidia in culture. However, these structures had not been documented in California vineyards. Spore traps were placed in selected vineyards throughout the state. Spores of *Pa. Chlamydospora* were trapped from coastal counties and correlated with rainfall events. Grapevine tissues were collected from vineyards where spores were previously trapped. Tissues were washed and cultured on media. Results showed that spores of *Pa. chlamydospora* were found on grapevine cordons and pruning wounds of 2-3-year-old spurs. Pycnidia were observed beneath bark and on old pruning wounds. These pycnidia were similar to those that were artificially produced in culture. Spores from pycnidia were shown to be viable. Pathogenicity tests of spores from artificially and naturally produced pycnidia were compared. ITS regions from pycnidiospores were compared to that of known isolates of the *Pa. chlamydospora*.

Development and use of degree-day models to improve management of corn flea beetles in the Stewart's disease pathosystem on corn. P. D. ESKER and F. W. Nutter, Jr. Iowa State University. Phytopathology 92:S24. Publication no. P-2002-0168-AMA.

The ability to accurately predict emergence of corn flea beetles (CFB) *Chaetocnema pulicaria* is important for timely application of insecticides to reduce the risk of Stewart's disease of corn. Using a developmental temperature threshold of 16°C, degree-days (DD) were used to determine the emergence of the overwintering, first, and second generations of CFB adults at six locations in Iowa. In 1999, overwintering CFB adults emerged between 38 and 64 DD, the first summer generation was observed between 290 and 360 DD, and the second summer generation emerged between 590 and 620 DD. In 2000, emergence of CFB adults occurred between 44 and 91 DD, the first summer generation was observed after approximately 320 DD, and the second summer generation occurred between 630 and 650 DD. Compared to other years, there was a three-week delay in accumulation of the first DD in 2001 and CFB was not found throughout much of the growing season. This information is being tested in field insecticide trials to determine if a DD model can be used to improve the management of Stewart's disease of corn.

The effect of previous crop residues and *Bacillus subtilis* on root rot of dry bean. C. ESTEVEZ DE JENSEN and J. A. Percich. Plant Pathology, University of Minnesota, St Paul, MN. Phytopathology 92:S24. Publication no. P-2002-0169-AMA.

Residues of canola, dry bean, pea, potato, soybean and sunflower were evaluated to sustain *Fusarium solani* f. sp. *phaseoli* (Fsp) and *Rhizoctonia solani* (Rs), the bean root rot pathogens. Each crop was grown in naturally infested soil and after harvest the residue was left on the soil and removed in the spring. One half of the residue was used for pathogen isolation and the remaining was pasteurized (45 C), inoculated with either Fsp or Rs, and incubated at 24°C for 1 mo. The residue was then inoculated with *Bacillus subtilis* GBO3 (Bs) (0.08 g/kg), mixed with pasteurized soil and sown with dry bean. Isolations indicated that Fsp survived in residues of dry bean, canola, soybean and peas and Rs in dry bean, potato and sunflower. Dry bean growing in contact with colonized Fsp residue of dry bean, canola and soybean resulted in root rot severity (DS) of 6, 6.3 and 6.7, respectively. Rs colonized residue of dry bean, potato and sunflower, resulted in dry bean DS of 6, 5.2 and 4.7, respectively. Bs applied on dry bean residue colonized with either Fsp or Rs decreased DS to 4.5 and 2.5, respectively. Residue management may enhance root rot control.

The effect of *Bacillus subtilis* and *Rhizobium* dry bean seed inoculation on bean root rot. C. ESTEVEZ DE JENSEN (1), J. A. Percich (1), and P. H. Graham (2). (1) Plant Pathology Dept.; (2) Soil, Water and Climate Dept, Univ. of Minnesota, St. Paul, MN. Phytopathology 92:S24. Publication no. P-2002-0170-AMA.

Irrigated dry bean production in central Minnesota is threatened by declining yields due to root rot caused by *Fusarium solani* f. sp. *phaseoli* and *Rhizoctonia solani*. *Bacillus subtilis* (strains GBO3, MBI600 and GB34) alone or in combination with *Rhizobium tropici* UMR 1899 and *R. leguminosarum* RCR 3622, were evaluated for reducing disease severity (DS) in pasteurized field soil inoculated with both pathogens simultaneously. The controls were standard seed treatment (SST) of Captan + streptomycin + Lorsban and, untreated-seed in pathogen free soil. The best treatment (GBO3 + RCR 3622) lowered DS (scale 1-9) to 4.0 when compared to the SST (DS=6.2). Also, seed treated with GBO3 or RCR 3622 had lower DS (5 and 4.5, respectively) than the SST. There was greater acetylene reduction activity for plants treated with GBO3 + RCR3622 (34) compared to the SST (22) or either treatment alone (26 and 24 umol/hr/plant, respectively). Bs GBO3, GB34 and MBI600 reduced DS and increased plant biomass compared to the SST. A combination of *Bacillus subtilis* and *Rhizobium* offers promise to control bean root rot.

Timing of inoculations of dryland wheat plots and the effect on *Fusarium graminearum* infection. C. K. EVANS and R. Dill-Macky. Dept. Plant Pathology, University of Minnesota, St. Paul, MN 55108. Phytopathology 92:S24. Publication no. P-2002-0171-AMA.

Eight wheat (*Triticum aestivum*) genotypes were assessed for reaction to FHB in a dryland inoculation study. The test was a randomized complete split-split block design. Main-plots were timings of inoculation (TOI), sprayed at anthesis (SAA), 3 days post-anthesis (DPA), and 6 DPA. Sub-plots consisted of two spray-inoculation treatments, inoculated once or twice, and sub-sub plots were the eight wheat genotypes. Significant differences were found among TOI treatments for % FHB severity (7.1 SAA, 7.7 sprayed 3 DPA, and 0.6 sprayed 6 DPA with *l.s.d.*_{0.05}=2.42) and deoxynivalenol accumulation (3.0 parts-per-million [ppm] SAA, 3.0 ppm sprayed 3 DPA, and 0.3 ppm sprayed 6 DPA with *l.s.d.*_{0.05} = 0.60). Average % FHB severities of wheat genotypes were 0.2 on BacUp, 0.2 on ND2710, 0.2 on Ingot, 0.2 on Forge, 0.8 on Oxen, 1.4 on Parshall, 5.3 on Norm, and 8.6 on Wheaton (*l.s.d.*_{0.05}=1.07). Our data demonstrate that dryland inoculations of wheat can be useful for screening germ plasm for reaction to FHB.

Biological control of *Monilia* pod rot in the field using epiphytic bacteria isolated from cocoa's pods. C. E. FALCONI, G. T. Paez, A. R. Oleas, and V. R. Yanez. Department of Agropecuarian Research, Faculty of Agropecuarian Science. The Army Polytechnic School, Telefax (593) 2 02 870 193, P.O. Box. Sangolqui, Ecuador, E-mail: cfalconi@espe.edu.ec. Phytopathology 92:S24. Publication no. P-2002-0172-AMA.

Experiments were conducted to determine the effect of epiphytic bacteria isolated from cocoa pods on development of *Monilia* pod rot. The research was carried out in a cocoa plantation (cv. Tenguel) located on the Oreacao experimental farm, Los Rios, Ecuador. The microorganisms evaluated were strains of *Pseudomonas putida*, *Pseudomonas cepacia* and *Bacillus subtilis*. The experiment was established in a randomized complete block with three replications. Five treatments were included: the three strains of antagonistic bacteria that consistently inhibited mycelial growth of *Monilophthora roreri* in petri plate inhibition assays, a positive control (0.05% sucrose), and an untreated control. The concentration of the bacteria inoculum was 2 × 10⁸ cfu/ml. The antagonists were applied every 2-3 weeks from March to August, 2001, and disease severity was measured routinely throughout the season. Disease severity data were summarized by computing the area under disease progress curve (AUDPC). Based on analysis of variance of AUDPC values, the strain of *B. subtilis* and *oP.cepacia* reduced disease by 62 and 55%, respectively, in comparison with the untreated control. Acknowledgments: This research was sponsored by The Programa de Modernizacion de los Servicios Agropecuarios (PROMSA).

Correlation of soil eubacterial diversity with variation in farming system and time; *Burkholderia* as a model system. D. C. FARGO and F. J. Louws. Dept. of Plant Pathology, North Carolina State University, Raleigh, NC 27695. Phytopathology 92:S24. Publication no. P-2002-0173-AMA.

The impact of five farming systems: successional, forestry, integrated crop-animal, organic, and conventional (BMP), on the dynamics of soil microbial populations were investigated. Extracted DNA from 500 soil samples collected from the various farming systems over 4 years was examined for

total eubacterial diversity. T-RFLP analysis of these extracted templates revealed a general, but not absolute, correlation of bacterial population structure and farming system. We investigated the population dynamics of culturable and non-culturable soil *Burkholderia* as a model system to enhance the understanding of bacterial population dynamics. The dynamics of total soil *Burkholderia* were studied at the species, genomovar, and sub-genomovar level using a series of genomovar-specific rDNA PCR primers. A subset of these PCR-amplified products was examined in more detail by DNA sequencing to confirm the specificity of the primers on environmental samples. *Burkholderia cepacia* complex genomovars I, II and V were identified from the soil extracts using this system. The diversity of rapidly growing cultureable *Burkholderia* was examined from the same 500 soil samples using BOX-PCR techniques and the diversity of BOX-PCR fingerprint patterns was investigated in relation to farming system and temporal changes.

Molecular phylogenetic relationships among *Septoria* species from woody perennials and development of markers for species identification by PCR. N. FEAU (1), R. C. Hamelin (2), and L. Bernier (1). (1) CRBF, Université Laval, Quebec, QC, G1K 7P4; (2) Canadian Forest Service, Ste-Foy, QC, G1V 4C7. Phytopathology 92:S25. Publication no. P-2002-0174-AMA.

Septoria species are usually distinguished by their occurrence on different host plants and by characters such as conidial size. However, wide variation and overlap in morphological characters has led to confusion in species concepts. This is the case for *S. musiva* and *S. populicola* which cause a leaf spot and stem canker disease of *Populus* species and hybrids in North America. We recovered 12 species of *Septoria* from various woody perennials, and carried out phylogenetic analyses using nuclear rDNA, the mitochondrial rDNA small subunit (SSU) and the beta-tubulin gene. A 499 bp group-I intron was identified and located in the SSU rDNA gene of *S. musiva*. Pairwise alignment of the ITS sequence from *S. musiva* and *S. populicola* nuclear rDNA revealed the occurrence of a few interspecific polymorphisms which were used to design specific PCR primers. In spite of the presence of the group-I intron in the SSU rDNA gene of *S. musiva*, phylogenetic inference about *Septoria* species revealed close relationships between species from poplars and willow.

Management of soilborne pests in strawberry and vegetable plasticulture systems. L. M. FERGUSON (1), F. J. Louws (1), G. E. Fernandez (2), D. C. Sanders (3), D. W. Monks (3), Z. Pesic-VanEsbroeck (1), E. B. Poling (3), P. M. Brannen (4), J. P. Smith (5), C. D. Safley (6), and O. B. Sydorovych (6). (1) Plant Pathology, North Carolina State Univ., Raleigh NC; (2) Hort. Science, NCSU, Plymouth, NC; (3) Raleigh, NC; (4) Plant Pathology, Univ. of Georgia, Athens, GA; (5) Clemson Ext. Service, Clemson Univ., Lexington, SC; (6) Agric. & Resource Economics, NCSU, Raleigh. Phytopathology 92:S25. Publication no. P-2002-0175-AMA.

Soilborne pathogens, nematodes and weeds can reduce yields when strawberry or vegetable crops are grown in non-fumigated fields. A team was formed to evaluate methyl bromide alternatives and engage growers, industry, researchers and extension agents in resolving problems of production without fumigation. Strawberry studies have included tests of alternatives, application methods, and cover crop/compost systems. We have also emphasized clean plant production to prevent introduction of pathogens into fumigated fields. Research in tomato, pepper and cucurbit production has focused on alternative chemicals, compost-based systems, and weed management to minimize losses. We will emphasize findings related to yield and quality, disease severity, and nematode/weed control.

Characterization of *Phytophthora infestans* in Michoacan, Mexico. S. P. FERNANDEZ-PAVIA (1), G. Rodriguez-Alvarado (1), E. Garay-Serrano (2), A. K. Sturbaum (2), N. J. Grünwald (3), J. M. Sanchez-Yañez (1), and H. Lozoya-Saldaña (2). (1) IIAF, Univ. Michoacana de San Nicolas Hidalgo, Morelia, Mexico, 58240; (2) PICTIPAPA, Metepec, Mexico; (3) USDA ARS, Prosser, WA. Phytopathology 92:S25. Publication no. P-2002-0176-AMA.

The state of Michoacan represents 8% of the total potato production in Mexico. Despite important yield reduction by potato late blight, information about *Phytophthora infestans* populations in the region is limited. Twenty-nine *P. infestans* isolates collected from cultivated potato fields near Patzcuaro, Michoacan, were analyzed for genetic variation by mating type, allozyme loci (*Gpi* and *Pep*) and DNA fingerprinting. Mitochondrial DNA haplotype analysis is in progress. To detect oospore formation in nature, potato leaflets with multiple lesions were collected where both mating types were found. Seventy five percent of potato leaflets contained oospores. Genotypic diversity was detected using DNA fingerprinting and allozyme analysis.

Mixed infection of papaya ringspot virus, zucchini yellow mosaic virus and papaya bunchy top affecting papaya (*Carica papaya* L.) in Puerto Rico. M. Ferwerda-Licha. USDA-ARS-Tropical Agriculture Research Station. Phytopathology 92:S25. Publication no. P-2002-0177-AMA.

Papaya trees with severe viral symptoms were observed in fields at two locations in Puerto Rico. Samples from each field were tested with ELISA for the presence of viruses using antisera to: *Papaya Mosaic Virus* (PMV), *Papaya Ringspot Virus* (PRSV), *Tobacco Ringspot Virus* (TRV), *Tomato Spotted Wilt* (ToSW), *Impatiens Necrotic Spot Virus* and potyvirus group antisera. Two of the samples tested positive for the potyvirus group test and negative for PRSV. The material was re-tested with antisera to PRSV, *Watermelon Virus-2* (WMV-2) and *Zucchini Yellow Mosaic Virus* (ZYMV). Results were negative for WMV-2 and positive for PRSV and ZYMV. A second set of 45 samples was collected at one of the locations. These plants had viral symptoms as well as symptoms characteristic of papaya bunchy top (PBT). All samples were tested for PRSV, ZYMV, and PBT. For PRSV 34% were positive and 55% were negative while 11% had elevated titers. For ZYMV 53% were positive and 27% were negative while 20% had elevated titers. Eighty-four percent of the samples tested positive for PBT.

New diagnostic assay based on DNA array for identification and detection of five bacterial pathogens of potato. A. Fessehaie (1), S. H. De Boer (2), and C. A. LEVESQUE (1). (1) Agriculture and Agri-Food Canada, Eastern Cereal and Oilseed Research Centre, 960 Carling Ave, Ottawa, ON, K1A 0C6, Canada; (2) Canadian Food Inspection Agency, Centre for Animal and Plant Health, 93 Mount Edward Rd, Charlottetown, PE, C1A 5T1, Canada. Phytopathology 92:S25. Publication no. P-2002-0178-AMA.

By sequencing the 3' end of the 16S rDNAs and the intergenic spacer (IGS) regions of pectolytic erwinias (*E. carotovora* subsp. *atroseptica* and *carotovora* and *E. chrysanthemi*) and using equivalent Genbank data of other potato pathogens (*Clavibacter michiganensis* subsp. *sepedonicus*, *Ralstonia solanacearum*), oligonucleotides were designed and formatted into an array by pin spotting on nylon membranes. Using conserved ribosomal primers, bacterial DNA was amplified and labelled simultaneously by PCR with digoxigenin-dUTP. Hybridization of amplicons to the array revealed different patterns that were distinct for each species and subspecies tested. Hybridization of amplicons was generally restricted to the appropriate homologous oligonucleotides and cross-hybridization with heterologous oligonucleotides was rare. The assay was used successfully with infected potato samples.

Mutants of *Fusarium verticillioides* impaired in fumonisin biosynthesis. J. E. FLAHERTY and C. P. Woloshuk. Dept. of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S25. Publication no. P-2002-0179-AMA.

Fusarium verticillioides is a pathogen of maize that produces fumonisins, a class of mycotoxins linked to human and animal diseases including cancer. Toward achieving a better understanding of the genetic regulation of fumonisin biosynthesis, we have used a restriction enzyme-mediated integration (REMI) procedure to generate mutants disrupted in fumonisin production. The REMI vector, WE67, contains the promoter region of *FUM5*, the polyketide synthase gene involved in fumonisin biosynthesis, fused to the GUS gene, resulting in a reporter of *FUM5* transcription. Over 1,000 REMI transformants were grown for one week on cracked maize kernels and screened for GUS activity. Two transformants that failed to produce detectable levels of GUS, exhibited >100-fold reduction in fumonisin accumulation compared to a transformant control. The transformants grew and conidiated similar to the wild-type strain. Plasmid rescue and complementation experiments are in progress to determine the genes disrupted in these mutants.

Identification and characterization of a PACC-like gene in *Fusarium verticillioides*. J. E. FLAHERTY and C. P. Woloshuk. Dept. of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S25. Publication no. P-2002-0180-AMA.

Fusarium verticillioides is an economically important pathogen of corn due to its ability to produce fumonisins, a class of mycotoxins known to cause animal disease and promote cancer. In the maize kernel, where pH is between 6.5 and 7, fumonisin biosynthesis by *F. verticillioides* requires a functional *FCC1* gene. Deletion mutants of *FCC1* fail to produce fumonisin on maize but produce wild-type levels on synthetic medium buffered at acidic pH (<5). To further understand the role of pH in fumonisin biosynthesis, we have cloned a PACC-like gene (*PACC-Fv*) from *F. verticillioides*. PACC genes encode the major transcriptional regulator of several pH-responsive pathways in filamentous fungi. Growth of the *PACC-Fv* disruption mutant was severely impaired at alkaline pH (8.4) while no

adverse effects on growth or conidiation was observed at acidic pH. Furthermore, the *PACC-Fv* mutant produced fumonisin on corn and on a synthetic medium buffered at acidic pH. While previous evidence indicates that pH is an important determinant for fumonisin biosynthesis, our data suggest that *PACC-Fv* is not directly involved.

Range grasses: Potential reservoirs for fungal pathogens. J. M. FLEER and J. E. Partridge. Dept. Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722. Phytopathology 92:S26. Publication no. P-2002-0181-AMA.

Twelve species of range grasses with leaf blight symptoms were collected in the summer of 2000 from corn and wheat field borders in the Sandhills region of Nebraska. Many fungal species were isolated from leaf blight symptoms of the range grasses and established in pure cultures. An isolate of *Bipolaris sorokiniana* from Switchgrass (*Panicum virgatum*) was selected for further study. The culture was grown on corn meal agar. Spores were washed from the agar plates and inoculated onto agronomic crops including: corn, wheat, sorghum, barley, oats, and ryegrass to determine host range. Corn and ryegrass were not; however, barley and oats were susceptible to this pathogen; sorghum and wheat were inconclusive. The isolate was biotyped for race on barley using differential varieties. We conclude that range grasses are potential reservoirs of fungal pathogens affecting crop species.

A nested PCR protocol to determine if *Sphaeropsis sapinea* is present in asymptomatic *Pinus nigra* tissue. J. L. FLOWERS, J. R. Hartman, and L. J. Vaillancourt. Dept. of Plant Pathology, University of Kentucky, Lexington, KY 40546. Phytopathology 92:S26. Publication no. P-2002-0182-AMA.

Surveys of diseased and asymptomatic *Pinus nigra* revealed that latent infections of asymptomatic tissues by the casual agent of pine tip blight disease, *Sphaeropsis sapinea*, were common. However, culturing asymptomatic pine tissues to isolate the fungus destroys the shoot, preventing further studies of the latent infections. A nested PCR protocol was developed to test for *S. sapinea* in asymptomatic terminal buds and bark samples. This protocol results in comparatively little harm to the latently infected shoot compared to the culturing technique. *S. sapinea* specific primers were developed from the ITS region of the rDNA gene cluster and the specificity of the primers was tested with other fungi that were isolated from *P. nigra* tissues in this region. *S. sapinea* DNA was the only DNA amplified by using these specific primers. Theoretically, this protocol can detect as little as one fungal genome in a pine tissue sample. This nested PCR protocol will make it possible to study latent *S. sapinea* infections in pine shoots which will lead to a better understanding of pine tip blight.

Wood chip mulch as a source of *Verticillium dahliae*. G. L. FOREMAN, D. I. Rouse, and B. D. Hudelson. Dept. Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S26. Publication no. P-2002-0183-AMA.

Non-composted wood chips from a *Verticillium dahliae*, (*Vd*)-infested sugar maple, (*Acer saccharum*), were placed in mesh bags and placed in non-infested wood chips to simulate a typical mulch layer found in an urban landscape. Bags were removed at 0, 2, 4, 8, 16, 32, 62 and 122 days after the start of the experiment and chips from these bags were placed onto Sorenson's medium. The number of chips showing growth of *Vd* was determined. After 122 days, *Vd* grew from 19% of the chips, compared to 99% at day 0. In a separate experiment, 27 day-old eggplants, (*Solanum melongena*), were transplanted into 10.2 cm pots filled with a mixture of Scott's potting medium and 300 mL of either *Vd*-infested or non-infested wood chips. Eggplants treated with infested chips wilted after 14 days, and *Vd* was recovered from 89% of these plants. Eggplants treated with non-infested chips remained asymptomatic and *Vd* was not recovered. A similar experiment is underway using amur maple (*A. ginnala*), redbud (*Cercis canadensis*), and green ash (*Fraxinus pennsylvanica*). Results from this work suggest that non-composted wood chips are a potential source of *Vd* inoculum in urban landscapes.

Spiral gradient dilution - A rapid, single-plate method for determining EC₅₀ values of fungicides. H. FORSTER, L. Kanetis, and J. E. Adaskaveg. Dept. Plant Pathology, Univ. of California, Riverside, CA 92521. Phytopathology 92:S26. Publication no. P-2002-0184-AMA.

In vitro toxicity of fungicides against target organisms is commonly expressed as EC₅₀ values that are based on measuring fungal growth or spore germination on culture media amended with serial dilutions of the fungicide. Because this procedure is time-consuming, the spiral gradient endpoint test (SGE) for bacteria was adapted for the use with fungi as a rapid, single-plate

assay. A fungicide solution was plated onto agar medium using a spiral plater, forming a 2.5-log dilution in a radial concentration gradient. Fungal mycelium or spores of up to 15 isolates were then plated along the gradient of a single plate. After incubation, the radial distance where 50% reduction of growth or spore germination occurred was converted into a local fungicide concentration using SGE software (Spiral Biotech, Inc.). EC₅₀ values of fludioxonil obtained for *Botrytis cinerea* and *Monilinia fructicola* and of myclobutanil for *Chondrostereum purpureum* were very similar to those obtained from a series of single-dilution plates. Spiral gradient dilution has potential for mass-screening of fungal isolates for establishing baseline sensitivities.

Phylogeny of *Gaeumannomyces graminis* based on sequence insertions in the small subunit rDNA. H. M. FOULY and H. T. Wilkinson. Department of Natural Resources and Environmental Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S26. Publication no. P-2002-0185-AMA.

The middle region of the small subunit ribosomal DNA of *Gaeumannomyces graminis* (*Gg*) (SSUrDNA) was amplified using universal primers NS5 and NS6. DNA sequences were determined for 28 isolates. The sequence insertions in this region were used to distinguish *Gg* isolates and resolve their phylogenetic relationship. Based on the presence of sequence insertions, *G. g. var. graminis* (*Ggg*) isolates were divided into 4 groups: group one did not have any sequence insertion; group two had a single insertion of 323-389 nucleotides; group three had a single insertion of 381 nucleotides; and group four had two insertions of 381 and 398 nucleotides, respectively. Isolates of *G. g. var. tritici* (*Ggt*) and *G. g. var. avenae* (*Gga*) had a single insertion homologous to that of *Ggg* group-3. These sequence insertions were aligned and cladistically analyzed. Data revealed a major clade. Within this clade, *Ggt* and *Gga* isolates formed two distinct subclades, respectively. *Ggg* isolates did not form distinct clade but were distributed between the *Ggt* and *Gga* subclades. Parsimony analysis of *Ggg* group 2 insertions revealed a distinct clade consisting of 2 monophyletic groups.

ITS ribosomal DNA Phylogeny of *Gaeumannomyces graminis*. H. M. Fouly. Department of Natural Resources and Environmental Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S26. Publication no. P-2002-0186-AMA.

Gaeumannomyces graminis causes take-all disease on cultivated cereal grasses. In the present study, the 5.8S and the internal transcribed spacers (ITS) regions of the ribosomal DNA from 21 isolates belonging to *G. graminis* varieties [*tritici* (5), *avenae* (4), and *graminis* (9)], *G. incrustans* (1), *G. leptosporous* (1), and *G. cylindrosporous* (1) were sequenced and compared to each other. Phylogenetic parsimony analysis among the isolates was carried out. Together, including gaps, ITS and 5.8S regions totaled 660 aligned sites, 220 of which were informative. Only 18 of the informative sites were located in the 5.8S rDNA. All of the isolates tested formed a major clade with high bootstrap support value (100%). *G. leptosporous* and *G. cylindrosporous* formed a distinct subclade. *G. graminis var. avenae* grouped together as sister isolates. *G. g. var. tritici* isolates formed two monophyletic groups. Four *G. g. var. graminis* isolates formed two distinct groups, while two additional isolates were associated with *Ggt* and *Gga* groups. The rest of the isolates were randomly positioned. These data support our previous results using Random amplified polymorphic DNA (RAPD) and Restriction fragment length polymorphism (RFLP) of rDNA.

Host range and distribution of a *Longidorus* sp. associated with stunted loblolly pine seedlings. S. W. FRAEDRICH (1), M. M. Cram (1), and Z. A. Handoo (2). (1) USDA Forest Service, Athens, GA 30602; (2) USDA ARS, Beltsville, MD 20705. Phytopathology 92:S26. Publication no. P-2002-0187-AMA.

An undescribed *Longidorus* sp. has been associated with severely stunted loblolly pine seedlings at a Georgia nursery. Various crop and weed species were inoculated with 100 or 200 *Longidorus* per container to evaluate host range. Nematode soil populations increased in containers with slash, loblolly and longleaf pine seedlings. *Longidorus* reduced the dry root weights of slash ($P = 0.008$) and loblolly ($P = 0.047$) but not longleaf ($P = 0.095$) pine compared to controls. Populations of *Longidorus* decreased on wheat, rye and sorghum as well as yellow nutsedge. A survey conducted in and around the nursery detected a morphologically identical *Longidorus* sp. in loblolly and slash pine seed orchards that border the nursery. Thus far, *Longidorus* has not been found in red cedar windrows, an oak seed orchard or pine stands adjacent to the nursery.

Harnessing the endophytic actinomycetes of cereal crops. C. M. M. FRANCO (1), J. T. Coombs (1,2), and P. M. Michelsen (1). (1) Dept. Biotechnology, Flinders University, Adelaide, Australia; (2) Dept. Plant Pathology, Cornell University, Ithaca, NY. *Phytopathology* 92:S27. Publication no. P-2002-0188-AMA.

Endophytic actinomycetes colonising healthy wheat and barley plants were isolated from surface-sterilised root tissue from cropping sites across South Australia. Over 100 distinct actinomycete isolates were obtained over the growing season, and on the basis of 16S rDNA sequencing were identified as belonging to a small group of genera including *Streptomyces*, *Microbispora*, *Micromonospora*, *Nocardioides*, *Streptosporangium* and *Tsukumarella*. The endophytic status was confirmed with a GFP-tagged strain developed to study the colonisation of wheat seedlings. Extensive screening in laboratory and glasshouse trials revealed a number of isolates that displayed significant activity against fungal root diseases caused by *Gaeumannomyces graminis* var. *tritici* and *Rhizoctonia*, and also as growth promoting agents. This is important as endophytic microorganisms used as inoculants for biocontrol and growth promotion will provide an advantage due to their location within plants.

Improved detection of *Phytophthora capsici* in a Florida pepper field. R. D. FRENCH-MONAR, D. J. Mitchell, P. D. Roberts, and J. B. Jones. Plant Pathology Department, University of Florida, Gainesville, FL 32611. *Phytopathology* 92:S27. Publication no. P-2002-0189-AMA.

Recovery of *Phytophthora capsici* from soil was evaluated in pepper plots located in southwest Florida. Treatments included methyl bromide fumigation, solarization, and their corresponding non-treated controls (control M and control S). Microwaved soil, infested with oospores, sporangia, and mycelia in wheat seed, was placed into acrylic membrane envelopes and buried 15 cm deep in each of six repetitions per treatment. Six envelopes per treatment were sampled at 0, 5, and 13 weeks after planting. Soil dilution plating (SDP) on a selective medium and a modified SDP (MSDP) in which clarified V8 juice agar was amended with rifampicin and ampicillin and layered over the 5-day-old dilution plates were used in detection assays. *P. capsici* was not detected in methyl bromide plots with either assay. After 13 weeks, using SDP and MSDP, *P. capsici* was detected in 2 and 6, 3 and 6, and 0 and 6 samples of the control M plots, control S plots, and solarization plots, respectively. For all samples, detection increased from 10 samples with SDP to 17 samples with MSDP in control M, 8 to 17 in control S, and 0 to 9 in solarization plots.

Virulence to pine in the progeny of a hybrid cross in the *Gibberella* mating population complex. C. J. FRIEL, S. C. Kirkpatrick, and T. R. Gordon. University of California, Davis. *Phytopathology* 92:S27. Publication no. P-2002-0190-AMA.

Gibberella circinata (anamorph = *Fusarium circinatum*), the causal agent of pitch canker on Monterey pine (*Pinus radiata*), is interfertile with an isolate of *Gibberella fujikuroi* mating population E (Netza 9), that was originally recovered from teosinte, the progenitor of maize. When inoculated into *P. radiata*, Netza 9 consistently produced very small lesions. The female fertile isolate was grown on carrot agar (CA); the male strain was grown on PDA. Conidia from the PDA culture were added to the CA cultures, which were incubated in a growth chamber. Single ascospores obtained from these hybrid crosses were evaluated for pathogenicity on Monterey pine, and found to be avirulent. A backcross of the F₁ generation to the virulent parent yielded progeny with a wide range of virulence. Further backcrosses to Netza 9 with selection for virulence are planned, in order to obtain isogenic lines differing in virulence to pine. This will facilitate a search for the genetic determinants of virulence in the pitch canker fungus.

Effect of transgenic anthuriums producing the Shiva-1 lytic peptide on beneficial bacteria. T. M. Fujii, A. ALVAREZ, R. Fukui, K. Obsuwan, and A. R. Kuehnle. University of Hawaii, Honolulu, HI 96822. *Phytopathology* 92:S27. Publication no. P-2002-0191-AMA.

Responses of non-target organisms to transgenic plants must be understood if biological control strategies are to be used in conjunction with genetic resistance. We have genetically engineered anthurium to express a secreted cecropin analogue Shiva-1 to control blight caused by *Xanthomonas campestris* pv. *dieffenbachiae* (Xcd). Inhibitory concentrations of Shiva-1 against 4 foliar biocontrol bacteria (BCAs) and Xcd were determined by dilution plating after 18 hours exposure to the lytic peptide. Populations were measured in guttation fluid 6 and 11 days post-inoculation. Three of four BCAs were statistically less sensitive to Shiva-1 ($P < 0.05$); Xcd was completely inhibited at 0.5 micromolar. Populations of two of four BCAs increased at 1 micromolar Shiva-1. Transgenic anthuriums, Paradise Pink and Mauna Kea,

did not inhibit BCAs more than non-transgenic anthuriums. Biological control with the BCAs, *Sphingomonas chlorophenolica*, *Microbacterium testaceum*, *Brevundimonas vesicularis*, and *Herpaspirillum rubrisubalbicans* is thus compatible with engineered genetic resistance to bacterial blight on anthurium.

Rhizobium species form biofilms on both biotic and abiotic surfaces. N. A. Fujishige (1), K. S. Jankaew (1), C. J. Butcher (1), and A. M. HIRSCH (1,2). (1) Department of Molecular, Cell and Developmental Biology; (2) Molecular Biology Institute, University of California, Los Angeles, CA 90095-1606. *Phytopathology* 92:S27. Publication no. P-2002-0192-AMA.

We found that transfer of *Sinorhizobium meliloti* nod genes to *Bradyrhizobium japonicum* and to *Rhizobium leguminosarum* bv. *viciae* allowed these strains, which normally nodulate soybean and pea, respectively, to form nodules on roots of transgenic alfalfa plants carrying either the soybean (SBL) or the pea (PSL) lectin genes. Moreover, significantly more bacteria attached to the transgenic roots than to the non-transgenic roots. From preliminary TEM studies, we observed that the transconjugant bacterial strains were decorated with what appeared to be pili or fimbriae. Because fimbriae are important for biofilm formation in numerous other bacteria, these observations led us to examine whether or not rhizobial species were capable of forming biofilms on abiotic surfaces as well as biotic surfaces. We focused our studies on *S. meliloti* because the genome has been completely sequenced. We also have data showing that *R. leguminosarum* bv. *viciae* forms biofilms in vitro. Conditions for obtaining *S. meliloti* strain Rm1021 biofilms on abiotic surfaces will be described as well as mutants affected in biofilm formation.

Cultivar-specific increase in 2,4-diacetylphloroglucinol-producing fluorescent pseudomonads associated with wheat. D. L. FUNNELL and M. Mazzola. USDA-ARS, Wenatchee, WA. *Phytopathology* 92:S27. Publication no. P-2002-0193-AMA.

Pseudomonas spp. producing 2,4-diacetylphloroglucinol (Phl) have been implicated in suppression of root diseases. It has been shown that wheat cycling alters populations of fluorescent pseudomonads, in a cultivar-specific manner, with an associated reduction of apple replant disease. Fluorescent *Pseudomonas* isolates from wheat roots or from soils planted with 1 of 5 different cultivars in 3 orchard soils were screened by PCR using primers to a gene (*phlD*) in the locus that confers Phl. From the rhizosphere of cultivars Lewjain and Penawawa, Phl-producers represented as high as 28.5% and 10% of the population, respectively, while such isolates were not detected in unplanted soil. Cultivars Eltan and Hill81 supported undetectable or low numbers (5 and 1.6%, respectively) of Phl-producers in different assays. Wheat cycling in orchard soil previously planted with wheat yielded the highest number of Phl-producers (5.5%), while soils planted only in apple resulted in lower percentages (1 to 2%). Colonization studies are being conducted to determine whether the association is maintained when Phl-producers are applied to seed or soil.

A profile of putative parasitism genes expressed in the esophageal glands of *Heterodera glycines*. B. GAO (1), T. Maier (2), E. L. Davis (3), T. J. Baum (2), and R. S. Hussey (1). Depts. of Plant Pathology, (1) University of Georgia, Athens, GA 30602; (2) Iowa State University, Ames, IA 50011; (3) N. C. State University, Raleigh, NC 27695. *Phytopathology* 92:S27. Publication no. P-2002-0194-AMA.

The most evolutionary advanced adaptations for plant parasitism by nematodes are the products of parasitism genes expressed in the esophageal glands and secreted through the stylet into host tissue. Microaspiration of cytoplasm from glands of 10 parasitic stages of *H. glycines* provided mRNA for RT-PCR to construct a long-distance cDNA library. Of 2,345 cDNAs sequenced, predicted protein sequences of 231 cDNAs were preceded by a secretion signal peptide and, thus, could have roles in plant parasitism. In mRNA in situ hybridizations, 40 cDNAs coding signal peptides specifically hybridized to mRNA within subventral (11) or dorsal (29) glands. PSORT II predicted 30 deduced proteins to be extracellular and 10 as nuclear localized. In BLASTp analyses, 25 predicted proteins were novel. Those with similarities to known proteins included beta-1,4-endoglucanases, a pectate lyase, a chitinase, and RanBPMs. Only two gland-expressed genes had homologues in *Caenorhabditis elegans*.

Differential gene expression in dimorphism in *Ustilago maydis*. M. D. GARCIA-PEDRAJAS, D. L. Andrews, and S. E. Gold. University of Georgia. *Phytopathology* 92:S27. Publication no. P-2002-0195-AMA.

Ustilago maydis displays dimorphic growth alternating between a budding haploid form and a filamentous dikaryon. This morphological switch is

critical for pathogenicity since only the filamentous dikaryon can infect corn plants. Previously, we have identified a role for the cAMP signal transduction pathway in dimorphism and pathogenicity. We are now using suppression subtractive hybridization PCR (SSH) to identify novel genes involved in dimorphism. Using this technique we have identified a number of genes upregulated during filamentous growth and have confirmed differential expression by northern blot analysis. Seeking genes specifically expressed in the budding form we have arrayed a library of 6400 clones and shown by reverse northern blots that the vast majority of them are upregulated during budding growth. We are now in the process of sequencing several hundred of these clones. We have isolated genomic clones of some of the highly differentially expressed genes and produced disruption mutants to study their roles in morphogenesis and pathogenesis.

Comparison of AFLP and ISSR genetic markers in the study of populations within *Pythium* species. C. D. GARZON, D. M. Geiser, and G. W. Moorman. Dept. Plant Pathology, The Pennsylvania State University, University Park, PA 16802. Phytopathology 92:S28. Publication no. P-2002-0196-AMA.

The AFLP technique has been widely used for identification purposes and to generate useful data for population genetic studies. Its reported advantages include the generation of high numbers of genetic markers and a high reproducibility of results. The success in the application of any technique to obtain informative data depends on the nature of the populations of the organism being studied. In previous work using AFLP fingerprinting we identified isolates of *P. aphanidermatum*, *P. irregulare* and *P. ultimum* to the species level using two AFLP primer combinations and detected a population structure in *P. irregulare*. *P. aphanidermatum* shows very little genetic variation among isolates from the US and around the world. In spite of the high levels of variation characteristic of AFLPs, little information was obtained that could be used to resolve groups within *P. aphanidermatum*, so alternative markers are being employed to investigate populations. We will compare the resolution power of AFLPs and internal simple sequence repeat (ISSR) markers and discuss the benefits and pitfalls of both approaches.

Analysis of the capsular exopolysaccharide from *Erwinia amylovora* and the Asian pear pathogen *E. pyrifoliae* and genetics of bioynthesis. K. GEIDER (1), W.-S. Kim (1), M. Schollmeyer (1), and M. Nimtz (2). (1) MPI für Zellbiologie, Ladenburg; (2) GBF, Braunschweig, Germany. Phytopathology 92:S28. Publication no. P-2002-0197-AMA.

Erwinia pyrifoliae causes Asian pear blight and produces exopolysaccharide (EPS) comparable to amylovoran of *E. amylovora* in sugar composition and linkages. The structure of the repeating units was determined by degradation of the EPS with a viral EPS-depolymerase, by methylation analysis and by ESI/MS, confirmed 1H-NMR spectra. EPS of *E. pyrifoliae* carried side chains, which were also terminated by acetyl and pyruvyl residues. A second side chain with glucose, found for up to 50% of the repeating units of amylovoran, was completely absent. The nucleotide sequences of five genes of the *cps*-operon of *E. pyrifoliae* encoding proteins for EPS synthesis were characterized and displayed high homology with the corresponding *ams* genes. Similar functions of the gene products are assumed. A *cpsB* mutant of *E. pyrifoliae* did not produce ooze on slices of immature pears nor symptoms on pear seedlings as *ams* mutants of *E. amylovora*. The *E. pyrifoliae* mutant was complemented with a gene cluster of *E. amylovora*, including *amsB*, for EPS-synthesis and virulence on pear slices.

Molecular analysis of the response of wild barley-derived lines to inoculation with *Rhynchosporium secalis* spores. R. K. GENGER (1), K. Oldach (2), and W. Knogge (2). (1) Centre for Plant Biodiversity Research, CSIRO Plant Industry, GPO Box 1600, Canberra ACT 2601 Australia; (2) Dept. of Plant Science, Waite Campus, Adelaide University, Glen Osmond SA 5064 Australia. Phytopathology 92:S28. Publication no. P-2002-0198-AMA.

Barley leaf scald is a serious disease in temperate barley growing regions worldwide. Breeding for resistance is the major control strategy, but no scald resistance genes have yet been cloned, and little is known about early molecular events after infection. The disease is caused by the deuteromycete *Rhynchosporium secalis*, which, in its development on the plant, forms a subcuticular mycelium secreting small necrosis-inducing proteins. One of these proteins, NIP1, determines avirulence on barley plants carrying the *Rrs1* resistance gene. Using the *Rrs1-nip1* system, a subtractive hybridisation approach was used to identify genes differentially expressed in resistant and susceptible plants. These clones were used to analyse gene expression post-inoculation in two lines that carry different scald resistant genes derived from wild barley, using two races of the fungus with differing virulences. Results of this analysis will be presented.

Development of molecular markers linked to scald resistance genes derived from wild barley, *Hordeum vulgare* ssp. *spontaneum*. R. K. GENGER, A. H. D. Brown, and J. J. Burdon. Centre for Plant Biodiversity Research, CSIRO Plant Industry, GPO Box 1600, Canberra ACT 2601 Australia. Phytopathology 92:S28. Publication no. P-2002-0199-AMA.

Leaf scald is a serious disease widespread in the temperate barley growing areas of the world. Generally, single gene resistance to scald is not durable, due to the high levels of pathogenic variability exhibited by the causal organism, *Rhynchosporium secalis*. Wild barley is a rich source of scald resistance; molecular markers for these scald resistance genes will facilitate pyramiding, a strategy likely to increase resistance durability. We previously used wild barley as the non-recurrent parent to generate scald resistant third backcross lines in a susceptible cultivar, and identified markers linked to resistance loci on 1H, 3H, 6H and 7H. Here we describe the development of markers for scald resistance genes in two lines, for which the locations of the resistance genes were previously unknown. Wild barley also contains unwanted traits such as head shattering. One scald resistant line contains a shattering gene closely linked to the resistance locus, which is marked by the isozyme marker, *Idh1*. We have used *Idh1* to select non-shattering recombinant lines.

Vegetative and sporulating structures of *Pasteuria penetrans* from *in vitro* production. J. F. Gerber (1), T. E. Hewlett (2), M. L. SMITHER-KOPPERL (2), and J. H. White (2). (1) CDG Laboratories, Inc., Gainesville, FL; (2) Entomos, LLC, 4445 SW 35th Terr., Suite 310, Gainesville, FL 32608. Phytopathology 92:S28. Publication no. P-2002-0200-AMA.

The endoparasitic bacterium *Pasteuria penetrans* is a highly effective biological control agent for the root knot nematode, *Meloidogyne* spp. Due to the previous inability to grow the bacterium *in vitro*, the vegetative and sporulating structures have been deduced only by observations of infected nematodes. *In vitro* culture has allowed direct observation of *P. penetrans* structures by light and scanning electron microscopy. The early vegetative stage was identified as pleomorphic structures, including cocci and rods, which grow through elongation and division. The rod shaped bacteria differentiate into mycelial balls, which in turn form thalli and mature endospores. The endospores attach to *M. arenaria* J2 nematodes and are infective. The early vegetative stage had not previously been identified with confidence from observation of *P. penetrans* in infected nematodes. The ability to produce endospores of *P. penetrans* from vegetative stage cells will enable wider implementation of this bacterium for biological control of nematodes.

Nematode transmission: An effective method for screening blackberry germplasm for resistance to Tobacco ringspot virus. R. C. GERGERICH, S. J. Troxell, and S. L. Wickizer. Dept. of Plant Pathology, University of Arkansas, Fayetteville 72701. Phytopathology 92:S28. Publication no. P-2002-0201-AMA.

In 2000, Tobacco ringspot virus (TRSV) was isolated in Arkansas from cultivated blackberry plants showing a severe mosaic symptom on leaves of primocanes and floricanes. Nine blackberry cultivars were tested for resistance to TRSV using nematode transmission. Populations of *Xiphinema americanum* were given access to TRSV-infected cucumber for 10 days, after which the nematodes were transferred to the roots of young blackberry plants (~ 150 nematodes/plant) grown in fine river sand. After 6 weeks, the plants were planted in a commercial potting mix, and one year post-inoculation (after dormancy), roots of test plants were evaluated for TRSV infection using ELISA. All tested cultivars including 'Arapaho', 'Apache', 'Chester', 'Chickasaw', 'Choctaw', 'Kiowa', 'Navaho', 'Shawnee', and 'Triple Crown' were susceptible to infection by TRSV by both graft and nematode inoculation. Use of nematode transmission for resistance screening allows evaluation for susceptibility to the virus as well as susceptibility to nematode transmission of the virus.

Control of Fusarium scab with puroindoline-containing transgenic wheat. S. A. Gerhardt, C. Balconi, and J. E. SHERWOOD. Montana State University, Bozeman, MT. Phytopathology 92:S28. Publication no. P-2002-0202-AMA.

Head scab, a disease of wheat and barley caused by either *Fusarium graminearum* or *F. culmorum*, results in premature ripening and white heads. The puroindoline proteins PINA and PINB, which are found in the wheat endosperm and contribute to grain softness, also have *in vitro* and *in vivo* anti-fungal properties. The growth of both *Fusarium* spp. was inhibited by PIN using *in vitro* bioassays. Wild-type and transgenic HiLine wheat that over-express the *pinB* gene driven by the constitutive maize ubiquitin promoter or the endosperm-specific glutenin-promoter were inoculated with

F. culmorum in field and glasshouse studies. The plants were analyzed for scab by visual inspection. Between 30-60% of control plants were heavily infected, with 40-70% infected spikelets/head, while only 4-20% of *pinB*-transgenic plants were infected at the same level. The percentage of tombstones produced by the *pinB*-transgenic lines was reduced by 30-90% compared to controls. Experiments are in progress with *F. graminearum* as the disease agent. These data suggest that PIN may provide protection to wheat against Fusarium scab.

Effect of soybean planting date on incidence of Bean pod mottle virus. L. GIESLER (1), A. Ziemis (1), and T. Hunt (2). Dept. Plant Pathology (1) and Dept. Entomology (2), University of Nebraska, Lincoln, NE. Phytopathology 92:S29. Publication no. P-2002-0203-AMA.

Soybean producers commonly adjust planting dates for machinery management and in some cases yield advantage. *Bean pod mottle virus* (BPMV) is a common soybean pathogen that has increased significantly in the North Central Region where the majority of the U.S. soybean crop is grown. On-farm research projects were conducted to explore the effect of planting date on BPMV incidence. Replicated strips of April and May planting dates with eight or twelve 30 in. rows at two locations were arranged in a randomized complete block design. BPMV incidence was measured by collecting 66 trifoliates from each strip and assaying 10 sets of six leaflets for each pooled sample. Incidence for each strip was calculated from the number of Elisa positive samples for each set of 10 subsamples processed. Planting date affected BPMV incidence at both locations ($P = 0.06$ and $P = 0.002$) with more virus in April planted than May planted soybean. May planted soybean had a 42% lower BPMV incidence ($P < 0.01$) than April planted soybean in mid-vegetative growth stages at one location. We conclude that early-planted soybean has more BPMV than later planted soybean.

Interaction of glyphosate tolerance with soybean cyst nematode resistance. L. GIESLER (1), G. Graef (2), J. Wilson (3), J. Schimelfenig (1), and T. O. Powers (1). (1) Dept. Plant Pathology and (2) Dept. Agronomy and Horticulture, University of Nebraska, Lincoln, NE; (3) Burt County Cooperative Extension, Tekamah, NE. Phytopathology 92:S29. Publication no. P-2002-0204-AMA.

A study was conducted to determine if differences in response to SCN, measured in yield and nematode reproduction, exist among soybean cultivars differing in SCN resistance and glyphosate tolerance. Twelve glyphosate tolerant and 12 conventional soybean cultivars were tested, with 6 SCN-resistant (R) and 6 SCN-susceptible (S) cultivars in each group of 12. They were planted in a randomized complete block design in an SCN infested field. There was a significant interaction in yield between glyphosate tolerance and SCN resistance ($P = 0.04$) with a difference of 3 and 11 bu./A between SCN-R and SCN-S in conventional and glyphosate tolerant cultivars, respectively. Average yield among conventional SCN-S cultivars was 6 bu./A higher than glyphosate tolerant SCN-S cultivars. No significant interaction was found when measuring SCN reproduction. There appears to be an interaction of glyphosate tolerance with SCN susceptibility for some soybean cultivars.

Aphid vector competence and transmission of Pennsylvania isolates of plum pox virus from infected peach fruit. F. E. GILDOW (1), V. D. Damsteegt (2), A. L. Stone (2), and D. G. Luster (2). (1) Dept. Plant Pathology, Penn State University, University Park, PA 16802; (2) USDA-ARS Foreign Disease-Weed Science Research Unit, Ft. Detrick, MD 21702-5023. Phytopathology 92:S29. Publication no. P-2002-0205-AMA.

Eight species of aphids collected from a peach orchard in Adams Co., PA were tested for their ability to transmit two Pennsylvania isolates of plum pox potyvirus recovered from infected trees in Adams (PPV-R1) or Franklin Co. (PPV-FF). Clones of each aphid species were reared on herbaceous host plants. Several acquisition and inoculation protocols were tested to enhance non-persistent transmission of PPV. Four species, *Myzus persicae*, *Brachycaudus persicae*, *Aphis spiraeicola*, and *A. fabae* consistently transmitted PPV. *Rhopalosiphum padi* transmitted occasionally, and *Macrosiphum euphorbiae*, *A. pisum* and *A. glycines* did not transmit PPV in replicated tests. *M. persicae* and *B. persicae* consistently transmitted PPV to peach seedlings from mature peach fruits collected from ELISA positive trees in orchards. Results prove that PA isolates of PPV can be transmitted by aphid species occurring in Prunus orchards and verifies the ability of these aphids to transmit PPV from infected fruits.

New method for screening cowpea germ plasm for resistance to cucumber mosaic virus. A. G. Gillaspie, Jr. USDA, ARS, Plant Genetic

Resources Conservation, Griffin, GA 30223. Phytopathology 92:S29. Publication no. P-2002-0206-AMA.

Utilizing cowpea [*Vigna unguiculata* (L.) Walp.] germ plasm line, GC-86L-98, a greenhouse screening method was developed to find potential sources of CMV resistance in the USDA cowpea germ plasm collection. GC-86L-98, the first CMV-resistant cowpea germ plasm line, was released by ARS in August 2001. A uniform source of CMV inoculum (freeze-dried cowpea tissue) was diluted to give an infection rate in GC-86L-98 similar to that observed under field conditions. More concentrated inoculum caused infection in all of the plants. Seedlings of cowpea test lines as well as GC-86L-98 and susceptible cultivar, Coronet, (both lines were included for comparison in each test) were mechanically inoculated and tested individually by DAC-ELISA after 2 weeks. In another test, the plants were sampled with a number 10 core borer, the leaf disks were combined in groups of 3 to 5 disks for ELISA, and the absorbance readings averaged for each test line. These averaged results did not always correlate with the percent infected numbers. The greenhouse percent infection screening method was used since it gave results most similar to those in field tests.

Variation in fungicide sensitivity of *Colletotrichum acutatum* isolates from blueberries. J. M. Gillett, A. M. C. SCHILDER, and R. W. Sysak. Dept. of Plant Pathology, Michigan State University, East Lansing, MI 48824. Phytopathology 92:S29. Publication no. P-2002-0207-AMA.

The sensitivity of *C. acutatum* isolates from blueberry to benomyl, thiophanate-methyl, azoxystrobin, and pyraclostrobin was assessed. In a preliminary test with two isolates, PDA was amended with 0, 0.001, 0.01, 0.1, 1, and 10 ppm azoxystrobin or pyraclostrobin; or with 10, 100, and 1000 ppm benomyl. SHAM at 100 ppm was always added to strobilurin plates to prevent an alternative respiration pathway. Azoxystrobin and pyraclostrobin at 1 ppm reduced conidial germination >95% while benomyl did not at any rate. In a larger experiment, hyphal growth of 50 isolates was measured on PDA amended with either 1 ppm azoxystrobin, 1 ppm pyraclostrobin, 10 ppm benomyl or 10 ppm thiophanate-methyl. Controls consisted of PDA and PDA + SHAM. Pyraclostrobin was consistently most effective at inhibiting hyphal growth. Azoxystrobin, benomyl, and thiophanate-methyl also inhibited hyphal growth but to a lesser extent. Isolates varied in sensitivity to each fungicide but not sufficiently to indicate resistance. In two field trials, applications of pyraclostrobin resulted in the lowest levels of anthracnose.

Performance of a warning system for sooty blotch and flyspeck on apple using on-site wetness measurements and site-specific wetness estimates. M. L. GLEASON (1), M. Babadoost (2), P. S. McManus (3), S. N. Wegulo (1), and S. J. Helland (1). (1) Dept. Plant Pathology, Iowa State University, Ames, IA 50011; (2) Dept Crop Sciences, University of Illinois, Urbana, IL 61801; (3) Dept Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S29. Publication no. P-2002-0208-AMA.

A warning system for sooty blotch and flyspeck (SBFS) on apples, developed in North Carolina (T.B. Sutton) and Kentucky (J.R. Hartman), was evaluated in five field trials in Iowa, Illinois, and Wisconsin from 1998-2001. The system delayed the second-cover fungicide spray until 175 hours of leaf wetness duration (LWD) had elapsed. Inputting LWD data from a sensor placed beneath the apple canopy saved an average of two fungicide sprays/year compared to a protectant spray program. Disease incidence was equivalent for the two spray schedules except for FS at one site. Using site-specific LWD estimates (SkyBit Inc.) saved an average of 0.8 sprays/year, with disease control equivalent to the protectant program. The results suggest site-specific technology overestimated within-canopy LWD, and that the warning system could benefit apple growers in the Upper Midwest.

Characterization of a *Fusarium verticillioides* seedling pathogenicity factor. A. E. GLENN, F. I. Meredith, and R. T. Riley. USDA, ARS, Toxicology & Mycotoxin Res. Unit, Russell Res. Center, Athens, GA 30604. Phytopathology 92:S29. Publication no. P-2002-0209-AMA.

As part of a larger research program focusing on the capacity of *Fusarium verticillioides* to infect and endophytically colonize corn, we have examined an apparent seedling pathogenicity factor produced by the fungus. Genetic analysis of field isolates indicated a single locus segregates for ability to cause disease. Strains carrying the non-pathogenic allele did not cause any disease symptoms, yet still infected and endophytically colonized the corn seedlings. Mutant strains that were greatly attenuated in their ability to infect seedlings nonetheless caused severe symptoms, suggesting the pathogenicity factor may be a translocated phytotoxin. Fumonisin B1 (FB1) production was assessed among the parental and progeny strains and was also found to segregate as a single locus. Linkage between pathogenicity and fumonisin production was supported since only the pathogenic strains produced FB1.

Further experiments are underway to better quantify this linkage, examine the molecular nature of the mutation resulting in fumonisin non-production, and also examine the dynamics of FB1 production in soils.

Functional and species composition of soil microbial communities from organic and conventional field soils in North Carolina. D. L. GLENN and J. B. Ristaino. North Carolina State University. Phytopathology 92:S30. Publication no. P-2002-0210-AMA.

A comparative study of field soils from organic and conventional farms was begun in 2001 to better understand the biological, physical, and chemical properties of these soils. Three soil samples were collected from five organic and five conventional farms that grew pepper or tomato. Population densities of culturable *Trichoderma* sp., fluorescent *Pseudomonas* sp., and thermophiles were similar among production systems but varied among individual farms. Total bacterial biomass was highest in conventional field soils while the ratio of active/total bacterial biomass was highest in organic field soils. Microbial community profiles based on carbon substrate utilization were distinct among production systems. Communities differed in utilization of D-galacturonic acid, D-mannitol, N-acetyl-D-glucosamine, and L-threonine. Two soils from organic farms differed from others by their low bulk density, high soil water content, CEC, and labile carbon values. Soils are being evaluated for their ability to suppress sclerotia germination and disease caused by *Sclerotium rolfsii* on bell pepper.

Characterization of a new virulent race of *Tilletia controversa*. B. J. Goates. USDA-ARS, Aberdeen, ID 83210. Phytopathology 92:S30. Publication no. P-2002-0211-AMA.

Dwarf bunt of wheat, caused by *Tilletia controversa*, is controlled with resistant cultivars where the disease has the potential to occur regularly. In 1999, up to 10% bunted spikes occurred in a few southern Idaho farms on two cultivars that historically were extremely resistant. Isolates of this bunt were used to inoculate 12 highly resistant cultivars, 2 partially resistant cultivars, and 15 differential cultivars in two tests performed in 2000 and 2001 to determine their potential virulence. The new race caused 35 to 90% bunted spikes in the cultivars Boundary, Hansel, Manning, Promontory, Survivor, Utah 100, and Weston which have had very little infection in commercial fields and in nurseries inoculated with a broad composite of pathogenic races. Partially resistant cultivars Eltan and Knor had markedly increased disease. The new race caused little or no increase in disease in the cultivars Blizzard, Bonneville, Garland, Golden Spike, Lewjain, Luke, and Winridge. The virulence of this new race appears primarily due to combined virulence to the resistance genes *Bt3*, *Bt9*, and *Bt10*, but low virulence to *Bt8* could also be contributing. Resistance to the new race apparently comes from *Bt12*, but also from unknown factors.

Eradication of sudden oak death in Oregon. E. M. Goheen (1), E. M. HANSEN (2), A. Kanaskie (3), M. G. McWilliams (3), N. Osterbauer (4), and W. Sutton (2). (1) USDA Forest Service, SW Oregon Insect and Disease Service Center, Central Point, OR 97502; (2) Oregon State University, Dept. Botany and Plant Pathology, Corvallis, OR 97331; (3) Oregon Dept. Forestry; (4) Oregon Dept. Agriculture, Salem, OR 97310. Phytopathology 92:S30. Publication no. P-2002-0212-AMA.

Sudden oak death, caused by *Phytophthora ramorum*, was found in July 2001 near Brookings Oregon, killing tanoak (*Lithocarpus densiflorus*). This is the only US report of the disease outside of the San Francisco Bay area, California, 300 km to the south. Nine disease centers were located in mixed tanoak/Douglas-fir forests via the cooperative aerial survey flown by the USDA Forest Service and Oregon Dept. of Forestry. Disease centers included 5 to 40 diseased trees. *P. ramorum* was isolated from tanoak stem cankers and from foliage and shoots of native *Rhododendron* and *Vaccinium*. All lands within 1 mile of the disease centers are subject to Oregon and APHIS quarantine, barring the transport of host materials. An eradication effort followed by intensive monitoring of treated and perimeter areas is underway. All symptomatic and adjacent host plants in the treated areas have been cut and burned on a total area of about 16 ha.

Influence of time of planting on the isolation frequency of *Pythium* spp., *Theilaviopsis basicola* and *Rhizoctonia solani* on cotton. N. G. GOMAA and C. Rothrock. Dept. Plant Pathology, University of Arkansas, Fayetteville, AR 72701. Phytopathology 92:S30. Publication no. P-2002-0213-AMA.

The change in the isolation frequency of *Pythium* spp., *Theilaviopsis basicola*, and *Rhizoctonia solani* on cotton, from the day after planting until 14 days after planting, was examined for two field soils over a range of temperatures and soil water contents under controlled environmental conditions.

Pythium spp. were isolated from cottonseeds one day after planting and the isolation frequency tended to be similar over the experimental period. *Pythium* isolation frequency was favored by wet soils, > -10 joules/kg compared to < -24 joules/kg. *T. basicola* was not isolated until five days after planting and increased dramatically between the fifth day and the tenth day after planting. *R. solani* was isolated from cotton the first day after planting and increase gradually reaching a maximum at 14 days after planting. *R. solani* was favored by the higher temperature treatment, 20/25°C compared to 16/21°C.

Genetic structure of *Phytophthora infestans* populations from Costa Rica. L. GOMEZ, A. C. Café-Filho, and J. B. Ristaino. North Carolina State University. Phytopathology 92:S30. Publication no. P-2002-0214-AMA.

Phytophthora infestans causes late blight and is the most devastating disease of potato worldwide. In Costa Rica, ca. 3000 ha of potato are grown every year mostly under agro-climatic conditions conducive for the disease. In recent years, disease management has become increasingly difficult and could be associated with changes in the population structure of this pathogen. Forty-one isolates of *P. infestans*, including three isolates from wild *Solanum*, were analyzed from two geographically distinct growing regions of Costa Rica to determine the genetic diversity and structure of pathogen population's using allozyme genotyping with *Glucose 6-phosphate isomerase* (*Gpi*) and mitochondrial DNA haplotyping. All isolates tested thus far were the Ia mtDNA haplotype. Allozyme analysis of 29 isolates with *Gpi* revealed the 100/100 genotype. Isolates from wild *Solanum* species were all 100/100. Research is in progress to more fully characterize additional isolates by allozyme genotyping with peptidase, DNA fingerprinting, mating type analysis and metalaxyl sensitivity.

Characterization of isolates of *Colletotrichum gloeosporioides* and *Glomerella cingulata* from apple. E. GONZÁLEZ (1), T. B. Sutton (1), and J. C. Correll (2). (1) Dept. Plant Pathology, North Carolina State University, Raleigh, NC 27695; (2) Dept. Plant Pathology, University of Arkansas, Fayetteville, AR 72701. Phytopathology 92:S30. Publication no. P-2002-0215-AMA.

Isolates of *Colletotrichum* spp. and *G. cingulata* from fruit showing bitter rot symptoms and from leaves with Glomerella leaf spot symptoms from the US and Brazil, were characterized based on morphology, mtDNA RFLPs and VCGs. Isolates of *C. gloeosporioides*, *C. acutatum* and *G. cingulata* were distinguished based on colony color, conidial shape and the ability of monosporic isolates to produce perithecia in culture. Different mtDNA RFLP haplotypes were found within each species, and Brazilian isolates of each of the species could be distinguished from the US isolates. Vegetative compatibility analysis revealed multiple VCGs within some of the mtDNA RFLP haplotypes for foliar isolates of *G. cingulata* from Brazil, indicating a considerable degree of genetic diversity within some of the mtDNA haplotypes for these isolates. These results indicate that mtDNA RFLPs and VCGs are useful for characterizing genetic diversity among populations of *Colletotrichum* species from apple in the US and Brazil.

The barley pathogen *Septoria passerinii* probably has an unobserved sexual cycle. S. B. GOODWIN (1), C. Waalwijk (2), G. H. J. Kema (2), J. R. Cavaletto (1), and G. Zhang (1). (1) USDA-ARS, Purdue Univ., W. Lafayette, IN 47907; (2) Plant Res. Intl., Wageningen, Netherlands. Phytopathology 92:S30. Publication no. P-2002-0216-AMA.

To test for possible sexual reproduction of the barley speckled leaf blotch pathogen *Septoria passerinii*, a putative mating-type locus was cloned and compared to that of its close relative *Mycosphaerella graminicola*. Sequence analysis of the corresponding region from *S. passerinii* confirmed that it is homologous to the mating-type locus of *M. graminicola* and contained *MAT-1* and *MAT-2* idiomorphs of approximately 3 kb. A multiplex PCR test was developed that allowed rapid identification of the mating types of isolates of *S. passerinii*. Both mating types were present in approximately equal frequencies in fields in MN and ND, often among multiple isolates from the same leaves. Analyses with isozyme and RAPD markers revealed that each isolate had a unique genotype. The common occurrence of both mating types on the same leaf and high levels of genotypic diversity indicate that sexual reproduction probably plays an integral role in the life cycle of *S. passerinii* and may be much more important than believed previously in this and possibly other "asexual" species of *Septoria*.

Development of reliable and sensitive detection methods for the diagnosis of major viruses and viroids infecting vegetables, grape and citrus in Tunisia. F. Gorsane, I. Fekih-Hassen, A. Elleuch, F. Djilani, L. Jandoubi, M. Marrakchi, and H. FAKHFAKH. Faculty of Sciences of Tunis, Tunisia. Phytopathology 92:S30. Publication no. P-2002-0217-AMA.

Vegetables, grapes and citrus crops are among the most important horticultural commodities in Tunisia. However, heavy losses caused by viruses and viroids are observed. These pathogens were identified by host range studies, serological (ELISA, tissue-blotting) and molecular techniques (RT-PCR-RFLP, hybridization, sequencing). Through seasonal surveys, the biological and molecular variability of viral and viroidal isolates were studied. Peppers and tomatoes were infected with S-I and S-II groups of *Cucumber mosaic virus*, mild isolates of *Pepper vein mottle virus* and different isolates of *Potato virus Y* and *Tomato yellow leaf curl virus*. Lettuce was infected with *Lettuce mosaic virus* and potatoes were mainly infected with *Potato leaf roll virus* and *Potato virus X*. Viroids detected with molecular methods were *Grapevine yellow speckle viroid* and *Australian grapevine viroid* from grapes and *Citrus exocortis viroid* from citrus. Our results showed that virus and viroids are important pathogens of these crops and that appropriate disease management strategies are necessary.

Differential expression of genes in *Fusarium graminearum* strains with low and high virulence for wheat. R. S. GOSWAMI (1), F. Trail (2), J. R. Xu (3), and H. C. Kistler (4). (1) Dept. Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108; (2) Dept. Botany and Plant Pathology, Michigan State Univ., E. Lansing, MI 48824; (3) Dept. Botany and Plant Pathology, Purdue Univ., West Lafayette, IN 47907; (4) USDA-CDL, St. Paul, MN 55108. Phytopathology 92:S31. Publication no. P-2002-0218-AMA.

Fusarium graminearum is a species complex containing at least eight biogeographically structured lineages. Initial studies were conducted on diverse strains to ascertain differences in aggressiveness (spread of scab symptoms and the fungus from the point of inoculation) and ability to produce mycotoxin when inoculated onto susceptible wheat spikes. Strains varied significantly in their virulence and mycotoxin production. Two strains with high (NRRL 31084) and low (NRRL 28303) virulence were selected for genomic studies. cDNA libraries were created by subtractive hybridization to compare mRNA populations from wheat heads inoculated with the two strains in order to identify genes specific to each interaction. Preliminary results revealed marked differences in gene expression among strains of this pathogen. Ultimately, we hope to understand gene function related to pathogenicity.

Citrus canker: Plant pathology versus public policy. T. R. GOTTWALD (1) and T. S. Schubert (2). (1) USDA, ARS, Ft. Pierce, FL; (2) Florida Department of Agriculture and Consumer Services, Gainesville, FL. Phytopathology 92:S31. Publication no. P-2002-0219-AMA.

Increasing international travel and trade has resulted in an unprecedented number of plant pathogen introductions, including *Xanthomonas axonopodis* pv *citri*, (Xac), the bacterium that causes citrus canker. The disease affects commercial and dooryard citrus, and has far-reaching political and socioeconomic impact. Xac is a leaf and fruit-spotting pathogen that can cause crop loss, but also triggers quarantine of infested areas, disrupting national and international trade. The only known eradication method is removal of diseased and nearby asymptomatic trees suspected to have sub-clinical infections. This has elicited conflict between regulatory agencies, commercial citrus growers, and homeowners resulting in legal proceedings to resolve the disputes. Eradication policy decisions are at times influenced by public perception and legal challenges that are not always in agreement with pathogen biology. Researchers working on Xac find themselves in situations in which scientific information does not take precedence. The effects of public policy decisions on Xac epidemics and implications on research will be discussed.

Control of citrus bacterial diseases by induced systemic resistance. J. H. GRAHAM (1), R. P. Leite, Jr. (2), and D. L. Drouillard (1). (1) CREC, Univ. Florida, Lake Alfred 33850; (2) IAPAR, Londrina, Parana, Brazil. Phytopathology 92:S31. Publication no. P-2002-0220-AMA.

Induced Systemic Resistance (ISR) compounds, Harpin (Messenger) and Acibenzolar-s-methyl (Actigard 50WG), were evaluated in a greenhouse assay using *Xanthomonas axonopodis* pv. *citrumelo*, the cause of citrus bacterial spot (CBS). Lesion numbers were reduced 40-60% when the bacterium was infiltrated into Swingle citrumelo leaves 7-30 days after foliar spray with ISR compounds. Based on activity against CBS, ISR sprays with and without copper hydroxide (CuOH) and copper oxychloride (CuOCl) were evaluated against *X. axonopodis* pv. *citri*, the cause of citrus canker, in field trials in Parana, Brazil. CuOCl sprays (every 30-45 days) were more effective than CuOH for reducing incidence of foliar and fruit lesions on 4 yr-old trees of a midseason sweet orange variety under low and a late season orange under moderate disease pressure. Under low pressure, ISR compounds, either with or without Cu, reduced fruit lesions from detectable to non-detectable. Under moderate pressure, ISR compounds did not provide additional disease control compared to CuOCl alone.

Characterization of defense-related genes in soybean. M. Y. GRAHAM and T. L. Graham. Dept. Plant Pathology, Ohio State University, Columbus, OH 43210. Phytopathology 92:S31. Publication no. P-2002-0221-AMA.

Public availability of the soybean EST database prompted us to begin a systematic characterization of soybean defense-related genes. We first focused on the key enzymes of isoflavonoid metabolism and the pathogenesis-related (PR) protein genes. Next we organized genes potentially involved in defense signaling processes, including transcription factors and redox enzymes. To catalog these genes, we searched the soybean EST database using existing annotations or homologies to prototype defense-related genes from other plants. We assembled the ESTs into contigs representing possible gene family members. Microbial interaction and related EST libraries facilitated selection of those most likely to be defense-related. Primer sets were designed from the selected contigs and PCR products were used as probes in Northern blots from various elicitor/chemical treated or infected soybean tissue samples. As the number of probes increases, we are currently attempting to implement more efficient methods to carry out the expression analyses. From the kinetics and pattern of gene induction we hope to define the signal-response and regulatory networks for soybean defense.

Selection for resistance to *Sclerotinia sclerotiorum* (Ss) within soybean germplasm. C. R. GRAU, A. J. Peltier, and N. C. Kurtzweil. Dept. of Plant Pathology, University of Wisconsin-Madison 53706. Phytopathology 92:S31. Publication no. P-2002-0222-AMA.

Soybean lines are reported to express degrees of partial resistance to Ss. A petiole inoculation technique (PIT) was used to evaluate soybean germplasm for higher levels of resistance to Ss. Cultivars, breeding lines and plant introductions (461) were characterized for reaction to Ss by rate of plant death and percent plant survival. Surviving plants from 52 lines were advanced to seed production. Progeny of selected plants were evaluated for levels of observed resistance and compared to the source line. The cultivars Maple Arrow and MN1401 and breeding lines M90-184111 and M91-196123 expressed high resistance. Plant introductions that expressed high resistance were 153.235, 184.042, 427.143, 507.792, 561.285B, 561.345, 567.157A and 567.157B. Progeny of a single plant selection from MN1401 had 47% survival after challenge with Ss compared to 12% survival for the source line. The PIT provided sources of resistance to Ss and evidence that some soybean lines are heterogeneous for resistance to Ss. Heterogeneous lines are a source of unique plants for breeding purposes and genetic studies of resistance to Ss.

***Monarda didyma* and control of damping off of tomato.** S. E. Greene (1), K. D. GWINN (1,2), D. J. Trently (1), S. L. Hamilton (2), and B. H. Ownley (1,2). (1) Dept. Entomol. & Plant Pathol.; (2) Bioactive Natural Products Center of Excellence, University of Tennessee, Knoxville, TN 37996-4560. Phytopathology 92:S31. Publication no. P-2002-0223-AMA.

Herbage of *Monarda didyma*, a plant with high concentrations of antifungal compounds, was added to greenhouse growth medium to determine if seedling losses caused by *Rhizoctonia solani* could be reduced. The experiments were designed as factorials with 2 rates of *Monarda* herbage, 0 or 10% (v/v) and 2 rates of *R. solani* inoculum, 0 or 2% (v/v) with 20 replicates in a randomized complete block design. Treatments were added to greenhouse germination mix. Three seeds were planted per cell. Seedling emergence and height were measured at one week. Amending germination mix with herbage from 'Marshall's Delight' increased seedling height and germination above that of controls regardless of *R. solani* infestation. For 'Elsie's Lavender', shoot height and germination were reduced in treatments containing only herbage but not in those containing herbage + *R. solani*; the disease index of herbage or herbage + *R. solani* was less than pathogen alone but greater than uninfested, no herbage control. Amendment with 'Sioux' herbage did not protect against *R. solani*.

Mycofumigation for control of soilborne diseases. E. GRIMME, A. M. Stinson, N. K. Zidack, B. J. Jacobsen, and G. A. Strobel. Department of Plant Science and Plant Pathology, Montana State University, Bozeman, MT 59717. Phytopathology 92:S31. Publication no. P-2002-0224-AMA.

The fungi *Muscodor albus* and *M. roseus* produce volatile gases toxic to a wide range of fungi including the genera *Aphanomyces*, *Pythium*, *Rhizoctonia* and *Verticillium*. We are currently testing the efficacy of *Muscodor* sp. as 'mycofumigants' for methyl bromide replacement. These fungi show no phytotoxicity/pathogenicity to a wide range of plants, including barley, sugarbeet, eggplant and peppermint. In greenhouse pot assays *Muscodor* sp. were tested for control of *Rhizoctonia solani*, *Pythium ultimum* and *Aphanomyces cochlioides*, all pathogenic to sugarbeet. We also tested a chemical cocktail of the volatile compounds produced by *M. albus*. Two

weeks after planting, seedling establishment of sugarbeet in the uninoculated control soil for experiments with all three pathogens was from 83 to 97%. Pathogen alone treated soil had 32% emergence in the *Aphanomyces* test, 38% for the *Rhizoctonia*, and 2% for the *Pythium*. Pots treated with formulations of the mycofumigants or a cocktail of the gas components of *M. albus* resulted in equivalent emergence of sugarbeet seedlings compared to the uninoculated control for all pathogens.

A shift in the strains of sorghum mosaic virus causing mosaic in Louisiana sugarcane. M. P. GRISHAM and Y.-B. Pan. USDA, ARS, SRRC, Sugarcane Research Unit. Phytopathology 92:S32. Publication no. P-2002-0225-AMA.

Mosaic of sugarcane is caused by strains of sugarcane mosaic virus (SCMV) or sorghum mosaic virus (SrMV). During the first half of the Twentieth Century, mosaic in Louisiana sugarcane was caused by strains of SCMV. SrMV strains H, I, and M were identified in 1956, 1966, and 1973, respectively. In field surveys of plants with mosaic symptoms conducted between 1978 and 1995, more than 90% were infected with SrMV strain H, the remainder with SrMV strains I and M. No plant was found infected with SCMV. Surveys were discontinued because of the large amount of labor required to identify strains using host differentials and the results had changed little in 10 years. A survey conducted in 2001 using reverse transcription-polymerase chain reaction-based restriction fragment length polymorphism (RFLP) analysis to identify SCMV and SrMV strains indicated a shift in the population of strains. SrMV strain I and strain H were associated with approximately 65% and 21% of the sugarcane plants with mosaic symptoms, respectively. The remainder of the plants (14%) with mosaic symptoms appeared to be infected by a new strain with a distinctive RFLP banding pattern. Nucleotide sequencing is being conducted to identify the virus strain.

Genetic instability in field isolates of *Phytophthora infestans*. C. L. GROVES and E. R. Champaco. USDA-ARS, New England Plant, Soil, and Water Laboratory, University of Maine, Orono, ME 04469. Phytopathology 92:S32. Publication no. P-2002-0226-AMA.

Several *glucose-6-phosphate isomerase (Gpi)* genotypes of *P. infestans* have been identified from field collections in Maine in recent years. These *Gpi* genotypes have RG57 DNA fingerprints typical of the US-8 clonal lineage but possess a variety of alleles at the *Gpi* locus. Single zoospore progeny obtained from five field isolates of *P. infestans* recently collected in Maine were examined to assess the role of genetic instability in generating new genotypes. Genetic instability, characterized by the loss or recombination of alleles at the *Gpi* locus, or a change in mating type or self-fertility, was detected among single zoospore progeny from all five field isolates. *Gpi* patterns typical of the genotypes recently collected from the field in Maine were observed among the aberrant single zoospore progeny obtained from all five field isolates. Segregation for mating type and for self-fertility in culture was observed among single zoospore progeny from at least four of the five field isolates used. Our data suggest that genetic instability may be a plausible mechanism for generating genetic diversity in field populations of *P. infestans*.

Evaluating lettuce (*Lactuca spp.*) germplasm for resistance to *Sclerotinia minor*. R. C. GRUBE and E. J. Ryder. U. S. Department of Agriculture, Agricultural Research Service, Salinas, CA. Phytopathology 92:S32. Publication no. P-2002-0227-AMA.

Several lettuce cultivars and accessions were evaluated for resistance to *Sclerotinia minor* using natural and artificial inoculation techniques in field and greenhouse trials. In replicated field tests, differences were identified among genotypes and results were consistent in different locations and years. Supplementing the existing soilborne inoculum with *S. minor*-infested rye seeds each field season insured high disease incidence without altering the relative performance of control genotypes. Responses to artificial inoculation in some greenhouse tests were strongly correlated with field tolerance. Variability between greenhouse tests, which presumably results from the influence of environmental factors on infection and disease development, has slowed breeding progress and genetic studies of resistance. We have examined the effects of varying quantity and type of inoculum and plant material used in order to better understand, and ultimately, to better control, the factors influencing *S. minor*-lettuce interactions.

Managing potato late blight at the center of origin: integrating durable resistance with a decision support system. N. J. GRÜNWARD (1) and W. E. Fry (2). (1) USDA ARS, 24106 N. Bunn. Rd., Prosser, WA 99350; (2) Dept. Plant Pathology, Cornell University, Ithaca, NY 14853. Phytopathology 92:S32. Publication no. P-2002-0228-AMA.

Management of potato late blight in the highland tropics is costly and difficult. Integrating the use of resistant cultivars with a fungicide forecasting system could lower the number of fungicide applications. The Mexican national potato program has produced several cultivars with high levels of field resistance. We evaluated durability of resistance to potato late blight of a selection of 12 cultivars (1960-1999) and found that field-resistance in the Mexican germplasm is durable. Previously, we evaluated the fungicide forecasting system SimCast for use with these Mexican cultivars and found that it predicted too many fungicide applications for cultivars of moderate to high levels of resistance. We adapted SimCast and field validations conducted in 1999 and 2000 show that SimCast resulted in good disease control on cultivars ranging from susceptible to highly resistant. The number of fungicide applications forecast for cultivars with moderate to high levels of resistance was reduced. Precipitation was the environmental variable responsible for most of the forecasts. A user-friendly decision support system consisting of just a rain-gauge and the exclusive use of SimCast's fungicide units could be a valuable and affordable tool in managing potato late blight in the highland tropics.

Molecular characterization of genetically distinct strains of *Bean pod mottle virus*. H. GU, C. Zhang, and S. A. Ghabrial. Dept. Plant Pathology, University of Kentucky, Lexington, KY 40546. Phytopathology 92:S32. Publication no. P-2002-0229-AMA.

BPMV (genus *Comovirus*), an economically important pathogen of soybean, has a positive sense single stranded bipartite RNA genome, designated RNA1 and RNA2. We have previously reported the occurrence in nature of two distinct subgroups of BPMV strains (subgroups I and II), as well as reassortants between the two subgroups, that can be clearly distinguished based on nucleic acid hybridization and nucleotide sequence analyses. Now we report on the production of infectious full-length cDNA clones of genomic RNAs 1 and 2 from representative strains of the two subgroups and reassortants. Interestingly, the reassortant strains induced severe to very severe symptoms on their soybean host plants, whereas strains in either subgroup I or II induced only mild or moderate symptoms. This phenomenon was also observed when pseudorecombinants were constructed in the laboratory from heterologous infectious transcripts. The availability of the infectious cDNA clones from genetically distinct strains makes it possible to generate the appropriate chimeric constructs that allow mapping the determinants of symptom severity.

Relationship between high resolution IKONOS satellite image intensities and soybean stress caused by soybean cyst nematode. J. GUAN, A. J. A. Moreira, G. L. Tylka, and F. W. Nutter, Jr. Iowa State University. Phytopathology 92:S32. Publication no. P-2002-0230-AMA.

Satellite images may provide a fast, nondestructive, objective, and affordable method to quantify plant stresses for crops grown over large areas. The main goal of this research was to investigate whether satellite images can be used to quantify plant stress caused by SCN. IKONOS satellite images (4-m resolution) were obtained for Iowa State University Woodruff Research Farm for three dates during the 2000 growing season and ERDAS IMAGINE and ArcView (GIS) were used to analyze these images. Linear regression was used to relate soybean yield, protein and oil concentrations, and initial and final SCN population densities to satellite image intensities. Satellite image intensities explained up to 89% of the variation in soybean yield, 54% of the variation in soy protein, 55% of the variation in soy oil, and 82% of the variations in initial and final SCN population densities, respectively. This indicates that satellite images can be used to detect soybean stress caused by soybean cyst nematodes.

Susceptibility of grapevine pruning wounds to infection by *Phaeoemoniella chlamydospora* and *Phaeoacremonium spp.* W. D. GUBLER, A. Eskalen, S. N. Rooney, A. J. Feliciano, and A. Khan. Dept of Plant Pathology, UC Davis, CA. Phytopathology 92:S32. Publication no. P-2002-0231-AMA.

Spores of *Pa. chlamydospora* (Pc), *Phaeoacremonium aleophilum* (Pa) and *Pm inflatipes* (Pi) were shown to be aeri ally disseminated from pycnidia and unknown sources, respectively in CA vineyards. These pathogens are responsible for causing black measles (esca) and Petri disease. Petri disease is a disease of 1-5 yr-old grapevines and black measles, which is a disease of older grapevines has in recent years become more prevalent on young grapevines in CA. Spore release occurred with rainfall events during the late winter and spring when pruning was done. *Pa. chlamydospora*, *Pm inflatipes* and *Pm. aleophilum* were capable of penetrating 1-day-old to 120-day-old pruning wounds. Susceptibility was highest in fresh wounds and decreased with age of the pruning wound. In Thompson Seedless and Cabernet Sauvignon, vascular streaking moved down to the bud positions below the pruning wound thus effecting new shoot positions. Symptoms occurred as

dark vascular streaking and stunted shoot growth having foliar symptoms similar to those of black measles.

Differences in substrate specificity and antimicrobial activity of potato aspartic proteases. M. G. GUEVARA (1), P. Verissimo (2), E. Pires (2), C. Faro (2), A. Mayoral (1), and G. R. Daleo (1). (1) University Nacional De Mar Del Plata, Mar Del Plata, Argentina; (2) Coimbra, Portugal. Phytopathology 92:S33. Publication no. P-2002-0232-AMA.

A few studies have shown that proteases are important in plant defense. We have reported the induction of potato APs in infected tissues with *P. infestans*. Here we analyzed the substrate specificity of potato APs using oxydized insuline α -chain. Potato tuber AP had two majoritary and six minority cleavage positions, whereas potato leaves AP had one majoritary and four minority cleavage positions. We shown that both APs have one majoritary and three minority common cleavage positions. These cleavage positions are common with other plant APs. Other majoritary and two minority cleavage positions of potato tuber AP were not common with potato leaves AP or with other plant APs described. Potato leaves AP was able to inhibit 50% the germination of *P. infestans* at 3.3 μ g/ml whereas potato tuber AP is able to inhibit 100% the germination at 0.33 μ g/ml. Antimicrobial activities were inhibited by pepstatin A. We suggest that the differences in the antimicrobial activity of these proteases is associated with the diferent substrate specificity found for these enzymes.

Evaluation of drought tolerance and relationship with aflatoxin contamination. B. Z. GUO (1), Y. Cao (2), A. E. Coy (3), R. D. Lee (3), C. C. Holbrook (4), and R. E. Lynch (1). (1) USDA-ARS, Crop Protection and Management Research Unit, Tifton, GA; (2) Department of Entomology; (3) Department of Crop and Soil Sciences, University of Georgia, Tifton, GA; (4) USDA-ARS, Crop Genetics and Breeding Research Unit, Tifton, GA. Phytopathology 92:S33. Publication no. P-2002-0233-AMA.

Corn and peanuts become contaminated with aflatoxins when subjected to prolonged periods of heat and drought stress. The effect of drought tolerance on aflatoxin contamination is not clear. The objectives of this research were to evaluate preharvest aflatoxin contamination in commercial corn hybrids known to have drought tolerance and to determine the correlation of drought tolerance with aflatoxin contamination. Gene expression under drought stress had been studied using DD-RT-PCR. Fifteen genotypes with different levels of drought tolerance were grown in two locations in Georgia in 2000 and 2001. Each location had two treatments, irrigation and not irrigation. The field evaluation on drought tolerance and aflatoxin production have demonstrated that drought tolerant commercial lines, in general, had lower aflatoxin contamination in drought condition. One poly(A)-anchored oligonucleotides and 10 arbitrary primers are used to differentiate gene expression. Polymorphic mRNA transcripts have been identified. Some cDNA fragments, up- or down-regulated by induced drought stress, have been cloned and sequenced. Further Studies are needed to characterize these cloned fragments in relationship with drought tolerance and aflatoxin formation.

Occurrence of ring nematode on creeping bentgrass putting greens in Alabama. A. K. HAGAN, K. McLean, and M. E. Rivas-Davila. Dept. Entomology and Plant Pathology, Auburn University, AL 36849. Phytopathology 92:S33. Publication no. P-2002-0234-AMA.

High populations of ring nematode (*Mesocriconema* sp.) have been associated with the summer decline of creeping bentgrass (*Agrostis paulstris*) putting greens on several golf courses in Alabama. During extended periods of hot, dry summer weather, damage appears as a gradual thinning or sudden wilting and death of large irregular patches of turf. In several instances, ring nematode numbers on symptomatic greens have exceeded 2000 to 4000 per 100 cc of soil. A combination of improved ventilation, aggressive core aeration, and timely nematicide treatments has suppressed symptom onset and ring nematode populations. Intensive sampling has shown that localized 'hot spots' of high nematode populations were randomly distributed across one bentgrass green on two golf courses. In 2001, nematode populations gradually increased through the summer months, peaked between September and November, and then declined in the winter. In 2002, ring nematode populations will be monitored on selected bentgrass greens at three golf courses. Digital imaging will be used to correlate ring nematode populations with the vigor of bentgrass.

Eradication of *Pseudomonas syringae* pv. *syringae* in sweet cherry bud sticks. T. W. HALL, M. C. Heidenreich, R. Ciccirelli, R. L. Andersen, and W. W. Turechek. Cornell University, Geneva, NY 14456. Phytopathology 92:S33. Publication no. P-2002-0235-AMA.

Bacterial canker of sweet cherry can be a serious problem in young orchards. Initial introduction into an orchard could be the result of systemically infected propagation material, and/or post planting acquisition from an indigenous field source. In western New York (NY), levels of detectable systemic infection of dormant Royalton sweet cherry buds ranged between 5–40%. Various hot water treatments were applied to dormant bud sticks to determine an appropriate treatment for eradicating the pathogen while minimizing damage to the bud sticks. Immersion of bud sticks in 52°C water followed by immediate cooling by immersion in 21°C water was found to eliminate detectable systemic infections of the pathogen. This hot water treatment was applied to dormant Royalton, Hedelfingen, and Bing bud sticks collected from NY and the treated sticks were grafted onto established Mazzard rootstocks. Bud blast and canker development were monitored relative to untreated Royalton and Hedelfingen collected in NY and untreated Hedelfingen and Bing obtained from Prosser, WA.

***Aspergillus* and *Arabidopsis*, elucidating the role of the host in mycotoxin production.** T. M. HAMMOND, J. H. Ham, and N. P. Keller. Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S33. Publication no. P-2002-0236-AMA.

Aspergillus flavus secretes aflatoxin, a highly toxic and carcinogenic secondary metabolite. While there is much known about the biosynthetic pathway leading to aflatoxin, there is little known about its role in the pathogen-seed interaction. There is some evidence to suggest that the host genotype may positively or negatively influence the production of this toxin during the infection process. Unfortunately, a tractable model to carefully study the affect of plant genotype on toxin production is not readily available. To solve this dilemma we are developing a system using the model organism *Arabidopsis thaliana*. Our results show that *Aspergillus* species infect *Arabidopsis* seed and produce mycotoxin in a manner that is similar to infection of maize and peanut, the two most agriculturally important hosts of *Aspergillus*. Future tests using transgenic or mutant *Arabidopsis* may help us elucidate characteristics of the host genome responsible for increased or decreased aflatoxin production during the pathogen-seed interaction.

Log inoculations to assess tree susceptibility to sudden oak death. E. M. HANSEN and W. Sutton. Dept. Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331. Phytopathology 92:S33. Publication no. P-2002-0237-AMA.

Phytophthora ramorum, agent of sudden oak death, causes girdling cankers on tanoak (*Lithocarpus densiflorus*) in Oregon. As a test of potential host range, we inoculated freshly cut logs, 10–20 cm dia. and about 1 m long, of native conifer and angiosperm tree species. Plugs of bark (5 mm dia.) were removed to the cambium, and a matching plug of colonized agar (or sterile agar) was inserted, then covered by wet gauze and foil. Bark was stripped after 5 weeks and extent of phloem necrosis was measured. Isolations were made from canker margins. Necrotic area was greatest on tanoak and Port-Orford-cedar. Smaller lesions developed on California black oak, Oregon white oak, chinquapin, and canyon live oak. Average lesion areas were at least 10 times greater on tanoak than on the *Quercus* species tested. Lesion breadth on tanoak was nearly as great as lesion length, but lesions were much narrower on other hosts. Lesion extent depended on time of year that logs were collected. Other hardwoods and conifers did not support expanding lesions. *P. ramorum* was regularly reisolated from lesion margins. Four *P. ramorum* isolates tested did not differ significantly in lesion area.

Biological control of sugar beet damping-off with *Trichoderma* species. L. E. Hanson. USDA-ARS, NPA, SBRU, Fort Collins, CO 80526. Phytopathology 92:S33. Publication no. P-2002-0238-AMA.

Isolates of *Trichoderma virens* and other *Trichoderma* species are effective biocontrol agents for diseases of several crops. Control of damping-off, caused by *Rhizoctonia solani*, has been observed in a number of crop species. We examined the effect of seed treatment with *Trichoderma* preparations on the survival of sugar beet seedlings under severe *Rhizoctonia* pressure. All of the *Trichoderma* isolates colonized sugar beet radicles and young roots well. Some fungal isolates significantly improved seedling emergence and survival in greenhouse tests. There was no correlation between antibiosis against *R. solani* *in vitro* and biological control activity. Tests are ongoing to examine biological control activity under field conditions.

Use of phage display for isolation of potential geminivirus resistance genes. S. F. HANSON, B. N. Milde, and D. P. Maxwell. Dept. of Plant Pathology, Univ. Wisconsin-Madison 53706. Phytopathology 92:S33. Publication no. P-2002-0239-AMA.

Geminiviruses are a group of small ssDNA viruses that cause severe disease problems throughout the tropical regions of the world and for which sources of useful natural resistance are limited. Previous results have shown that pathogen-derived resistance to geminiviruses may be limited by the narrow spectrum of resistance conferred by geminivirus-derived resistance genes. To overcome this limitation and to address safety concerns associated with pathogen-derived resistance, alternative strategies to create non-pathogen derived resistance genes are being evaluated. Phage display was used to isolate small peptide ligands that bind to a highly conserved stem-loop structure within the geminivirus origin of replication, which has been shown to be the site where new DNA synthesis is initiated during replication. Ligand binding was specific for the stem-loop structure as none of the ligands bound significantly to the same sequence in dsDNA. These peptide ligands may be useful for the construction of broad-spectrum geminivirus resistance genes, since the stem-loop sequence is almost perfectly conserved among all known geminiviruses.

Vegetative compatibility groups of *Verticillium dahliae* isolates from strawberry in California. J. J. HAO, J. M. Duniway, and D. M. Dopkins. Dept. Plant Pathology, Univ. of California, Davis, CA 95616. Phytopathology 92:S34. Publication no. P-2002-0240-AMA.

About 80 isolates of *Verticillium dahliae* were collected from strawberry in all major production areas of California. Pure cultures were obtained by single sporing. Spontaneous nit-mutants were selected using either minimal medium with chlorate or water agar chlorate medium. Vegetative compatibility group (VCG) was determined using standard VCG tester strains. Groups VCG2A and VCG2B were found only in the northern production area near Watsonville. VCG4A and VCG4B were found in both the southern (Oxnard, Irvine, Santa Maria) and the northern production areas. VCG1 and VCG3 were not detected. All but 2 of 35 isolates tested by root dip inoculation were pathogenic to strawberry, although the virulence varied among isolates. There was no correlation between pathogenicity and VCG. Molecular methods are being used to further examine the variation among isolates of *V. dahliae* on strawberry in California.

Effects of rhizobacteria on inhibition of soilborne pathogens and growth of strawberry. J. J. HAO (1), J. M. Duniway (1), D. M. Dopkins (1), and C. L. Xiao (2). (1) Dept. Plant Pathology, Univ. of California, Davis, CA 95616; (2) Washington State Univ., Tree Fruit Res. and Ed. Ctr., Wenatchee, WA 98801. Phytopathology 92:S34. Publication no. P-2002-0241-AMA.

Bacteria were isolated from the rhizospheres and root tissues of strawberry grown in methyl bromide fumigated and non-treated fields, as well as from bulk soil. Following fumigation, fluorescent *Pseudomonas* spp. quickly reached high populations in soil and on roots. Isolates were identified using the MIDI-GC Microbial Identification System and tested for antibiosis in cultures with *Verticillium dahliae*, *Phytophthora cactorum*, and species of *Pythium*, *Rhizoctonia*, *Fusarium*, *Cylindrocarpum*, and *Colletotrichum* isolated from strawberry. Many bacteria, especially *Pseudomonas* spp. from rhizospheres, had antibiosis to one or more fungi, but few isolates inhibited *Pythium* sp. and *P. cactorum*. Among bacterial isolates, there was a correlation between antibiosis and growth promotion of inoculated strawberry in natural soil under controlled conditions. Bacterial growth promotion of strawberry following inoculation in the field was variable and depended on soil background treatment and/or location, as well as isolate.

Effect of triiodobenzoic acid on broomrape (*Orobanche ramosa*) infection and development on tomato plants. A. M. Harb (1), K. M. HAMEED (1), and R. Shibli (2). (1) Dept. Applied Biological Sciences; (2) Dept. Plant Production, Jordan University of Science and Technology, Irbid 22110. Phytopathology 92:S34. Publication no. P-2002-0242-AMA.

Broomrape (*O. ramosa*) forms tubercles at point of attachments on its host plant which starts as undifferentiated callus. Indole-3-acetic acid (IAA) was found at early stages of tubercle development. Triiodobenzoic acid (TIBA), as an inhibitor of IAA polar transport, was used to investigate the role of IAA in tubercle development on tomato plant. TIBA, 10 mg/g in lanolin, was pasted at stem base of tomato, 2, and 4 wks after transplant and *Orobanche* inoculation. Seven weeks later, no differences in infection incidence per pot observed, but number of emerged *Orobanche* and attachments on root per plant was reduced by 60%, & 45% on TIBA treated plants, respectively. Xylem differentiation within tubercles on TIBA treated plants was delayed. But, when it happened, vessels were not in continuously aligned, and they were wider in diameter, and shorter than the control, with very little secondary wall deposition. Therefore, host is the source of IAA to tubercles in order to develop proper xylem vessels.

Evidence for the biological nature and host specificity of mature watermelon vine decline. R. HARIKRISHNAN, D. S. Egel, K. K. Rane, and R. D. Martyn. Dept. of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S34. Publication no. P-2002-0243-AMA.

Mature watermelon vine decline (MWVD), a disease of unknown etiology is characterized by root rot, wilting and collapse of mature vines. In Indiana, MWVD has been observed in both fumigated and non-fumigated fields and appears restricted to watermelons. In the greenhouse, 19-L pots filled with problem field soil were either fumigated (5g of Dazomet/pot) or non-fumigated and were transplanted (TP) with seedlings (3-wk old) of watermelon or muskmelon. Only watermelon in non-fumigated pots consistently wilted by the end of the study (9-wk after transplanting). A preliminary cucurbit host range study also was conducted in the greenhouse using 4-L pots filled with the problem soil. Three wk old seedlings of watermelon, muskmelon, squash, and pumpkin were TP into the soil. Plants were subjected to one of two different moisture regimes; normal (watered to promote growth) and wet (soil saturated weekly for 10 to 12 h). Only watermelon subjected to wet conditions consistently wilted 4-wk after transplanting. Based on these results, it appears that MWVD is biological in nature and is restricted to watermelon.

Influence of primary inoculum on epidemics of gray leaf spot on perennial ryegrass. P. F. HARMON and R. Latin. Department Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S34. Publication no. P-2002-0244-AMA.

Winter survival of *Magnaporthe grisea* (anamorph = *Pyricularia grisea*), has a large effect on primary inoculum, which in turn affects disease development during the summer. This investigation examines gray leaf spot epidemics in 2000 and 2001 at the Purdue Turf Research Center in West Lafayette, IN. Epidemic severity was assessed by counting airborne conidia collected over a four-month period with a volumetric air sampling device. The 2000 epidemic was initiated with infested residue on July 1, 2000. The 2001 epidemic was allowed to develop from inoculum surviving the winter. In both years conidia were detected in early July, however the 2000 epidemic was more severe. Despite the difference in severity, environmental conditions appeared equally favorable for disease development during both years. Efforts to recover the pathogen from infested residue during the winter and spring months of 2001 showed poor pathogen survival. It is likely that differences in primary inoculum levels contributed to differences in the disease epidemics.

Immunocapture RT-PCR as a confirmation tool for ELISA. A. M. Harness, J. C. JOHNSON, B. P. Kulemeka, C. L. Sutula, and M. D. Bandla. Agdia, Inc., 30380 County Road 6, Elkhart, IN 46514. Phytopathology 92:S34. Publication no. P-2002-0245-AMA.

Reliable plant diagnostic techniques are necessary for good crop management. Occasionally an ELISA may not give a definitive result, requiring confirmation using a different diagnostic method. Immunocapture reverse transcription PCR (IC-RT-PCR) allows for direct confirmation of an ELISA result in a simple, cost-effective manner. We found you can use the captured antigens from a microwell of a completed ELISA as a template in IC-RT-PCR, eliminating the need for traditional extraction methods and reducing the number of steps involved, costs incurred, and sampling variations. The ability to use an ELISA result for IC-RT-PCR provides an alternative when no additional plant tissue is available for confirmation testing. A person with a questionable ELISA result can send the ELISA microwell to a laboratory to be confirmed by IC-RT-PCR. This provides flexibility and leads to greater confidence in results. Experiments show that a variety of pathogens can be detected using ELISA in the IC-RT-PCR process. The ease of use, cost effectiveness, and added flexibility of IC-RT-PCR make it an important alternative for confirmation of ELISA results.

Incidence and identification of potential aphid vectors of Soybean dwarf virus in Illinois. B. HARRISON (1) and L. L. Domier (2). (1) Dept. of Crop Sciences; (2) USDA-ARS, University of Illinois. Phytopathology 92:S34. Publication no. P-2002-0246-AMA.

Soybean dwarf virus (SbDV) causes an important disease of soybeans in Japan and is persistently transmitted by aphids. In the United States, SbDV infects clovers. In 2000, *Aphis glycines*, an aphid species that colonizes soybeans, was reported in the Midwest. In 2001, we surveyed six species of clovers in 33 counties in the state of Illinois and tested them for the presence of SbDV by triple antibody sandwich enzyme-linked immunosorbent assay and polymerase chain reaction. We detected SbDV only in red clover (*Trifolium repens*), in 62% of the plants sampled. We also analyzed 87

soybean plants collected in Illinois in 2000, and 981 plants collected in 2001, but none tested positive for SbDV. We conducted transmission studies with aphid species that were found colonizing clover and soybean plants. Aphids of the species *Nearctaphis bakeri* and *Aphis craccivora* vectored SbDV among red clovers, and *N. bakeri* transmitted SbDV from red clover to soybean. However, *A. glycines* did not transmit SbDV; neither did two other clover-infesting aphid species, *Acyrtosiphon pisum* and *Therioaphis trifolii*.

Geographical distribution of Pierce's disease in North Carolina's winegrowing region. U. J. HARRISON, O. Anas, and T. B. Sutton. Dept. Plant Pathology, NC State Univ., Raleigh, NC 27695. Phytopathology 92:S35. Publication no. P-2002-0247-AMA.

Pierce's disease (PD) of winegrapes, caused by *Xylella fastidiosa*, is well documented in the Coastal Plain of NC. A survey was conducted in 2001 to identify the range of the pathogen in vineyards at 300 to 600 m elevation in the Piedmont and Mountain regions, where a continuum of low to moderate average winter temperatures occurs. In 15 of 21 vineyards, PD symptoms were identified, and *X. fastidiosa* was confirmed with ELISA. Negative ELISA results were largely limited to vineyards in the Mountains where winters are more severe. On this basis, and the fact that vinifera grapevines have existed at high elevations (>600 m) in NC for 20+ years with no history of PD, it is our hypothesis that areas of transition exist in NC where *X. fastidiosa* dies out or its population is reduced in infected vines during the winter. Various measures of minimum temperatures from 44 weather stations in the Mountains and Piedmont were analyzed using ArcView GIS 3.2. Data are presented which identify high-risk areas according to specific isotherms. The incidence of PD in the Piedmont may be related to a shift in isotherms due to an increase in mean temperature over the past 25 years.

Reducing root disease problems in dry beans by new methods aimed at alleviating soil compaction. R. M. HARVESON, J. A. Smith, E. S. Blehm, and R. Gatch. University of Nebraska Panhandle Research and Extension Center, Scottsbluff, NE 69361. Phytopathology 92:S35. Publication no. P-2002-0248-AMA.

A study was conducted at the Panhandle Research and Extension Center in Scottsbluff, NE in 2001 to investigate integrated methods for reducing root health problems in dry-edible beans in the presence of a purposely-compacted field. Eight treatments were utilized that included four tillage treatments and four combinations of tillage and/or fungicide (Quadris) applications. Tillage treatments consisted of a compacted control with no additional tillage, formation of beds 10 cm above soil surface, zone tillage with an implement using in-row shanks, and a combination of zone tillage and bedding. Fungicide treatments included one, two, and three applications combined with both zone tillage and bedding, and fungicide alone. Mortality counts and disease ratings were performed in late August, and plots were harvested in mid-September. Disease ratings, plant mortality, and seed yield were all significantly improved compared to the compacted control using zone tillage, but not bedding. Quadris applications did not provide any added advantages for improving plant health or performance.

***gacA* regulates plant interaction, motility and extracellular polysaccharide of *Erwinia amylovora*.** H. HASEGAWA, Y. Cui, A. Chatterjee, and A. K. Chatterjee. Dept. of Plant Microbiology and Pathology, Univ. of Missouri, Columbia, MO 65211. Phytopathology 92:S35. Publication no. P-2002-0249-AMA.

gacA and *gacS*, genes for a conserved two-component regulatory system, occur in *E. amylovora* (Ea) and *E. carotovora* subsp. *carotovora* (Ecc). To determine the phenotypes controlled by GacA, we constructed by marker exchange a GacA⁻ derivative of Ea strain E9. The GacA⁻ mutant, compared to E9, produces reduced levels of EPS and exhibits reduced swarming motility. The mutant requires higher cell density than the GacA⁺ parent to elicit hypersensitive response (HR) in tobacco and *Arabidopsis* leaves. The mutant, compared to the wild type, produces low levels of *hrpL* (alternative sigma factor), *hrpN* (harpin), *rsmB* (regulatory RNA), *dfoA* (desferrioxamine synthesis) and *rpoS* (sigma-S) transcripts. Motility, EPS production, and the ability to elicit the HR are restored in the mutant by the GacA⁺ plasmid, pAKC2000. In Ea and Ecc, the regulatory effects of GacA appear to be channeled via *rsmB* RNA.

Population dynamics of *Burkholderia cepacia* in organic soil. J. S. HAUDENSHIELD, N. A. Gundersheim, G. L. Mark, and J. W. Lorber. Dept. Plant Pathology, Cornell University, Ithaca, NY 14853. Phytopathology 92:S35. Publication no. P-2002-0250-AMA.

Onion producers suffer persistent losses from infection by *B. cepacia*, currently the most important bacterial pathogen of onions in New York. Soilborne *B. cepacia*, deposited in the leaf axial by wind or rainsplash, infects the plant, causing bacterial canker and, as infection advances into the bulb, sour skin. We hypothesize that cultivation of non-onion rotations, cover crops, or green manures may reduce soilborne inoculum. Onion field surveys spanning several consecutive seasons show a transient rise in *B. cepacia* counts. Similar surveys of soils historically planted to non-onion crops and of onion fields rotated with other crops show very low levels of *B. cepacia*. We have devised a cultivation method, miniculture, that facilitates quantification of the entire rhizosphere population of *B. cepacia* for individual plantlets grown in a known amount of soil with a known amount of inoculum. Various crops were grown in miniculture, and in some cases the crop was incorporated back into the soil as a green manure. The results of this research show a great influence on the bacterial population by some crops, generally supporting the hypothesis.

Spatial distribution of *Pratylenchus crenatus* in a carrot field and effect on yield and quality. F. S. HAY and S. J. Pethybridge. University of Tasmania. Phytopathology 92:S35. Publication no. P-2002-0251-AMA.

The effect of *Pratylenchus crenatus* on carrot was assessed in a trial consisting of 42 plots planted with variety 'Koyo' on 1/1/2001. Nematodes were extracted from soil and roots at 58 and 116 days after planting (DAP). The average, minimum and maximum number of *Pratylenchus*/400 ml soil at 58 DAP was 173, 40 and 446 respectively, and at 116 DAP was 160, 30 and 490 respectively and for *Pratylenchus*/g dry weight of root at 58 DAP was 99, 0 and 261 respectively. At 58 DAP, there was a correlation ($P = 0.05$) between *Pratylenchus*/g dry weight of root and dry weight of roots ($r = -0.475$) and foliage/plant ($r = -0.428$) at 58 DAP, and with the weight of twisted carrots ($r = 0.438$) at 123 DAP. There was a negative correlation ($P = 0.05$) between *Pratylenchus*/400 ml soil at 58 DAP and plants/m row ($r = -0.450$) and estimated total yield (t/ha) ($r = -0.378$) at 123 DAP. Numbers of *Pratylenchus* were not correlated with average carrot weight, percentage marketable carrots or weight of small, stunted, forked, cracked or constricted carrots. Spatial analysis by SADIE indicated an aggregated distribution of *Pratylenchus* in the soil at all times ($I_a = 1.53-1.64$, $P = 0.002-0.008$).

Salicylic acid-induced priming for potentiated responses to Fusarium infection in *Asparagus officinalis*. C. Y. HE and D. J. Wolyn. Dept. Plant Agriculture, University of Guelph, Guelph, ON N1G 2W1, Canada. Phytopathology 92:S35. Publication no. P-2002-0252-AMA.

Pre-inoculation of asparagus roots with nonpathogenic *Fusarium oxysporum* (npFo) led to induction of systemic resistance and protection against *F. oxysporum* f. sp. *asparagi* (Foa). As a first step towards elucidating the defense pathways through which npFo-mediated resistance was induced, the influence of SA on activation of defense responses in roots was investigated. *In vitro* assays indicated no effects of SA (3,000 ppm) on fungal mycelial growth. After soil drench pretreatment with SA, then inoculation with Foa, roots exhibited enhanced Foa resistance in a dose-dependent manner but no hypersensitive epidermal cell death. Induction of peroxidase and phenylalanine ammonia-lyase activities and lignification was minimal with SA treatment, however, SA-treated plants were predisposed to respond to Foa infection. Higher activities for POX and PAL and lignification were observed after Foa infection of SA-treated plants compared to those with no SA treatment. The ability to respond to Foa was induced within 8 h of SA treatment and the potentiated state was maintained for at least 48 h. Similar responses were observed after roots were pretreated with npFo, then infected with Foa. Interestingly, the potentiation by npFo pre-inoculation was not found in plants treated with a SA synthesis inhibitor. The results suggest both SA and/or npFo-induced priming is an important cellular mechanism for induced systemic resistance in asparagus.

Discovery and characterization of a necrotic response on leaves of *Nicotiana trigonophylla* induced by the tobacco blue mold pathogen, *Peronospora tabacina*. E. P. HEIST, W. C. Nesmith, and C. L. Schardl. Dept. Plant Pathology, University of Kentucky, Lexington, KY 40546. Phytopathology 92:S35. Publication no. P-2002-0253-AMA.

Peronospora tabacina causes downy mildew disease on several species of *Nicotiana*, including *N. tabacum* (tobacco). The primary objective of this study was to examine several wild *Nicotiana* for novel responses to *P. tabacina* infection. After examining 14 different species of *Nicotiana*, we discovered that one species, *N. trigonophylla*, produced a necrotic response 5-6 days following foliar inoculation with *P. tabacina* sporangiospores. This necrotic response was characterized by rapid necrosis, coincided with the time required for sporulation of the pathogen and could be induced by multiple isolates of *P. tabacina*. Plants needed to be at least one month old

before necrotic response could be induced and inoculation with water or killed spores did not result in the response. This reaction to *P. tabacina* infection appears both interesting and relevant for future research and might prove useful as a source of resistance if moved into commercial tobacco.

Gene expression profile of pathogenicity factors in *Xylella fastidiosa*. R. HERNANDEZ-MARTINEZ, C. K. Dumenyo, and D. A. Cooksey. Dept. Plant Pathology, University of California, Riverside, CA 92521. Phytopathology 92:S36. Publication no. P-2002-0254-AMA.

Xylella fastidiosa (*Xf*) strains cause several economically important plant diseases including, Pierce's disease on grape (PD), citrus variegated chlorosis (CVC), and oleander leaf scorch (OLS). Although the whole genomic sequence of the CVC strain has been published, no functional characterization of *Xf* genes has been performed. To identify genes involved in pathogenicity in the PD strain, the sequence of the CVC strain was used to select open reading frames specifying putative pathogenicity and virulence factors. DNA fragments of these genes were obtained by PCR amplification of the genome of the PD strain. Currently we are constructing arrays of these genes to perform analyses of gene expression between PD and OLS strains, pathogenic and non-pathogenic strains, and host responses in grape and oleander. This study will help in the determination of the genes whose expression changes following infection and to understand bacterial adaptation to varied environmental conditions thus providing an insight into the disease process.

A low titer RNA entity in a *Citrus tristeza virus*-infected Mexican lime plant with similarities to oat blue dwarf virus and grapevine fleck virus. C. M. HERRON (1,2), B. M. da Graça (2), J. V. da Graça (1), R. G. Shatters Jr. (3), and T. E. Mirkov (2). (1) Texas A&M University-Kingsville Citrus Center, Weslaco, TX 78596; (2) Dept. Plant Pathology and Microbiology, Texas A&M University Agric. Expt. Stn., Weslaco, TX 78596; (3) USDA, ARS U.S. Horticultural Research Laboratory, Ft. Pierce, FL 34945. Phytopathology 92:S36. Publication no. P-2002-0255-AMA.

A cDNA with high similarity to part of the *Oat blue dwarf virus* (OBDV; genus *Marafivirus*) genome was obtained from dsRNA extracts of a *Citrus tristeza virus*-infected plant. Northern analyses using the source plant total RNA and RNA extracts from virus purification fractions revealed a genomic RNA of at least 7.5 kb. OBDV coat protein (CP) antibodies react weakly with protein extracts from the source plant and produce a specific band of approximately 28 kDa. Grapevine fleck virus (GFkV; unassigned virus) has many features common to marafiviruses and a 7.5 kb genomic RNA. GFkV CP antibodies do not react with protein extracts from the source plant by ELISA. The entity appears non-graft transmissible to citrus, and a host range study to herbaceous plants concluded the entity could not be mechanically transmitted.

Assessments of Rio Red grapefruit scions with a *Citrus tristeza virus* untranslatable coat protein transgene for resistance to the virus. C. M. HERRON (1,2), Z. N. Yang (2), J. Molina (2), J. V. da Graça (1), S. P. van Vuuren (3), and T. E. Mirkov (2). (1) Texas A & M University-Kingsville Citrus Center, Weslaco, TX 78596; (2) Dept. Plant Pathology and Microbiology, Texas A&M University Agric. Expt. Stn., Weslaco, TX 78596; (3) Institute for Tropical and Subtropical Crops, Nelspruit, South Africa. Phytopathology 92:S36. Publication no. P-2002-0256-AMA.

Transgenic scions representing separate transgenic events using an untranslatable coat protein sequence (*uncp*) from *Citrus tristeza virus* (CTV) isolate SY568 have been generated. Under controlled conditions in Texas with wild type scions as controls, *uncp* scions have been grafted to virus-free rootstocks in duplicate, and challenged with a severe Texas CTV isolate. Five *uncp* sources have also been graft inoculated with mild and severe South African CTV isolates under quarantine conditions in South Africa. Assessments are by CTV coat protein ELISA. Some of the transgenic scions have a relative decrease in CTV titer compared to controls. Northern hybridizations of transgenic scion total RNA to a probe made from the transgene also had weak signals for these samples.

Evaluation of three bacteriocins in antagonism of T3 strains to T1 strains of *Xanthomonas campestris* pv. *vesicatoria*. A. P. HERT (1), S. Tudor (1), P. D. Roberts (2), G. V. Minsavage (1), and J. B. Jones (1). (1) Univ. of Florida, Gainesville; (2) Univ. of Florida, Immokalee. Phytopathology 92:S36. Publication no. P-2002-0257-AMA.

Xanthomonas campestris pv. *vesicatoria* tomato race 3 (Xcv T3) strain 91-118 produces at least three different bacteriocin-like compounds (BCN-A, BCN-B, BCN-C) antagonistic to Xcv tomato race 1 strains. Bacteriocin activity was disrupted by either transposon mutagenesis or PCR rearrange-

ment assisted mutagenesis and marker exchanged into a T3 strain using a suicide vector method. In greenhouse experiments the wild-type (wt) T3 strain and mutants expressing all possible combinations of bacteriocins were evaluated for antagonism toward T1 strains. All mutant and the wt T3 strains reduced the T1 population when inoculated in combination; however, wt T3 and BCN-AC were the most effective. A field trial was conducted in the spring of 2001 in which the wt T1, wt T3, BCN-AB, BCN-AC, BCN-BC, and a triple knockout (BCN⁻) were inoculated on the same plants. Lesions were sampled periodically and strains were isolated. Nine weeks after inoculation, BCN-AC was recovered from 80% of the lesions, whereas all other strains were less than 10%. T1 strains were not recovered.

Molecular evidence for multiple origins of decline isolates of CTV in Florida. M. E. Hilf. USDA-ARS-USHRL, Ft. Pierce, FL. Phytopathology 92:S36. Publication no. P-2002-0258-AMA.

Citrus tristeza virus (CTV) is widely dispersed in commercial citrus in Florida, yet the origin of CTV in Florida is unknown. A CTV-induced incompatibility of trees grafted on sour orange rootstock causes economic loss in Florida due to decline in tree productivity, tree vigor and eventual death of the tree. We have developed a PCR-based genetic marker assay that discriminates between CTV isolates by amplification of sequence specific genetic markers. Using marker analysis, we determined that in Florida, the graft incompatibility is associated with infection by virus isolates having a T36 marker profile (genotype). An evaluation of an international collection of CTV isolates indicated that the T36 genotype, while common in Florida, is rare worldwide. In Florida and elsewhere, the T36 genotype also was found in Meyer lemon trees, a citrus variety that is common as a dooryard plant. Sequence analysis of markers amplified from commercial trees in Florida, Meyer lemon trees from diverse locations and calamondin sources in the international CTV collection has provided evidence for two possibly separate introductions of the T36 genotype into Florida.

Resistance to the soybean aphid in soybean and other legumes. C. B. HILL (1), C. R. Ferro (1), Y. Li (1), and G. L. Hartman (1,2). (1) Dept. Crop Sciences, University of Illinois, Urbana, IL 61801; (2) USDA-ARS. Phytopathology 92:S36. Publication no. P-2002-0259-AMA.

The soybean aphid (*Aphis glycines* Matsumura) was first reported in the midwestern USA in 2000 and quickly became an important pest of soybean (*Glycine max* (L.) Merrill). It is the only aphid that can colonize soybean and reduces crop yield by its feeding activity and the transmission of viruses. Resistance to the aphid would be desirable to include with other control measures. In a greenhouse screen, less than 2% of 798 MG II and III commercial soybean cultivars appeared to be resistant while over 75% of the cultivars were very susceptible. In another test, four of 95 soybean ancestral lines appeared to be resistant. A test of other cultivated legumes revealed that white clover, red clover, and alfalfa, were susceptible to the aphid. These tests indicated that the availability of resistance to the soybean aphid was limited in this germplasm and the scope of evaluation needs to be expanded.

Fungal diversity in woody roots of east-slope Cascade Mountain Douglas-fir and ponderosa pine. J. A. HOFF (1,2), N. B. Klopfenstein (1), G. I. McDonald (1), J. R. Tonn (1), P. J. Zambino (1), J. D. Rogers (2), T. L. Peever (2), and L. M. Carris (2). (1) USDA Forest Service/RMRS, Moscow, ID; (2) Dept. Plant Pathology, Washington State University, Pullman, WA. Phytopathology 92:S36. Publication no. P-2002-0260-AMA.

Fungal diversity within woody roots of Douglas-fir (*Pseudotsuga menziesii*) and ponderosa pine (*Pinus ponderosa*) in the dry forests of the east-slope Cascades was studied using increment cores. Examination of rDNA internal transcribed spacer sequences and morphology of the cultured fungi delineated 26 fungal genera. Two groups predominated: *Umberlopsis*-like species (22% of isolations), and *Byssochlamys* species (21% of isolations). The predominance of these genera was not affected by host or habitat type. More information is needed regarding the ecological role of these fungi, their relationship with fire, and their potential for biological control of forest pathogens.

Validation of a strawberry powdery mildew risk model. L. E. HOFFMAN, T. C. Miller, K. J. Dell, and W. D. Gubler. Dept. of Plant Pathology, University of California, Davis, CA 95616. Phytopathology 92:S36. Publication no. P-2002-0261-AMA.

To validate a powdery mildew (PM) risk model, a trial was conducted in a 'Camarosa' strawberry field in Oxnard, CA. The risk index (RI) accumulates 20 points for every day that temperature is between 18-27°C for at least 4 hrs. When these conditions are not met, 10 points are deducted from the RI. Rally 40W alternated w/Benlate 50 WP was applied within 24 hr. anytime

the RI hit 60. Fungicides were applied 5x in this treatment as compared to 12x for the standards applied on a 14-d schedule. During the final disease assessment, the PM incidence in the model treatment (2.7%) was not significantly lower than in the untreated check (4.9%), but was significantly greater than the BAS 516 and Quintec standards (0.1 and 0.6%, resp.). However, marketable yield was not significantly different between the fungicide treatments, and all 3 were significantly greater than the untreated check. These results indicate that the PM risk model may be useful in reducing the number of fungicide applications required. Because Rally/Benlate applied on a 14-d schedule provided significantly less control than BAS 516 or Quintec, these latter compounds may be better choices for use with the model.

Tagging flowers to validate an infection risk model for managing strawberry gray mold. L. E. HOFFMAN, K. J. Dell, and W. D. Gubler. Dept. of Plant Pathology, University of California, Davis, CA 95616. Phytopathology 92:S37. Publication no. P-2002-0262-AMA.

Gray mold (*Botrytis cinerea*) at harvest is likely a result of flower infections that remain latent unless environmental conditions are conducive for symptom development prior to harvest. To test this hypothesis and validate an infection risk model, the fruit of appr. 500 unsprayed plants (cv. San Juan) were picked weekly and rated for botrytis. Onsite weather was recorded with an Adcon A730 station. In regression analyses, avg. leaf wetness duration during the week prior to harvest was a better predictor of rot ($R^2 = 0.62$) than the avg. temperature during wetness ($P = -0.5$) or the Botrytis risk index (combining both variables, $P = -0.22$). To calculate the botrytis incidence for each flowering \times harvest period, fresh flowers were tagged weekly with colored tape and the number of each color tag recovered was recorded during rating. When botrytis incidence was regressed against avg. leaf wetness prior to harvest and the avg. risk index during the week of flowering, the R^2 value was low (0.49) indicating that other variables may influence the botrytis incidence at harvest.

Susceptibility of watermelon cultigens to ambient ozone in eastern North Carolina. G. J. HOLMES (1) and J. R. Schultheis (2). (1) Dept. Plant Pathology, NC State Univ., Raleigh, NC 27695; (2) Dept. Horticultural Science, NC State Univ., Raleigh, NC 27695. Phytopathology 92:S37. Publication no. P-2002-0263-AMA.

Foliar ozone injury is common on watermelon grown in eastern North Carolina. In 2000 and 2001, we evaluated 93 cultivars and breeding lines (i.e., cultigens) for their sensitivity to visible foliar injury. One tetraploid, 42 diploid and 50 triploid cultigens were evaluated. Injury developed on all cultigens in both years. Visible foliar injury (% surface area necrotic/chlorotic) was rated one week after the first harvest in both years. Mean injury for all cultigens was 39% in 2000 (range = 16 to 66%) and 20% in 2001 (range = 2.5 to 60%). This corresponded to greater seasonal ozone levels in 2000 (58 ppb) than in 2001 (52 ppb). Triploid cultigens were less susceptible to injury than diploid cultigens. Mean injury for all triploids was 31% in 2000 and 16% in 2001, while injury on diploids was 47% in 2000 and 25% in 2001. Injury level was consistently negatively correlated with average number of days between transplanting and harvest. Correlations were also detected between injury and total harvested weight, mean fruit weight and length-diameter ratio, but not for soluble solids.

Effect of some pesticides on the mycoherbicide, *Epicoccossorus nematosporus* and synergistic effect in combination with herbicides on *Eleocharis kuroguwai* control in rice paddy field. Y.-K. HONG (1), S. B. Song (1), B. C. Lee (1), S. C. Kim (1), and J. Y. Uhm (2). (1) Plant Environment Division, National Yeongnam Agricultural Experiment Station (NYAES), RDA, Milyang 627-803, Korea; (2) Kyungpook National Univ. Daegu, 702-701, Korea. Phytopathology 92:S37. Publication no. P-2002-0264-AMA.

This study was initiated to determine the effects of the factors related to synergistic effect of the fungus with herbicide on the control of *Eleocharis kuroguwai*. We examined the effect of pesticides on mycelial growth, appressorial formation and the synergistic control efficacy of *E. nematosporus* with a herbicides, bentazon against water chestnut each treatment. *Epicoccossorus nematosporus* was more sensitive to the fungicides; tricyclazole and neosozin than both insecticides; phanthoate and BPMC and a herbicide; dithiopyr. EC_{50} values of *E. nematosporus* for tricyclazole and neosozine were 0.26 and 0.24 $\mu\text{g/ml}$, respectively. Two fungicides, tricyclazole and neosozin, affected appressorial formation of the fungus continuing until 15 days after application, while an insecticide phanthoate, BPMC and a herbicide, dithiopyr, had no effect after 3 days. In three years' field observations, a sequential application of the fungicides, tricyclazole and neosozine inhibited natural occurrence of the fungus. Weeding efficacy were much differences resulted from the combinations of conidia inoculum of *E. nematosporus* with a herbicide bentazon [3-(1-methylethyl)-(1H)-2,1,3-

benzothiadiazin -4(3H)-one]. The highest weeding efficacy was obtained in the mixture of bentazon (0.3%) and conidial inoculum comparing to that off the treatment of bentazon (0.4%) alone. Combined treatments of the conidial inoculum ($6.0 \times 10^5/\text{ml}$) with a herbicide bentazon (0.3%) killed most of weeds, and also suppressed number of underground tubers. Two applications of the conidial inoculum at 3-day-interval were the most effective for the weed control with plant mortality of 95.6%.

Geostatistical analysis of dollar spot: towards a new understanding. B. J. HORVATH (1), A. N. Kravchenko (2), and J. M. Vargas, Jr. (1). (1) Dept. of Plant Pathology, Michigan State University E. Lansing, MI 48824; (2) Dept. of Crop and Soil Sciences, Michigan State University, E. Lansing, MI 48824. Phytopathology 92:S37. Publication no. P-2002-0265-AMA.

Dollar spot, caused by *Sclerotinia homoeocarpa*, is the major turfgrass pathogen on cool season turfgrasses in Michigan. The disease is characterized by small, silver dollar sized spots of infected tissue that result in an uneven surface affecting golf ball roll. As fungicides become more limited in their ability to treat for this disease, it is critical that we understand the epidemiology of this disease. Our objective was to observe the dollar spot epidemic and determine if dollar spot incidence is clustered or random. Our study was conducted on a 30' \times 60' area of untreated turf at the Robert Hancock Turfgrass Research Center at MSU. Dollar spot was allowed to develop without fungicide applications and diseased spots were counted at regular intervals on a grid overlaying the site. Geostatistical analyses were conducted on the count data. The analysis indicates that dollar spot does appear to be clustered in appearance. Further analysis will indicate what other factors might be responsible for this clustering.

Cloning putative parasitism genes expressed in the esophageal glands of *Meloidogyne incognita*. G. HUANG (1), B. Gao (1), T. Maier (2), E. L. Davis (3), T. J. Baum (2), and R. S. Hussey (1). Depts. of Plant Pathology, (1) University of Georgia, Athens, GA 30602; (2) Iowa State University, Ames, IA 50011; (3) North Carolina State University, Raleigh, NC 27695. Phytopathology 92:S37. Publication no. P-2002-0266-AMA.

Cloning parasitism genes coding proteins secreted from the esophageal glands and injected through the stylet into plant tissue is the key to understanding the molecular basis of nematode parasitism of plants. Microaspiration of cytoplasm from the glands of 43 parasitic stages of *M. incognita* provided mRNA for RT-PCR to construct a long-distance cDNA library. Of 1,200 cDNAs sequenced, deduced protein sequences of 141 cDNAs were preceded by a secretion signal peptide and, thus, may function in nematode parasitism of plants. In mRNA in situ hybridizations, 21 cDNAs coding signal peptides specifically hybridized with mRNA within the subventral (7) or dorsal (14) glands. PSORT II predicted 18 deduced proteins to be extracellular and 3 as nuclear localized. In BLASTp analyses, 19 predicted proteins were novel and two had similarities to a transcription factor and an avirulence protein. None of the gland-expressed genes had homologues in *Caenorhabditis elegans*.

Identification of a promoter region from *Citrus yellow mosaic virus*. Q. HUANG (1) and J. S. Hartung (2). USDA-ARS, (1) USDA, Floral and Nursery Plants Research Unit; (2) Fruit Laboratory, Beltsville, MD 20705. Phytopathology 92:S37. Publication no. P-2002-0267-AMA.

Bacterial diseases often result in significant losses in the production and quality of ornamental crops, and are very difficult to control. The use of genetically engineered crops offers a novel and effective strategy for control of bacterial diseases. Commercialization of such crops, however, is limited by the use of promoters that have licensing restrictions. To obviate licensing problems, we isolated a promoter fragment from a cloned isolate of *Citrus yellow mosaic virus* (CYMV). This was done by fusing different regions of the CYMV genome to the coding region of the GUS gene and a CYMV region with strong promoter activity was identified by analyzing the amount of transient GUS gene expression in tobacco leaves transformed by particle bombardment. We are in the process of comparing the strength of the CYMV promoter with that of the *Cauliflower mosaic virus* 35S promoter, and determining the tissue specificity of the CYMV promoter in transgenic tobacco plants. These studies are important steps toward the development and successful commercialization of transgenic ornamental crops for bacterial resistance.

Molecular characterization of a mutation in *Aspergillus flavus* causing the dominant repression of aflatoxin biosynthesis. G.-H. Huh (1), J. E. Flaherty (2), and C. P. WOLOSHUK (2). (1) Center for Genetic Engineering, Inje University, 607, Kimhae, Korea; (2) Dept. Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S37. Publication no. P-2002-0268-AMA.

Aspergillus flavus strain 649 has a genomic DNA deletion greater than 150 kb including the 75 kb aflatoxin biosynthesis gene cluster. Diploids formed between strain 649 and aflatoxigenic strain 86 do not produce aflatoxin. To address the possibility that the phenotype results from the expression of a repressor gene in strain 649, a vector with the regulatory gene *afIR* and a *ver1* promoter fused to the GUS reporter gene was inserted into the genome of strain 649. One transformant having a single copy of the reporter was crossed with strain 86 to generate diploids. The diploid produced aflatoxin and stained positive for GUS. The data indicate that strain 649 does not produce a repressor, leaving the possibility that the repression phenotype results from inactivation of *afIR* due to homologous pairing of chromosomes. To further investigate this hypothesis, we isolated genomic DNA cosmids that contain the break-junction regions in strains 649 and 86 and sequenced across the break-junction region in each strain.

Effect of planting date, tillage and burning of residue on eyespot of winter wheat. R. M. HUNGER (1), L. L. Singleton (1), E. G. Krenzer (2), R. Sidwell (2), and M. E. Payton (3). Depts. of (1) Ent. and Plant Pathology, (2) Plant and Soil Sciences, and (3) Statistics, Oklahoma State University, Stillwater, OK 74078. Phytopathology 92:S38. Publication no. P-2002-0269-AMA.

This study evaluated the effect of planting date, tillage (disk and moldboard plow), and burning or no burning of residue on eyespot of winter wheat caused by *Pseudocercospora herpotrichoides*. Hard red winter wheat (cv. 2137) was planted early (20 Sep 00) and late (22 Nov 00) in a split-block with six replications in a field in which eyespot occurred uniformly; however, planting dates were not randomized correctly in order to facilitate sowing, tillage, and residue burning. Results indicated that eyespot severity, height, and fertile heads were greater in early-planted wheat, and that early planting in combination with disking and not burning residue increased lodging. Yield was highest in early-planted wheat because uncommonly cold weather for six weeks following the late planting inhibited tillering. Overall, these results confirm previous reports that tillage and burning of residue have a minimal, if any, effect on eyespot, and that eyespot is more severe in early-planted wheat.

begomovirus genomes associated with tomato leaf curl disease from the Nile Basin and evidence for interspecies recombination. A. M. IDRIS and J. K. Brown. University of Arizona. Phytopathology 92:S38. Publication no. P-2002-0270-AMA.

A full-length monopartite genome was cloned and the sequence was determined for three begomoviruses from tomato plants showing leaf curl symptoms in Central Sudan (SD). Two viral genotypes were identified from Gezira, SD, provisionally, Tomato leaf curl Sudan virus-1 (ToLCSDV1) and Tomato yellow leaf curl virus-Sudan (TYLCV-SD), and a third, ToLCSDV2, was identified from Shambat, SD. ToLCSDV1 shared ~90% nucleotide (nt) identity with its closest relative, ToLCSDV2, but the two diverged from TYLCV-SD, at ~82.8-83.1% nt identity. The closest relatives to ToLCSDV1 and ToLCSDV2 were members of the TYLCV begomovirus cluster at ~83%. TYLCV-SD from Gezira was ~93% identical with its closest relatives, TYLCV-Portugal and TYLCV-Japan. Recombination analysis (GENECONV) revealed two sites of recombination for ToLCSDV1 and TYLCV-SD, which were isolated from a mixed infection. Evidence for interspecies recombination in ToLCSDV1 and TYLCV-SD was corroborated by a topological shift in phylogenetic position for both species when predicted recombinatorial fragments were separately analyzed.

Aggressiveness of *Ophiostroma korrae* isolates from two genetically distinct populations to bermudagrass. F. B. IRIARTE (1), J. D. Fry (2), D. L. Martin (3), T. C. Todd (1), and N. A. Tisserat (1). (1) Dept. Plant Pathology; (2) Division of Horticulture, Kansas State University, Manhattan, KS 66506; and (3) Dept. Horticulture and Landscape Architecture, Oklahoma State University, Stillwater, OK 74078. Phytopathology 92:S38. Publication no. P-2002-0271-AMA.

Ophiostroma korrae is one of three reported causal agents of spring dead spot of bermudagrass. (*Cynodon spp.*). Based on AFLP analysis, isolates from the southeastern United States ('southern') clustered into a clade that was distinct from isolates collected in Kansas and Oklahoma ('northern'). On average, 19 southern isolates exhibited faster ($P = 0.0001$) radial growth than 15 northern isolates on potato dextrose agar at 25°C and 30°C. In greenhouse experiments, a susceptible (Tifgreen) and resistant (Midlawn) cultivar were inoculated with a mixture of southern or northern isolates. Plants inoculated with the southern isolates exhibited greater shoot mortality ($P \leq 0.05$) than plants inoculated with the northern isolates. No difference in shoot mortality among resistant and susceptible cultivars was detected. These results suggest there are regional differences in aggressiveness of *O. korrae* isolates.

Rapid Blight control on turf with trifloxystrobin and other registered fungicides. J. ISGRIGG III (1) and D. A. Spilker (2). Bayer, (1) Chesterfield, VA 23838, and (2) Kansas City, MO 64120. Phytopathology 92:S38. Publication no. P-2002-0272-AMA.

In winter 2000 and spring 2001, a new disease, Rapid Blight (proposed name), appeared and subsequently killed vast areas of perennial ryegrass, *Lolium perenne*, and rough bluegrass, *Poa trivialis*, over-seeded on several bermudagrass golf courses in South Carolina. The suspected pathogen for this new disease is a Chytridiomycete. Initial fungicide research conducted in California and at Clemson University showed that trifloxystrobin, Compass™, was one of the most effective treatments. Subsequent trials in fall 2001 on Kiawah Island, S.C., revealed treatments containing trifloxystrobin alone at 0.125 oz AI/ft² or in combination with either fosetyl-AI at 4.8 or 6.4 oz AI/1000 ft² or triadimefon at 0.25 or 0.5 oz AI/1000 ft² provided >85% control of Rapid Blight with increased turf quality. A tank-mix of trifloxystrobin at the aforementioned rate plus propamocarb hydrochloride at 2.25 or 3 oz AI/1000 ft² gave only modest control (>75%). Lower rates and other tested compounds provided poor control.

Antifungal glycoproteins in red-light irradiated broadbean leaflets. S. Z. ISLAM (1), Y. Honda (2), Y. Sawa (2), and M. Babadoost (1). (1) Dept. Crop Sci., Univ. of Illinois, Urbana, IL 61801; (2) Fac. Life and Env. Sci., Shimane Univ., Matsue, Japan. Phytopathology 92:S38. Publication no. P-2002-0273-AMA.

Red-light treatment of broadbean (*Vicia faba*) leaflets resulted in production of water soluble and heat stable substance(s) against *Botrytis cinerea*. Aliquots of 50- μ l of *B. cinerea* spore suspension (2×10^5 spores/ml) were placed on the surface of leaflets in moist plastic boxes, incubated at 24°C under continuous irradiation of red light (600-700 nm). After 48 h of irradiation, infection droplets (ID) were recovered and centrifuged. The antifungal substance(s) in ID was positively charged, as the antifungal constituent was removed by the cation exchanger CM-cellulose. Proteinase-k and glycosidases treatment of ID eliminated its antifungal activity, suggesting that both protein and carbohydrate are the active components of the substance(s). The HPLC:gel column analysis of ID resulted in four fractions, and all of them showed antifungal activity. This suggests that more than one antifungal compounds may be produced in red-light irradiated broadbean leaflets. The molecular weight of antifungal compound in fraction-IV was estimated to be 38.5 kDa by SDS-PAGE analysis.

Effects of *Heterodera glycines* population levels on *Fusarium solani* f. sp. *glycines* colonization of near-isogenic soybean lines differing in resistance to each pathogen. T. A. JACKSON (1), T. L. Niblack (1), and G. S. Smith (2). (1) Dept. of Crop Sciences, University of Illinois, Urbana 61801; (2) Pest Management Center Project Leader, Missouri Dept. of Agriculture, Jefferson City, MO 65102. Phytopathology 92:S38. Publication no. P-2002-0274-AMA.

Soybean sudden death syndrome (SDS), caused by *F. solani* f. sp. *glycines* (Fsg), is increasing in both severity and incidence in the Midwest resulting in greater yield losses. SDS often occurs in fields infested with the soybean cyst nematode (SCN), *H. glycines*, and is frequently exacerbated when both pathogens are present, although their interaction is not well understood. A study was conducted during 2000 and 2001 in a naturally infested field with 4 soybean isolines varying in resistance to each pathogen. Fsg colonization at flowering was very low and SDS foliar symptoms were absent during 2001, both of which may be attributed to late planting. SCN population densities were low to moderate at planting (239 eggs/100 cm³ soil), but increased markedly by harvest, particularly in SCN-susceptible isolines (maximum 41,104 eggs/100 cm³ soil). Fsg colonization at harvest and yield data will also be presented.

Aspects of UVR sensitivity and survival strategies of relevance to phyllosphere bacteria. J. L. JACOBS (1), C. D. Miller (2), T. L. Carter (1), A. J. Anderson (2), and G. W. Sundin (1). (1) Dept. Plant Pathology and Microbiol., Texas A&M University, College Station, TX 77843; (2) Dept. Biology, Utah State University, Logan, UT 84322. Phytopathology 92:S38. Publication no. P-2002-0275-AMA.

We examined the survival of a collection of *Pseudomonas* species and other phyllosphere bacteria following exposure to UV-C (direct DNA damage) or UV-A (indirect effects, oxidative damage) radiation. Plant-pathogenic *P. cichorii* and *P. syringae* were more tolerant to UV-A and UV-C exposure than pseudomonads isolated from animal hosts and soil. The UV-A survival of *P. syringae* pv. *syringae* B728a *sodAB* and *rpoS* mutants was reduced ~100-1,000 fold. Pigmented phyllosphere bacteria such as *Brevibacterium* sp., *Clavibacter michiganensis*, and *Pantoea agglomerans* were highly

tolerant to UV-A exposure. Results of field studies showed significant reductions in populations of a pigment-deficient mutant of *C. michiganensis* G7 compared to the wild-type, and that populations of UV-C sensitive *C. michiganensis* strains were consistently lower than those of UV-C tolerant strains. Our results demonstrate the importance of several classes of genes in conditioning UV survival and epiphytic fitness.

Prescribed burning effects on urban woodlands and *Armillaria*. K. A. JACOBS (1), M.-S. Kim (2), P. Gallegos (1) and M. L. Bowles (1). (1) The Morton Arboretum, Lisle, IL 60525; (2) Dept. Forest Resources, University of Idaho, Moscow, ID 83844. Phytopathology 92:S39. Publication no. P-2002-0276-AMA.

Prescribed burning is an important restoration tool in urban woodlands. We quantified tree vigor, prevalence of *Armillaria* and its genetic composition in 10 annually burned and 10 unburned 0.025 hectare plots in oak woodlands at The Morton Arboretum. The intergenic spacer (IGS-1) of ribosomal DNA sequences from *Armillaria* cultures derived from mushrooms and rhizomorphs indicated two species predominate: *A. mellea* and *A. calvescens* or *A. gallica*. Both were present in unburned plots, but only the latter was found in the burned stand. The prescribed burn in 2001 killed over 90 percent (%) of buried rhizomorphs and *Armillaria* abundance was less in the burned stand (2.3 mushroom clusters per burned plot versus 8.0 per unburned plot). Root colonization by *Armillaria* and somatic compatibility of isolates remain under study. Trunk cankers were also common, especially in burned (40% incidence) versus unburned (27%) plots and genera considered vulnerable to fire (*Tilia*, *Acer*, *Ostrya*) had higher disease severity ratings in burned than unburned plots.

A new member of the clover proliferation phytoplasma group associated with elm yellows in Illinois. K. A. Jacobs (1), I. M. LEE (2), H. M. Griffiths (3), F. Miller (4), and K. D. Bottner (2). (1) The Morton Arboretum, Lisle, IL 60532; (2) Molecular Plant Pathology Lab., USDA-ARS, Beltsville, MD 20705; (3) Cornell University, Ithaca, NY 14853; (4) Joliet Junior College, Joliet, IL 60431. Phytopathology 92:S39. Publication no. P-2002-0277-AMA.

A disease appearing similar to elm yellows (EY) has killed over 1000 American elm trees during the last decade in the Chicago metropolitan area. Irregular symptoms and negative detection test results led us to sample trees during 1998-2000 for EY phytoplasma. Nested PCR using universal primer pairs revealed that 14 of 15 trees sampled had phytoplasma and although they expressed a range of symptoms, all died within two years. Ten asymptomatic control elms tested negative. Phloem scraped from bark was superior to wood shavings or foliage for detecting phytoplasma. RFLPs and DNA sequencing of 16S rDNA indicated that the Illinois phytoplasma is not related to the EY phytoplasma group, but instead is a new member of the clover proliferation phytoplasma group (16SrVI). Neither EY, nor the newly designated Illinois elm yellow (ILEY) phytoplasmas were detected in leafhopper samples trapped from elms daily between May and September, 2000.

Aflatoxin contamination of commercial cottonseed in south Texas from 1997 to 2000. R. JAIME-GARCIA and P. J. Cotty. USDA-ARS-SRRC, 1100 Robert E Lee Blvd, New Orleans, LA 70124. Phytopathology 92:S39. Publication no. P-2002-0278-AMA.

Aflatoxins are toxic metabolites produced by *A. flavus*. Federal regulations limit the use of aflatoxin-contaminated cottonseed. Cottonseed with aflatoxin content of 20 ppb or higher may not enter the profitable dairy market. Between 4,472 to 9,949 truckloads of cottonseed from 31 to 35 gins in South Texas were analyzed for aflatoxin content each year from 1997 to 2000 upon receipt at the Valley Co-op Oil Mill in Harlingen, TX. Results revealed that aflatoxin contamination of commercial cottonseed presents both temporal and spatial variation. Highest levels of contamination occurred in 1999 with an average aflatoxin content of 112 ppb and 66% of the cottonseed exceeding 20 ppb. Years 1997 and 2000 had the lowest levels of aflatoxin contamination with an average of 24 ppb. The lowest occurrence of contamination was in 1997 with 16% of the truckloads exceeding 20 ppb. Geostatistical analyses revealed that the greatest contamination occurred in the northern and eastern Coastal Bend and the southern Upper Coast areas. The Rio Grande Valley area had the lowest contamination levels. In general, aflatoxin contamination increased as the ginning season progressed.

Role of sugars and other nutrients in germination of *Penicillium expansum* conidia with implication to biological control of postharvest decays on apple. W. J. JANISIEWICZ and T. J. Tworowski. USDA-ARS, Appalachian Fruit Research Station, Kearneysville, WV 25430. Phytopathology 92:S39. Publication no. P-2002-0279-AMA.

Competition for exogenous nutrients has been described as a putative mechanism of biological control of necrotrophic pathogen, *P. expansum*, which causes blue mold of apples after harvest. However, the role of the major carbon and nitrogen sources and various growth factors occurring at the wound site has not been established. We found that of the four main sugars in apple (glucose, fructose, sucrose and sorbitol), sucrose, by far, was the most stimulatory to conidia germination within the first 24 h in a well test. The germination occurred in the presence of the Hutner's solution containing various microelements. Rapid removal of sucrose by antagonists may be an important factor in preventing germination of *P. expansum* conidia at the wound site as many effective antagonists utilize sucrose rapidly. This may be of particular importance in preventing infections from taking place during fruit handling in packinghouses, where an antagonist can be present at the wound site at the same time or shortly after the wounds are made.

Genomic approach to analyze the defense gene expression during the rice and rice blast interaction. C. JANTASURIYARAT (1), G. Lu (1), B. Zhou (1), E. Mazur (1), H. Kim (2), Y. Yu (2), R. Wing (2), and G. L. Wang (1). (1) Department of Plant Pathology, The Ohio State University, Columbus, OH 43210; (2) Clemson University Genome Institute, 100 Jordan Hall, Clemson, SC 29634. Phytopathology 92:S39. Publication no. P-2002-0280-AMA.

Rice blast, caused by the fungus *Magnaporthe grisea*, is one of the most important diseases in rice production worldwide. Host resistance is the most effective way to control the disease. To understand the molecular basis of the host resistance to rice blast, a genomic approach is being used to profile gene expression at early infection stages. Leaf tissue was harvested from Nipponbare infected with compatible and incompatible isolates and from the partially resistant cultivar IR36 6 and 24 hours after inoculation. Using RNA isolated from these tissues, six libraries are being constructed. Another library is being constructed using RNA isolated from leaf tissue of the lesion mimic mutant *sp111*. About 5,000 clones from each library are being randomly picked and stored in 384-well plates. Two libraries were shipped to the Clemson University Genomic Institute for sequencing. A BLAST search will be conducted to identify what type of genes are involved in early defense response to rice blast.

A phylogenetic study of the Coniophoraceae, a family of wood decay fungi. C. A. JASALAVICH and J. Jellison. Dept. of Biological Sciences, University of Maine, Orono, ME 04669. Phytopathology 92:S39. Publication no. P-2002-0281-AMA.

Coniophoraceae is a family of wood decaying homobasidiomycetes that are primarily brown rot fungi. The family includes a number of important brown rot species capable of causing extensive damage to wood in service, e.g. buildings, railroad ties, telephone poles, and mine timbers, as well as many species known to decay downed timber and slash in the forest ecosystem. Some species may also attack living trees. We are currently examining the phylogenetic relationships among members of this family based on sequence data for the nuclear internal transcribed spacer region and a portion of the mitochondrial small subunit ribosomal DNA.

Induced systemic resistance by PGPR against multiple diseases under field conditions in Thailand. K. JETIYANON (1) and J. W. Kloepper (2). (1) Dept. Agricultural Sciences, Phitanulok, Thailand 6500; (2) Dept. Entomology and Plant Pathology, Auburn University, Auburn, AL 36849. Phytopathology 92:S39. Publication no. P-2002-0282-AMA.

Combinations of plant growth-promoting rhizobacteria (PGPR) (including *Bacillus amyloliquefaciens* strain IN937a and *B. pumilus* strain IN937b; *B. pumilus* strains IN937b and SE34; *B. pumilus* strain IN937b and SE49; *B. pumilus* strain T4 and INR7); and strain IN937a alone were evaluated for capacity to elicit induced systemic resistance (ISR) under field conditions in Thailand. Diseases tested included southern wilt of tomato caused by *Sclerotium rolfsii*; anthracnose of long cayenne pepper (*Capsicum annum* var. *acuminatum*) caused by *Colletotrichum gloeosporioides*; and cucumber mosaic virus (CMV) on cucumber. All PGPR treatments elicited significant protection ($P = 0.05$) against CMV and anthracnose, compared to the control. Significant protection against all three pathogens resulted from application of strain IN937a alone and in mixture with strain IN937b. Additionally, correlative responses in cumulative marketable yields were observed in some PGPR treatments.

Comparison of soybean differential reactions following inoculation with four races of *Phytophthora sojae* from Ohio and Indiana. H. JIA (1), A. E. Dorrance (1), T. L. Richards (2), and T. S. Abney (2). (1) Dept. Plant Pathology, The Ohio State University, Wooster, OH 44691; (2) USDA-ARS, Dept. Botany and Plant Pathology, Purdue University, W. Lafayette, IN 47907. Phytopathology 92:S39. Publication no. P-2002-0283-AMA.

Hypocotyl inoculation is commonly used to determine pathotypes of *P. sojae* and to identify *Rps* genes. Differences in reactions for isolates have been reported. This may be due to changes in virulence in the pathogen or to different sources of resistance in the differential. Isolates of *P. sojae* representing races 1, 3, 4, 7 and 25 from IN and OH were compared on three separate sets of soybean differentials using three separate seed sources. Races 1, 3, 4, 7, and 25 had the expected reaction on all three sets of differentials for *Rps1b*, *Rps1c*, *Rps1k*, *Rps2*, *Rps3a*, *Rps4*, *Rps5*, *Rps7* and differentials Harlon, L59-731, and Union for *Rps1a*. Differentials L88-8470 for *Rps1a* and L93-3302 for *Rps1d* did not have the expected response. Additional races are needed to differentiate reactions on *Rps3b*, *Rps3c*, and *Rps6*. Utilizing different sources of resistance for *Rps* alleles may account for some of the differences in reactions among *P. sojae* isolates.

Resistance of *Sclerotinia homoeocarpa* to thiophanate-methyl and propiconazole in Ohio. Y. JO, J. W. Rimelspach, and M. J. Boehm. Dept. Plant Pathology, The Ohio State University, Columbus, OH 43210. Phytopathology 92:S40. Publication no. P-2002-0284-AMA.

Dollar spot is the most widespread and chronic turfgrass disease on golf courses in Ohio. It is well known that dollar spot fungus (*Sclerotinia homoeocarpa*) develops resistances to benzimidazole and demethylation inhibitor (DMI) fungicides. Dual resistance to both chemical groups is also reported. Samples included 41 isolates from 36 different golf courses and 37 baseline isolates from the university research field in Ohio. ED₅₀ was determined by in vitro fungal growth on fungicide-amended media with thiophanate-methyl (benzimidazole fungicide family) or propiconazole (DMI fungicide family). ED₅₀ of the baseline population was 0.7 ppm of thiophanate-methyl and 0.008 ppm of propiconazole. It was found that 54% of Ohio isolates were resistant to thiophanate-methyl (ED₅₀ > 500 ppm), 29% intermediately resistant to propiconazole (0.05 < ED₅₀ < 0.08 ppm) and 17% strongly resistant to propiconazole (ED₅₀ ≥ 0.08 ppm). The dual resistance was 2.5 times more common than the single resistance. More than half of the isolates were still sensitive to both or either of the fungicides.

Molecular differentiation of *Erwinia amylovora* strains and two Asian pear pathogens. S. Jock and K. GEIDER. MPI für Zellbiologie, Ladenburg, Germany. Phytopathology 92:S40. Publication no. P-2002-0285-AMA.

The fire blight pathogen *Erwinia amylovora* was distinguished by PFGE analysis revealing spread from England to Central and Western Europe as well as from Egypt to Turkey and the Balkans. The ordered pattern types could indicate its rare escape from North America and rare establishment of the disease by trade of plant material. Sequential long distance spread dominates, but the pattern type 3 in Northern Italy and Central Spain was associated with import of contaminated plants. Raspberry and fruit tree strains from North America were diverse in PFGE analysis, similar to *Erwinia pyrifoliae*, detected in Korea, and the related *Erwinia* strains from Japan. The diversity could reflect long pathogen persistence in these countries for accumulation of genomic changes. Nucleotide sequence analysis of genes including *hrpN* revealed a close relationship of the Asian pear pathogens and a distance to *E. amylovora*. The short sequence DNA repeats (SSRs) of *E. amylovora* 'ATTACAGA' varied from 3 to 15 iterations and 'GGATTCTG' of the *Erwinia* strains from Japan from 15 to 25. The SSR numbers of *E. amylovora* strains were unrelated to the origin of isolation and changed under stress conditions.

Allozyme and DNA sequence analysis of *Ceratocystis fimbriata* isolates reveal geographic groupings and host associated lineages. J. A. JOHNSON (1), C. J. Baker (1), T. C. Harrington (1), and J. D. Nason (2). (1) Dept. of Plant Pathology; (2) Dept. of Botany, Iowa State University, Ames, IA 50011. Phytopathology 92:S40. Publication no. P-2002-0286-AMA.

The fungus *Ceratocystis fimbriata* causes wilts and cankers of many tree species and roots of several root crops. Many strains of *C. fimbriata* show evidence of host specialization, but there is little morphological variation among isolates. Phylogenetic analyses of ITS rDNA sequences, and a portion of the *MAT-2* gene placed isolates into three major geographic clades, one centered in Latin America infecting many hosts, one in Asia infecting *Ficus* and *Colocasia*, and a third in North America infecting *Prunus*, *Quercus*, *Populus*, and *Carya*. Allozyme analysis of 12 enzymes distinguished the six host-associated groups in the Asian and North American clades, while there was little variation within the Latin American clade. Allozymes provide further phenotypic markers for delimiting host-specialized groups or species within the *C. fimbriata* complex.

Varietal resistance evaluation for control of Phytophthora blight of pepper. S. A. Johnston, W. L. Kline, M. L. FOGG, and M. D. Zimmerman.

Rutgers University. Phytopathology 92:S40. Publication no. P-2002-0287-AMA.

Phytophthora blight (*Phytophthora capsici*) is the most important disease of peppers in New Jersey. Cultural practices and chemical applications are the primary methods of control utilized in commercial production; nevertheless, high disease incidence prevails under high soil moisture conditions. Effective disease management requires the availability of varietal resistance. Commercial varieties and experimental breeding lines were evaluated during the summer of 2001 in *P. capsici* infested fields on a commercial farm and at the Rutgers Agricultural Research & Extension Center. The majority of infection at both sites was due to the crown rot phase of the disease. 'Paladin', a commercial variety, and RPP 9430 UP (Rogers) resulted in lowest incidence of crown rot compared to the commercial standard 'Camelot'. SVR 2670 390B and SVR 2670 383 (Semini) also experienced low disease incidence at the commercial location and at the extension center, respectively. Conversely, 'Conquest', reported to be blight resistant, consistently experienced high levels of disease at both sites.

The effects of plant architecture in canola on sclerotinia stem rot (*Sclerotinia sclerotiorum*) avoidance. C. J. JURKE (1) and W. G. D. Fernando (2). (1) Advanta Canada Inc. and (2) University of Manitoba. Phytopathology 92:S40. Publication no. P-2002-0288-AMA.

Resistance to sclerotinia stem rot caused by the fungus *Sclerotinia sclerotiorum* (Lib.) de Bary has not been recorded in canola (*Brassica napus* L. and *B. rapa* (L.) Thell. Emend. Metzger), but a range in incidence has been noted in various canola cultivars. Field trials at three locations evaluating eleven morphologically different canola cultivars on the level of sclerotinia infection and on numerous plant architectural components were set up to investigate why there is a consistent difference in infection. Correlations of the architectural components with sclerotinia incidence revealed significant relationships between disease and petal drop and canopy density. Principle component analysis based on 13 architectural components further revealed that there is significant influence of certain plant architecture and resulting canopy structure on the degree of sclerotinia infection.

Epistasis analysis of *dnd1*-mediated resistance responses. G. JURKOWSKI, A. Rettler, and A. Bent. University of Wisconsin-Madison. Phytopathology 92:S40. Publication no. P-2002-0289-AMA.

Mutation of *DND1*, a cyclic nucleotide-gated ion channel of *Arabidopsis thaliana*, leads to heightened levels of disease resistance but elimination of the hypersensitive response in response to pathogens expressing a recognized *avr* gene. These data support a role for ion fluxes in activation of disease resistance pathways. We have used a genetic approach to elucidate the molecular mechanisms that contribute to *dnd1*-mediated resistance. Specifically, we have generated double and triple mutants in the *dnd1* genetic background by introducing *npr1*, a key mediator of both salicylic acid (SA)-dependent and SA-independent resistance pathways, *ndr1*, a regulatory protein involved in LZ-NBS-LRR *R* gene-mediated resistance pathways, and *nahG* (a gene whose expression leads to SA degradation). We observe that *dnd1* activates signaling that is *NPR1*-independent. We find that *NDR1* acts not only in *R/avr*-activated signaling, but also in the broad-spectrum defense signaling activated by *dnd1*. Work is in progress using *ein2* and triple mutant lines. We will present a genetic model for defense signaling that incorporates the above findings.

Crop rotation for Verticillium wilt management in conventional and organic strawberry. Z. KABIR (1), K. V. Subbarao (1), F. N. Martin (1), and S. T. Koike (2). (1) Dept. Plant Pathology, UC DAVIS/USDA, Salinas, CA 93905; (2) UCCE, Salinas, CA 93901. Phytopathology 92:S40. Publication no. P-2002-0290-AMA.

Wilt caused by *Verticillium dahliae* is a major disease in organic and in non-fumigated conventional strawberry production systems in California. The influence of rotations and residue incorporation on wilt, growth and yield of strawberry was studied in the two systems. In both systems, two successive crops of broccoli or lettuce, and residue incorporation were followed by strawberry. After rotation, the numbers of microsclerotia (MS) in broccoli plots decreased 44% and 18% from the initial level of 39-40 MS g⁻¹ soil in conventional and organic systems, respectively, in contrast to increasing 27% in conventional and 31% in organic lettuce plots. In both systems, wilt was less severe when rotated with broccoli than with lettuce. Strawberry canopy diameter and shoot weight were greater in broccoli plots than in lettuce plots while no significant difference in root length. Cumulative strawberry yield was highest in fumigated control, followed by plots rotated with broccoli and lettuce. Rotation with broccoli could thus be used for *Verticillium* wilt management in both production systems.

Assessment with microsatellite markers of diversity of *Sclerotinia sclerotiorum* from potato fields in the Columbia Basin of Washington. Z. KANAAN-ATALLAH, D. A. Johnson, and X. Chen. Dept. Plant Pathology, Washington State Univ., Pullman, WA 99164. Phytopathology 92:S41. Publication no. P-2002-0291-AMA.

Sclerotinia sclerotiorum the causal agent of stem rot is widely distributed on various crops in Washington state and is not satisfactorily managed on potatoes. Because of the ubiquitous nature of the fungus and the size and crop diversity of this region, understanding the population diversity may help in developing control strategies. Isolates of *S. sclerotiorum* were obtained from potato stems in three distant fields in the Columbia basin. Sixty isolates were analyzed for genotypic differences using 25 specifically developed microsatellite primer pairs. Isolates were scored based on banding patterns of their respective alleles. Clustering analyses yielded two main groups with 73 percent similarity of closely related isolates. Genotypes from the two clusters seemed to be equally distributed amongst the locations sampled. These results, from a subset of collected isolates, seem to denote a restricted diversity and indicate a rather limited movement of genotypes from outside the Columbia basin.

A PCR-based assay for specific detection of *Acremonium implicatum*, an endophytic fungus in species of *Brachiaria*. S. KELEMU (1), H. Dongyi (1), H. Guixiu (2), and Y. Takayama (3). (1) CIAT, A. A. 6713, Cali, Colombia; (2) CATAS, Danzhou City, People's Republic of China; (3) 19-1-208, Minamitomigaoka, Nara-shi, Nara 631-0023, Japan. Phytopathology 92:S41. Publication no. P-2002-0292-AMA.

Brachiaria is a pan-tropical genus with about 100 species. Some of the African species are economically important forage grasses in tropical America. An endophytic association of *Acremonium implicatum* with species of *Brachiaria* has been identified. DNA from isolates of *A. implicatum* was amplified with arbitrary 10-mer primers. A 500-bp polymerase chain reaction (PCR) product amplified with primer OPAK10 and common to most of the isolates of *A. implicatum* was cloned, and sequenced. Two specific primers, P1 (5'-TTCGAATGATAAGGCAGATC-3') and P4 (5'-ACGCATCCAC-TGTATGCTAC-3') were synthesized. The primer pair amplifies a single fragment of about 450-bp from DNA of isolates of *A. implicatum* whether from pure culture or in association with *Brachiaria* plants. No amplification product was detected using DNA from endophyte-free plants or from pathogenic and non-pathogenic fungi associated with *Brachiaria*. This assay allows precise and rapid detection of endophytes in *Brachiaria* plants and permits differentiation between endophytic and non-endophytic fungi.

Refinement of DMCase, a predictor of grapevine downy mildew (*Plasmopara viticola*). M. M. KENNELLY (1), R. C. Seem (1), D. M. Gadoury (1), W. F. Wilcox (1), and P. A. Magarey (2). (1) Cornell University Dept. of Plant Pathology; (2) South Australian Research and Development Institute, Loxton 5333 SA. Phytopathology 92:S41. Publication no. P-2002-0293-AMA.

DMCase, a previously-described (Vitic. Enol. Sci. 52:182-189) model of grapevine downy mildew, predicts the occurrence and severity of infection periods based on temperature, relative humidity, surface wetness, and light. We replaced the original primary infection component with simple rainfall-, temperature-, and phenology-based thresholds (rain > 0.25 mm; temperature > 11°C; and phenology > Eichorn and Lorenz stage 12), and we present 19 years of data to validate the modification. The secondary infection component, which deals only with foliar infection, accurately predicts the occurrence of infection, but not the severity of disease following infection events. We found that berries strongly expressed ontogenic resistance approximately 2-3 weeks after bloom, but the rachis remained susceptible several weeks longer. These factors were not previously considered in DMCase. A number of modifications incorporating spore mortality under field conditions, current disease severity, and current host phenology are proposed to more precisely align DMCase predictions with the actual risk of infection.

Characterization of a mobilizable pathogenicity island in *Streptomyces turgidiscabies*. J. A. KERS, K. D. Cameron, and R. Loria. Department of Plant Pathology, Cornell University, Ithaca, NY 14853. Phytopathology 92:S41. Publication no. P-2002-0294-AMA.

Horizontal transfer of an undefined region containing the thaxtomin biosynthetic gene cluster (*txtAB*, *txtC*), an independent virulence gene (*necl*), and other putative pathogenicity genes, has resulted in the evolution of new pathogenic *Streptomyces* species in agricultural systems. We have demonstrated simultaneous transfer of *necl* and the *txtAB*, *txtC* regions, from the pathogen *S. turgidiscabies* to the nonpathogen *S. lividans* via conjugation.

Cosmids have been isolated from a *S. turgidiscabies* cosmid library that hybridize to the transferred region. By pulsed-field gel analysis and Southern hybridization we have determined that the transferred region is at least 400 kb in size and integrates into the *S. lividans* chromosome. Sequence analysis of the transferred region has revealed many cryptic genes and transposases, features typical of pathogenicity islands (PAIs). A number of genes on the PAI have homologs that suggest a role in secretion, environmental sensing and regulation of toxin production. We are currently characterizing the border regions of the PAI.

Systemic resistance induced in cucumber against *Phytophthora crown rot* and blight by *Trichoderma hamatum* 382. J. KHAN, L. V. Madden, and H. A. J. Hoitink. Department of Plant Pathology, The Ohio State University, OARDC, Wooster, OH 44691. Phytopathology 92:S41. Publication no. P-2002-0295-AMA.

T. hamatum 382 (T382) inoculated into a composted cow manure-amended potting mix (compost mix) significantly reduced the severity of *Phytophthora* leaf blight of cucumber compared to plants grown in the same mix not colonized by the biocontrol agent. This control did not differ significantly from that provided by a drench with benzothiadiazole (BTH). In split-root cucumber seedling bioassays, where one half of the root system was grown in the compost mix inoculated with T382, and the paired half in a disease-conducive Sphagnum peat mix infested with *P. capsici*, the severity of crown and root rot was significantly reduced compared to seedlings grown in the same split root system without T382. The effect of T382 in this bioassay on disease severity did not differ significantly from that provided by a drench of BTH or Mefenoxam. In both bioassays, the inducer (T382) remained spatially separated on the plant from the challenging pathogen (*Phytophthora capsici*). We conclude that T382 induced systemic resistance in cucumber against *Phytophthora* root and crown rot as well as leaf blight.

Managing *Cercospora* using the prediction model. M. Khan. North Dakota State University & University of Minnesota. Phytopathology 92:S41. Publication no. P-2002-0296-AMA.

Cercospora leaf spot is the most damaging foliar disease of sugarbeet in North Dakota and Minnesota. This research was conducted to determine the most effective and economical method for controlling *Cercospora* in sugarbeet. Research was conducted in 2001 using susceptible (HH Agate) and tolerant (Crystal 222) sugarbeet varieties at two locations. Fungicide applications were made based on a calendar basis, and the prediction model where daily infection values were calculated at relative humidities greater than 87% and greater than 90%. At Breckenridge, MN, it was not economical to apply fungicides in most of the treatments in the low disease conditions that prevailed. At St. Thomas, ND, disease severity was higher compared to Breckenridge. There was a significantly lower recoverable sugar per acre between the untreated and treated susceptible HH Agate. There was no significant difference in recoverable sugar per acre between the untreated and treated tolerant Crystal 222. It was economical to apply fungicides to the susceptible HH Agate but not always economical for the tolerant Crystal 222. There was no difference between the daily infection values calculated at RH > 87% and at RH > 90%.

Molecular evolution of the *Avr-Pita* gene family in *Magnaporthe grisea*. C. H. KHANG (1), S. Kang (1), and B. Valent (2). (1) Dept. Plant Pathology, The Pennsylvania State University, University Park, PA 16802; (2) Dept. Plant Pathology, Kansas State University, Manhattan, KS 66506. Phytopathology 92:S41. Publication no. P-2002-0297-AMA.

The host specificity of *Magnaporthe grisea* follows the gene-for-gene model. *Avr-Pita*, one of its avirulence genes, prevents *M. grisea* from infecting the rice cultivars that contain the *Pita* resistance gene by directly interacting with the *Pita* gene product. Genomic DNA blots of *M. grisea* isolates from various hosts and geographic locations revealed that *Avr-Pita* is a member of a gene family. At least three distinct members of the gene family were isolated from several *M. grisea* isolates that are not pathogenic on rice. Some members were functional as avirulence genes when introduced into a rice pathogen. Gene genealogies and the comparative study of genome organization of these family members suggest that the gene duplication events mediated by repetitive DNA elements have been involved in the evolution of the *Avr-Pita* gene family.

Development of a biocontrol product based on *Paecilomyces lilacinus* (strain 251). S. Kiewnick (1), P. Lueth (2), and R. A. SIKORA (1). (1) University of Bonn, Dept. of Soil Ecosystem Phytopathology and Nematology, Germany; (2) Prophyta Biologischer Pflanzenschutz GmbH, Malchow, Germany. Phytopathology 92:S41. Publication no. P-2002-0298-AMA.

In the past, commercialisation of biological control products often failed during the last step of development: mass production and marketing. *Paecilomyces lilacinus* (strain 251), is a unique strain with a wide range of activity against the most important plant parasitic nematodes. Although efficacy has been demonstrated for Bananas, Tobacco and Tomatoes, product availability was limited due to insufficient production capacity and a user unfriendly formulation with a high end user price. Based on the ability to rapidly produce high amounts of conidia on solid substrates, a solid state fermentation system was developed which significantly decreased production costs. Furthermore, to guarantee worker and applicator safety, the conidia are formulated as a water dispersible granule (WG) with glucose as carrier. In this formulation, conidia have a sufficient shelf life (at least nine month at 4-6°C).

Effect of various soil treatments on the viability of oospores of *Plasmopara viticola*, downy mildew of grapevine. B. X. KILLIGREW (1), K. Sivasithamparam (1), and E. S. Scott (2). (1) Dept. Soil Science and Plant Nutrition, University of Western Australia; (2) Dept. Applied and Molecular Ecology, University of Adelaide. Phytopathology 92:S42. Publication no. P-2002-0299-AMA.

Downy mildew of grapevines is a damaging disease worldwide, and has recently caused significant yield losses in Western Australia. The pathogen, *Plasmopara viticola*, is a Chromistan. Oospores, which overwinter in decaying leaf matter, are thought to be the main primary inoculum. Oospores in leaf material were placed between two nylon mesh sheets in soil under regimes including composts and herbicide and fungicide applications. The viability of the oospores was examined after several months by extracting oospores from leaf pieces from each regime, aspirating them onto a selective medium and assessing germination. Viability of oospores was related to individual treatment effects.

***Ralstonia solanacearum* Biovar 2, Race 3 in geraniums imported from Guatemala to Pennsylvania in 1999.** S. H. KIM (1), T. N. Olson (1), and N. W. Schaad (2). (1) Plant Disease Diagnostic Laboratory, PA Department of Agriculture; (2) USDA ARS Foreign Disease-Weed Science Research Unit. Phytopathology 92:S42. Publication no. P-2002-0300-AMA.

The PA Department of Agriculture Plant Disease Diagnostic Laboratory received geraniums infected with *Ralstonia solanacearum* Biovar 2, Race 3 (RsB2R3) from three greenhouses in Spring 1999 and two additional greenhouses in Spring 2000. The geranium cultivars originated from propagators in Guatemala or Mexico. The disease was identified by symptoms (wilting) and signs (bacterial oozing) and the organism by isolation on TTC (fluidal pink-centered colonies), ELISA (positive with Agdia antibody, BRA 33900/0500), Biolog database (identified as *R. solanacearum*), and utilization and oxidation of carbon sources (keyed to RsB2R3). The identity was confirmed by real-time PCR using RsB2R3 specific primers and probe and pathogenicity tests on 4-6 leaf stage plants of geraniums (+), tomatoes (+), eggplants (+), potatoes (+), and tobacco (-). The importation of RsB2R3 in geraniums propagated in Guatemala in 1999 constituted the first introduction to the US.

Possible role of a putative nuclear localization signal in *dspE*, a pathogenicity/avirulence gene of *Erwinia amylovora*. W. S. KIM and S. V. Beer. Dept. of Plant Pathology, Cornell Univ., Ithaca, NY 14853. Phytopathology 92:S42. Publication no. P-2002-0301-AMA.

PSORT and PROSITE profile analysis revealed that *dspE* contains a typical bipartite nuclear localization signal (NLS). To determine its role in the disease-specific/avirulence functions of *dspE*, the wild-type gene and three NLS site-directed mutants (SDMs), fused with the β -glucuronidase (GUS) reporter gene, were introduced into onion cells by microprojectile bombardment. All mutations were also marker-exchanged into *E. amylovora* and the resulting mutants were tested for pathogenicity on apple trees growing in the greenhouse. Gus activity was localized to the nuclei of onion cells with the three NLS SDMs as well as intact NLS, whereas a vector control resulted in cytoplasmic localization. The NLS SDMs were not affected in either nucleus localization or pathogenicity. In addition, pathogenicity study with two NLS deletion mutants is underway; these localized to the cytoplasm of onion cells. The wild-type *dspE* gene and the five NLS mutant genes will be transformed into *Pseudomonas syringae* pv. *glycinea*; the resultant cells will be inoculated to soybean and evaluated for avirulence function (HR elicitation).

Isolation and characterization of the second catalytic subunit of cAMP-dependent protein kinase in *Magnaporthe grisea*. Y. KIM, J. E. Hamer, and J.-R. Xu. Dept. Bot. and Plant Pathol. Purdue Univ., West Lafayette, IN 47907. Phytopathology 92:S42. Publication no. P-2002-0302-AMA.

Rice blast caused by *Magnaporthe grisea* is one of the most devastating diseases of rice in the world. The fungus uses strong turgor pressure built up inside appressoria to penetrate rice plants. Because adding exogenous cAMP induces appressorium formation on hydrophilic surfaces, it is likely that surface recognition and appressorium initiation are mediated by the cAMP signaling pathway in *M. grisea*. However, mutants deleted of *CPKA*, a gene encoding a catalytic subunit of cAMP-dependent protein kinase (PKA), still form appressoria and respond to exogenous cAMP on hydrophilic surfaces, indicating that there are multiple catalytic subunits of PKA in *M. grisea*. In this study we isolated the *CPK2* gene that encodes the second catalytic subunit of PKA in *M. grisea*. The *CPK2* gene is 52% identical to *TPK3*, 52% to *ukc1*. Preliminary data indicated that *CPK2* is dispensable for appressorium formation and penetration. Currently, we are characterizing the phenotypes of *cpk2* mutants and generating *cpkA*, *cpk2* double mutants.

Temperature-sensitive reaction to *Stagonospora nodorum* in winter wheat. Y. K. KIM and W. W. Bockus. Dept. Plant Pathology, Kansas State University, Manhattan, KS 66506. Phytopathology 92:S42. Publication no. P-2002-0303-AMA.

Stagonospora nodorum blotch (SNB) is an important foliar and head disease of wheat (*Triticum aestivum*) in many regions of the world. To determine the effect of temperature on resistance to SNB, seedlings of the winter wheat cultivars Newton, AGSECO 7853, and Heyne were inoculated with three isolates of *S. nodorum* and exposed to three temperature regimes [high (21-29 C), medium (17-25 C), and low (10-18 C)]. Heyne was resistant at all temperatures in all three experiments. The reaction of AGSECO 7853 to two of the isolates was intermediate between Heyne and Newton at high temperature, but as susceptible as Newton at medium and low temperatures. Therefore, the reaction of AGSECO 7853 relative to Newton was temperature-sensitive. Although temperature-sensitive reaction of wheat to rusts has been reported, this is the first report of temperature affecting the reaction of wheat to *S. nodorum*.

Characterization of Californian and Australian isolates of *Fusarium oxysporum* f. sp. *vasinfectum*. Y. KIM and R. M. Davis. University of California, Davis. Phytopathology 92:S42. Publication no. P-2002-0304-AMA.

Fusarium wilt, caused by *Fusarium oxysporum* f.sp. *vasinfectum* (FOV), affects cotton production both in CA and in Aust. However, strains in Aust. are reported to be more virulent than CA strains. Because of the importation of cotton seed into CA for cattle feed, there is concern over the introduction of foreign FOV strains into CA. To determine if Aust. isolates differ from those already found in CA, a portion of the gene for the translational elongation factor (EF) was sequenced, using primers EF1 and EF2, in 15 CA isolates and 6 Aust. isolates. A three-base deletion in the Aust. isolates, among other differences, separated the Aust. strains from the CA strains. No differences were found in the internal transcribed spacer (ITS) region sequences between isolates of both regions. Restriction enzyme digestion of amplified intergenic spacer (IGS) region with *SacI* and *RsaI* produced different banding patterns between CA isolates and Aust. isolates. These findings suggest that Aust. isolates are different from prevalent CA strains.

Co-inoculation of genotypes A and B of *Phialophora gregata* does not modify symptom expression of soybean. A. E. KINZIGER and C. R. Grau. University of Wisconsin-Madison. Phytopathology 92:S42. Publication no. P-2002-0305-AMA.

Genotypes A and B of *Phialophora gregata* (Pg) infect the same soybean genotype in the field, however the frequency of concurrent infection of individual plants is low. It is unknown if plant selectivity is due to differences in inoculum density or competition between pathogen genotypes. A susceptible cultivar (Corsoy 79) was inoculated with two A isolates and two B isolates in the greenhouse to determine if genotypes compete for infection of the host. Plants were inoculated using an inoculum density of 10^7 spores/ml at different proportions of (A:B) 1:1, 9:1, 1:9, 1:0 and 0:1. Plants inoculated with a 1:1 ratio had a mean foliar severity of 43% similar to plants inoculated with a proportion of 1:0 46%, and plants inoculated with 9:1 55%. Inoculation of 0:1 produced a mean foliar severity of 3%. Co-inoculation with genotypes A and B did not inhibit phenotypic expression of symptoms when compared to inoculation with either genotype alone. The presence of A and B genotypes will be determined by PCR.

Soybean in Pennsylvania infected by *Sclerotinia sclerotiorum* clones common to legumes and crucifers in New York and Canada. L. Kohn (1) and B. W. PENNYPACKER (2). (1) Dept. of Botany, Univ. of Toronto, Mississauga, ON, Canada L5L 1C6; (2) Dept. Crop and Soil Sciences, Penn State, Univ. Park, PA 16802. Phytopathology 92:S42. Publication no. P-2002-0306-AMA.

S. sclerotiorum sclerotia from soybeans in a field experiment on management of white mold in Lycoming Co. PA were genotyped. 48 plots (0.15 ha) were harvested; each grain sample representing a 5.6 m² area. Sclerotia were found in 20 of 48 grain samples. Seven isolates were selected, via preliminary mycelial compatibility analysis, for DNA fingerprinting and comparison to the Canadian database. One PA-isolate had the fingerprint of Clone 1, which represented 46 percent of 213 soy- and edible bean isolates in Ontario (O) and Quebec (Q) in 2000 and 2001. Clone 1 was first isolated in 1989 from O canola and was also sampled from NY cabbage. Two PA-isolates were Clone 2, common on canola across Canada. Three PA-isolates were Clone 752, found on NY cabbage and O/Q soybean and edible bean. The 7th PA-isolate is a new clone. Soybean in PA, as in O and Q, is infected largely by genotypes already in the region and associated with other crops, such as edible bean, cabbage and canola. The proximity of NY and Eastern Canada to PA may explain the common genotypes.

Mechanistic studies of pheromone analog mating and germination inhibitors from *Ustilago hordei*. P. J. KOSTED and J. E. Sherwood. Dept. Plant Science and Plant Pathology. Phytopathology 92:S43. Publication no. P-2002-0307-AMA.

Studies to determine the mechanism by which analogs derived from *Ustilago hordei* pheromones inhibit mating of *U. hordei* and germination of *Tilletia* spp. continue to yield interesting results. Previous results showed 4mer peptide analogs caused receptor binding competition and competitive inhibition of the processing enzyme farnesyltransferase. To determine if peptide and cysteine analogs were interfering with other steps of processing, methyltransferase, the enzyme responsible for carboxyl methyl esterification of the pheromones, was tested with farnesylated substrates. Radiolabeled S-adenosylmethionine was used as the methyl donor to detect methyl esterified product. To date the farnesylated peptides YSTC and AAAC, cysteine methyl ester and methionine appear to interfere with methylation of the pheromone peptide. Methyl esterification is dependant on the removal of three C-terminal amino acids of the pheromone peptide. It is possible these analogs are interfering with the -AAXing enzyme of this removal process also. Experiments are in progress to determine these interactions.

Oak mortality, crown condition, and decline-associated damage in Minnesota, 1974-1990. K. W. KROMROY (1), J. Juzwik (1,2), and P. D. Castillo (2). (1) Dept. Plant Pathology, University of Minnesota, St. Paul; (2) USDA Forest Service, North Central Research Station, St. Paul, MN 55108. Phytopathology 92:S43. Publication no. P-2002-0308-AMA.

Oak *Quercus* species occur on over five million hectares of land in seven states of the Upper Midwest USA. Although several reports on oak mortality in the region have been published, information on historical trends of oak tree health is lacking. In a preliminary effort for a regional study, three variables were examined as measures of oak health in Minnesota between 1974 and 1990. The variables (mortality, live crown ratio, damage) were created from USDA Forest Service tree and plot data from two inventory cycles (ending years: 1977 and 1990). Mortality was less than one percent in 1977 and greater than six percent in 1990 on plots with 25 percent or more oaks. For trees examined between May and September, 33 and 29 percent were in poor condition, having live crown ratios of 30 percent or less in 1977 and 1990 cycles, respectively. Damage was recorded for 52 percent of the oaks measured during this same monthly period for the 1990 cycle. Damage associated with decline occurred more frequently than other types of damage within each of three forest types.

The effect of crop sequence on root-rot severity and yield in irrigated dry bean. J. E. KURLE, C. Estevez de Jensen, and J. E. Percich. University of Minnesota. Phytopathology 92:S43. Publication no. P-2002-0309-AMA.

Average yields of dry bean (DB) grown under irrigation in Central Minnesota have declined from 2,464 kg/ha in 1990 to 1,456 kg/ha in 2000. Root rots, caused by *Fusarium solani* f. sp. *phaseoli*, *Rhizoctonia solani*, and *F. oxysporum*, are largely responsible for the yield decline. Soybean or potato grown in rotation with DB are alternative hosts for the pathogens and maintain disease inoculum. A three-year crop rotation study examined the effect of seven alternative crops (AC): alfalfa, barley, canola, corn, potato, rye, soybean, or wheat, on RRS and DB yield. Crops were planted in four sequences: AC/AC/DB, AC/DB/DB, DB/AC/DB, and DB/DB/DB. Specific AC affected RRS (P = 0.063). The greatest RRS (1=healthy to 9=dead) in DB was present following canola and the lowest following alfalfa (5.7 vs. 4.9). Yields of DB following alfalfa were 50% greater than yields of DB following DB (762 vs. 493 kg/ha; P = 0.27).

Effect of soil pH on symptom development and pathogen reproduction of *Phialophora gregata* in soybean. N. C. KURTZWEIL, A. E. Kinziger, and C. R. Grau. University of Wisconsin, Department of Plant Pathology, Madison, WI 53706. Phytopathology 92:S43. Publication no. P-2002-0310-AMA.

Phialophora gregata (*Pg*) is the cause of brown stem rot (BSR) of soybean. Previous studies showed BSR resistant cultivars have a yield advantage of 19 bu/a over BSR susceptible cultivars when grown in soil pH of 6.0, however when soil pH approaches 7.0, the advantage drops to 4 bu/a. A study was conducted to determine the effect of soil pH on BSR symptom severity and pathogen reproduction at a site with soil pH naturally ranging from 5.8 to 8.4. Severity of BSR internal stem browning averaged 13% for BSR resistant cultivars grown at low soil pH (5.8-6.4), and 2% at high soil pH (7.0-8.4). Foliar symptoms for BSR resistant cultivars were 4% and 1% in low and high soil pH, respectively. BSR susceptible cultivars had an average of 86% stem browning at low soil pH and 28% at high soil pH, while foliar severity was 92% at low soil pH to 34% at high soil pH. Population density of *Pg* was estimated in stems and roots using a dilution-plating assay. The highest colony forming units were associated with low soil pH, however *Pg* was detected at high soil pH in the absence of foliar or stem symptoms.

Transformation of sugar beet with a Cercosporin export gene, *cfp*. L. D. KUYKENDALL (1), T. M. Stockett (1), and J. W. Saunders (2). (1) Molecular Plant Pathology Lab, ARS, USDA, Beltsville, MD 20705; (2) Crop and Soil Sciences Dept., MSU, East Lansing, MI 48823. Phytopathology 92:S43. Publication no. P-2002-0311-AMA.

Cercospora leafspot disease of *Beta vulgaris* L. causes reduced tonnage, reduced sucrose content, and losses of up to 30% in recoverable sucrose with only moderate disease severity. R.G. Upchurch at NCSU discovered a light-induced cercosporin toxin export gene (*cfp*). The insertion of this gene into the sugar beet genome can be expected to produce plants with increased resistance to *Cercospora*. This biotechnological approach is important since germplasm screening and breeding has thus far yielded only moderate resistance which is multigenic. Hundreds of cotyledons from surface-sterilized sugar beet seeds were inoculated with *Rhizobium* (*aka Agrobacterium*) *radiobacter* EHA105 harboring a pBIN19 vector carrying *cfp*. The sugar beet seeds used were either biotechnology clone Re11, or C69 breeding line germplasm of Bob Lewellen, USDA, ARS, Salinas, CA. Light (weak), temperature (23.5C), and media conditions (1mg/ml BAP) used were those that we recently discovered as producing regeneration via direct adventitious shoots without a hormone-independent callus intermediate. Most putative transgenics selected on kanamycin had viability problems. Three distinct, PCR-verified transgenic plants possessing *cfp* are now being propagated to maturity for both disease resistance testing and for examination of the synthesis of the CFP toxin export protein using specific antibodies. Additional transgenic clones are also being produced.

The mechanism of forcible ascospore discharge in *Gibberella zeae*. C. KWON and F. Trail. Michigan State University. Phytopathology 92:S43. Publication no. P-2002-0312-AMA.

Little is known of the mechanism of spore discharge in ascomycetous fungi. *Gibberella zeae* (anamorph *Fusarium graminearum*), the causal agent of Fusarium Head Blight on wheat and barley, is being used as a model organism for investigations into the genetics and physiology of ascospore discharge. Analysis of the ascus fluid in *G. zeae* has revealed the presence of mannitol as the predominant sugar component in the eiplasm, implying a possible role for mannitol in the generation of turgor pressure within the ascus. Specific inhibitor studies have also implicated potassium ions in the spore discharge process. We hypothesize that mannitol and potassium ions contribute to rapid buildup of turgor pressure within the ascus resulting in forcible ascospore discharge. Data supporting this hypothesis will be presented.

Use of aggregation pheromones of sap beetles to study overland transmission of *Ceratocystis fagacearum*. J. F. Kyhl (1), J. JUZWIK (2), R. J. Bartelt (3), and S. J. Seybold (1). (1) Dept. Entomology, Univ. of Minnesota, St. Paul, MN 55108; (2) USDA Forest Service, 1561 Lindig St., St. Paul, MN 55108; (3) USDA-ARS, Peoria, IL 61604. Phytopathology 92:S43. Publication no. P-2002-0313-AMA.

Two sap beetles, *Coleopterus truncatus* (Cot) and *Carpophilus sayi* (Cas) (Coleoptera: Nitidulidae), are considered the principal insect species responsible for overland spread of the oak wilt pathogen, *Ceratocystis fagacearum* (Cf), in Minnesota. Flight traps baited with Cot and Cas aggregation pheromones were used to study seasonal flight activity of vectors in oak stands and the incidence and levels of Cf on the vectors. During a weekly

survey over two growing seasons (2000, 2001), greater than 98 percent of the Cot obtained in Cot pheromone baited traps were caught between 14 April and 1 June. Of the 243 Cot collected during this period in 2001, 14 percent yielded Cf based on fungal bioassays. The mean Cf popagule load per beetle on the Cf positive Cot was 9.4×10^3 . Similar studies were initiated in June 2001 using Cas baited traps; greater than 80 percent of Cas obtained during 2001 were caught between 8 June and 27 July.

Moisture sources in relation to conidial dispersal and infection by *Cladosporium carpophilum* within peach canopies. Z. LAN and H. Scherm. Department of Plant Pathology, University of Georgia, Athens, GA 30602. Phytopathology 92:S44. Publication no. P-2002-0314-AMA.

Peach scab, caused by *Cladosporium carpophilum*, is favored by rainfall, but it is unknown whether rain acts by effecting dispersal of conidia, by providing fruit surface wetness (FSW) for infection, or both. The relative importance of FSW sources including splash, twig runoff, and dew and their interaction with air- and waterborne conidia was studied in a peach orchard with blocks that were sprayed or unsprayed with fungicide in the previous season. The localized absence of scab twig lesions in the sprayed blocks implied that fruit could only be infected by airborne conidia, while both air- and waterborne conidia could contribute to infection in the unsprayed blocks. Individual fruit were left untreated, protected from splashing by rain shields, and/or protected from runoff by cotton wicks placed proximal to the peduncle. Rain shields were adjustable, allowing rain and/or dew to be excluded selectively. Results showed that airborne spores contributed little to fruit scab development, even in the presence of FSW caused by dew, runoff, and/or splashing. Splashing was considerably more important than twig runoff in effecting conidial dispersal.

Heat treatment as a virus diagnostic tool. L. C. Lane. University of Nebraska, Lincoln, NE 68583-0722. Phytopathology 92:S44. Publication no. P-2002-0315-AMA.

James Johnson in 1932 proposed that thermal inactivation point was a useful virus property. This was an important insight; but in practice TIP has proved laborious and imprecise. I have combined heat treatments (heating blocks at roughly 5°C intervals) with partial virus purification, analyzing the product (soluble macromolecules that sediment after heating) by SDS gel electrophoresis. Host proteins (especially rubisco) serve as internal standards. The technique is quick (about a day), reproducible, and semi-quantitative. One can separate viruses or study effects of variables, such as pH, on virus stability. Heat eliminates less stable viruses from mixtures (e.g. heating a mixture of STMV and TMGMV facilitates culturing of pure TMGMV).

Cost benefit of spray programs for managing foliar pathogens of onions. D. B. LANGSTON, JR. (1) and G. E. Boyhan (2). (1) Dept. Plant Pathology, University of Georgia, Tifton, GA 31793; (2) Dept. Horticulture, University of Georgia, Statesboro, GA 30460. Phytopathology 92:S44. Publication no. P-2002-0316-AMA.

Fungicide spray programs were evaluated in 2000-01 at the Vidalia Onion and Vegetable Research Center, located in Toombs Co., GA, for suppression of Botrytis leaf blight (*Botrytis squamosa*), purple blotch (*Alternaria porri*), and Stemphylium leaf blight (*Stemphylium vesicarium*). Azoxystrobin (AX), cyprodinil plus fludioxonil (C plus P), iprodione (I), and vinclozolin (2000 only) were applied late season and evaluated for cost effectiveness as compared to full season applications of chlorothalonil (C). Disease suppression was greatest in both years in plots treated with C plus P, AX, and I. All fungicide treatments significantly improved yield over the non-treated plots in 2000 while no significant yield improvements were noted with any fungicide program in 2001. Fungicide returns for fungicide programs in 2000 ranged from \$3,891.00 with full season C to \$5,068.00 with late season applications of C plus P. Returns in 2001 ranged from negative \$228.63 with late season alternations of AX and C plus P to \$273.50 with late season applications of I.

Production and liberation of secondary conidia by *Cercospora zeae-maydis*. C. L. LAPAIRE (1) and L. D. Dunkle (2). (1) Dept of Botany & Plant Pathology; (2) USDA-ARS, Purdue University, W. Lafayette, IN 47907. Phytopathology 92:S44. Publication no. P-2002-0317-AMA.

On a variety of nutrient-deficient substrates, conidia of the maize gray leaf spot pathogen, *Cercospora zeae-maydis*, germinate and develop secondary conidia (SC) on conidiophores produced from germ tubes or directly from conidial cells. A population of conidia increases its numbers more than 2-fold by 2 days on a water droplet and by 4-fold when primary conidia are attached to trichomes of monocot or dicot leaves. Upon transfer from high

humidity to a dry atmosphere, SC and conidiophores gradually dehydrate and collapse. Dehydrated SC can be liberated from the conidiophores by wind speeds about one-third (1.3 m/s) those required to liberate hydrated conidia (3.7 m/s). The dispersed SC are able to rehydrate and germinate normally on water agar. Because this microconidiation cycle (MC) occurs at the expense of endogenous reserves, the ability to produce SC is lost after 4 cycles without an intervening period of growth on nutrient media. The MC process may have epidemiological consequences by maintaining inoculum potential during periods of fluctuating relative humidity when primary conidia fail to establish successful infections.

Effects of different 3-yr cropping systems on soil microbial communities and soilborne disease of potato. R. P. Larkin. USDA, ARS, New England Plant, Soil, and Water Lab. Phytopathology 92:S44. Publication no. P-2002-0318-AMA.

Eight different crop rotations, consisting of soybean-canola, soybean-barley, sweet corn-canola, sweet corn-soybean, green bean-sweet corn, canola-sweet corn, barley-clover, and consecutive potato (nonrotation control) followed by potato as the third crop in all systems, were established in replicated field plots with two rotation entry points in Presque Isle, ME, 1998. The effects on soil microbial community characteristics and the development of soilborne diseases of potato were evaluated in 2000 and 2001. Incidence and severity of stem and stolon canker and black scurf of potato, caused by *Rhizoctonia solani*, were reduced for most rotations relative to the consecutive potato control. Potato crops following canola, barley, or sweet corn provided the lowest disease levels and best tuber quality. Soil bacterial populations and activity also were highest following barley, canola, and sweet corn crops. Other characteristics of the soil microbial communities, including substrate utilization profiles (Biolog plates) and fatty acid methyl ester (FAME) profiles demonstrated distinct differences among cropping systems.

A virus disease epidemic of snap bean occurring in the Great Lakes region of the United States. R. C. LARSEN (1), C. R. Grau (2), K. C. Eastwell (3), A. M. Mondjana (2), P. N. Miklas (1), and W. R. Stevenson (2). (1) USDA-ARS, Prosser, WA; (2) Dept. Plant Pathology, University of Wisconsin, Madison, WI; (3) Washington State University, Pullman, WA. Phytopathology 92:S44. Publication no. P-2002-0319-AMA.

A virus complex causing an array of leaf and pod symptoms on snap bean (*Phaseolus vulgaris* L.) became epidemic during 2001 in the Great Lakes region of the U.S. and Ontario, Canada. The sudden increase in virus incidence in this region coincides with a population explosion of the soybean aphid (*Aphis glycines*). The virus complex resulted in severe yield losses due to reduced pod number, pod twisting, pod necrosis, and decline of general plant health. Symptomatic snap bean samples from affected areas were evaluated for a selected panel of viruses either by ELISA, or PCR using virus-specific primers. The most frequently occurring viruses in samples from Wisconsin, the largest production area, were cucumber mosaic virus (90%) and alfalfa mosaic virus (82%). Other viruses identified were tobacco streak, white clover mosaic, clover yellow mosaic, and clover yellow vein virus. Emergency management strategies are being evaluated for the 2002-growing season.

Integrating fungicides and a *Bacillus mycoides* biological control agent to manage *Cercospora* leaf spot resistance to fungicides. B. J. LARSON and B. J. Jacobsen. Dept. Plant Science and Plant Pathology, Montana State University, Bozeman, MT 59717. Phytopathology 92:S44. Publication no. P-2002-0320-AMA.

Cercospora leaf spot, caused by the fungus *Cercospora beticola*, is a major foliar disease of sugarbeets in Montana. Losses in Montana over the last 5 years have averaged between 2 and 3 tons per acre and between 0.5 and 1.5 percent lower sugar. Current management strategies rely heavily on a handful of registered fungicides. FQPA review and the development of resistance to these fungicides may limit the availability of some of these fungicides. Studies of spore germination and mycelial growth inhibition over the last 3 years have indicated an increase in the level of resistance/tolerance of *C. beticola* to the benzimidazole, strobilurin, and triazole classes of fungicide. Field treatments of the *Bacillus mycoides* biological control agent, Bac J, alone and in combination or rotation with fungicides have performed well as a control of *Cercospora* leaf spot. *C. beticola* isolates taken from plots with these treatments have shown much lower resistance levels than isolates taken from other treatment plots. Bac J is an inducer of systemic resistance in sugarbeets.

A bioassay for photosensitizing perylenequinone cercosporin of *Cercospora beticola*. R. T. LARTEY and T. Caesar-TonThat. USDA-ARS, Northern Plains Agricultural Research Lab, Sidney, MT. Phytopathology 92:S45. Publication no. P-2002-0321-AMA.

Cercosporin is a broad-spectrum photosensitizing perylenequinone toxin that is produced by several members of the genus *Cercospora*. In addition to symptom development, cercosporin has been reported to be toxic to several organisms including mice, bacteria and fungi. This broad-spectrum toxicity may suggest a role in protection against potential antagonists. To explore its possible role on potential antagonists we have developed and present here a quantitative bioassay for cercosporin toxicity. Cercosporin extracts from *Cercospora beticola*, spread on potato dextrose agar (PDA) supported growth of streak-inoculated *E. coli* in darkness but not in light. Dark-incubated cultures were transferred to light and assayed for light-exposure effects on bacteria death at various times using fluorescence microscopy and automated image analysis. We employed simultaneous staining with two fluorescence probes to differentiate dead and living cells. Image analysis showed increased death of *E. coli* cells with increased exposure to light in the presence of cercosporin. The bioassay allows rapid determination of the degree of cercosporin toxicity on living cells.

A protocol for rapid detection of *Cercospora beticola* in infected sugar beet tissues. R. T. LARTEY and S. Bucklin-Comiskey. USDA-ARS, Northern Plains Agricultural Research Lab. Phytopathology 92:S45. Publication no. P-2002-0322-AMA.

Leaf spot by *Cercospora beticola* Sacc. is the most important foliar disease of sugar beet (*Beta vulgaris* L.). The pathogen overwinters as stromata in beet leaf residues. Under optimal conditions, overwintering propagules germinate and produce conidia that are dispersed as primary inoculum to initiate infection in sugar beet. Severe leaf spot occurs even under rotation, suggesting that there are other sources of inoculum such as secondary hosts or weeds. We present an Extract-N-Amp Plant PCR Kit (Sigma)-based protocol for rapid detection and identification of *C. beticola* in plant tissues. Leaf disks from diseased tissues were homogenized in dilution solution. Without DNA extraction, aliquot of the homogenates were added to PCR reactions and subjected to amplification using the *Cercospora actin* gene and ITS region based primers. The fragment sizes of amplified products correlated with the expected size of the control DNA extracts from *C. beticola* cultures. Alignment of sequences of the amplified products confirmed them to be that of *C. beticola*. The system will enable rapid screening for alternate hosts, including asymptomatic plants.

Induction of systemic defense potentiation in soybean. C. T. LARUE and T. L. Graham. Dept. Plant Pathology, Ohio State University, Columbus, OH 43210. Phytopathology 92:S45. Publication no. P-2002-0323-AMA.

Our lab has investigated two types of elicitor-induced resistance in soybeans. Distal defense potentiation, effective throughout an elicitor-treated organ, is triggered by glucan elicitors from *Phytophthora sojae* (*Ps*), and jasmonic acid (JA), but not by salicylic acid (SA). Here we report preliminary characterization of a truly systemic defense potentiation phenomenon in soybean. Soybeans treated with the herbicide lactofen in the field are more resistant to certain pathogens, particularly *Sclerotinia sclerotiorum*. We found that lactofen treatment of unifoliolate leaves of greenhouse plants leads, within 8 days, to upward systemic protection against leaf infection by *Pseudomonas syringae* pv. *glycinea* and downward systemic protection against hypocotyl infection by *Ps*. Removal of the treated unifoliolate at various times showed that defense-generating signals leave the treated leaf within 3 days. Lactofen required for these responses (120 μ M) is only one tenth that applied in the field. Preliminary results suggest JA and glucan elicitors from *Ps* may also induce protection, while SA is inactive. Comparisons to the distal defense potentiation response will be discussed.

interaction using functional genomics. C. B. LAWRENCE, R. A. Cramer, K. Morey, and J. Wang. Department of Bioagricultural Sciences & Pest Management, Plant-Fungal Genomics Laboratory, Colorado State University. Phytopathology 92:S45. Publication no. P-2002-0324-AMA.

Our laboratory is taking a multi-faceted, functional genomics-based approach to dissect the role of both pathogen and host genes in development of diseases caused by necrotrophic fungi. We have chosen the *Alternaria brassicicola* - *Arabidopsis thaliana* interaction as a model system. We are using a combination of genetic and genomic approaches to study this interaction. Methods include screening plant T-DNA mutants for increased resistance / susceptibility, map-based cloning of resistance-conferring genes, and characterization of defense gene expression in a variety of ecotypes and mutants during fungal infection. We have screened a multitude of host

ecotypes for differences in susceptibility and found dramatic differences. Screening of various F2 populations derived from crosses between resistant and susceptible parents indicates that resistance may be due to the presence of a single gene inherited in a dominant manner. Using a recombinant inbred line population we are in process of a map-based cloning approach to isolate a resistance conferring gene. Northern and microarray analyses of defense gene expression patterns indicate that the jasmonic acid (JA) -mediated defense pathway is being predominantly activated in resistant ecotypes while the salicylic acid (SA)-mediated pathway is being activated in susceptible ones. Results of these experimental endeavors will be presented.

Effects of strawberry plant extracts on conidiation and appressorial production by *Colletotrichum acutatum*. L. F. S. LEANDRO, M. L. Gleason, S. N. Wegulo, and F. W. Nutter, Jr. Dept. Plant Pathology, Iowa State University, Ames, IA 50011. Phytopathology 92:S45. Publication no. P-2002-0325-AMA.

Conidial suspensions of *Colletotrichum acutatum* were prepared in 1:27, 1:40 and 1:81 (w/v) dilutions of flowers and leaves from strawberry plants (cv. Tristar) in water. Leaves on intact strawberry plants and plastic cover slips were spray-inoculated with the suspensions, and incubated at 25°C and 100% relative humidity for 24 and 48 h. Conidial and appressorial populations were quantified. At both sampling times, all flower dilutions significantly ($P < 0.05$) increased the number of conidia on leaves and cover slips compared to a water control. Up to 10-fold increases in ungerminated conidia were observed. Leaf extracts increased conidial populations at the 1:27 and 1:40 dilutions only. Appressorial production did not differ among treatments. These results suggest that inoculum levels of *C. acutatum* on foliage may increase sharply during flowering of strawberry plants. The effects of plant extracts on *C. acutatum* populations maintained on leaves and cover slips under dry conditions for up to 6 weeks are also being investigated.

Recent occurrence trends of RSV and its detection using RT-PCR. B. C. LEE (1), D. Y. Kwak (1), Y. K. Hong (1), D. C. Lee (1), S. C. Kim (1), and K. W. Lee (2). (1) National Yeongnam Agricultural Experiment Station; (2) Kyungpook National University. Phytopathology 92:S45. Publication no. P-2002-0326-AMA.

Until now, occurrence of RSV is limited in southern part of Korea. However recently the occurrence of RSV is increasing and spreading in central part of Korea including chungcheong and kyonggi province. It is very difficult to distinguish RSV symptoms on virus symptom physiological damage of rice. The symptoms induced of infected plants includes general leaf striping, yellowing, a distinct white coloring of the leaf stripe. We detected RSV viral RNA from infected rice plant and its insect vector *Laodelphax striatellus* using reverse transcription(RT)-PCR. The result of RT-PCR, we observed specific band including RSV-polymerase(1,023bp) and CP(969bp) in both host of rice plant and insect vector. Furthermore, the cDNAs of CP was cloned and sequenced. It was 969bp long and coded for a protein composed of 322 amino acids.

Classification of phytoplasmas in the expanded elm yellows group (16SrV) based on 16S rRNA and ribosomal protein gene sequences. I. M. LEE (1), M. Martini (1), and C. Marcone (2). (1) USDA-ARS Molecular Plant Pathology Laboratory, Beltsville, MD 20705; (2) Dept. Biologia, Difesa e Biotecnologie Agro-Forestali, University of Basilicata, 85100 Potenza, Italy. Phytopathology 92:S45. Publication no. P-2002-0327-AMA.

Elm yellows (EY) group (16SrV) phytoplasmas are associated with several devastating diseases in elm, grapevine, blackberry, cherry, peach and several other plant species in America, Europe and Asia. The EY group now represents the third most diverse phytoplasma cluster, next to aster yellows (group 16SrI) and X-disease (group 16SrIII) phytoplasma groups. EY group phytoplasmas were categorized on the basis of 16S rRNA sequences. Subgroups were differentiated based on RFLP analyses of 16S rRNA and ribosomal protein (rp) sequences. Five subgroups were identified by analysis of 16S rRNA gene, while 12 subgroups were resolved by analysis of rp genes. Based on phylogenetic analyses of rp genes or 16SrRNA and rp genes combined, the EY group clearly consists of seven genetically distinct strain clusters. Each cluster appeared to evolve as a result of ecological constraints (e.g. specific vector or plant hosts).

Agronomic performance of soybeans (*Glycine max*) in the presence of Soybean mosaic virus and the herbicides glyphosate and imazamox. M. E. LEE, N. C. Kurtzweil, C. Boerboom, J. Gaska, and C. R. Grau. University of Wisconsin-Madison, 53706. Phytopathology 92:S45. Publication no. P-2002-0328-AMA.

Management practices such as row width and herbicides can have dramatic effects on pathogens and disease. Field and greenhouse studies were conducted to determine if interactions occur between two herbicides, glyphosate (Roundup Ultra®) and imazamox (Raptor®), and *Soybean mosaic virus* (SMV). Variety AG2101 was used in all experiments. Soybeans were inoculated with SMV and/or sprayed with water, glyphosate, or imazamox at growth stages V2 and V4. The V2 growth stage is designed to mimic seed transmission of SMV, and V4 aphid transmission. It was found that SMV and imazamox delayed maturity in the field and the greenhouse. In the greenhouse, imazamox at V4 decreased seed transmission ($P \leq 0.0001$). Also in the greenhouse, co-application of SMV and imazamox on soybeans decreased the number of plants infected with SMV. The titer of SMV in soybeans inoculated at V2 significantly increased 24 hours after treatment with imazamox at V4 ($P \leq 0.0001$). Further studies are planned to confirm these results and explore the use of imazamox to reduce the incidence and agronomic effects of SMV in the field.

Impact of soybean canopy on aphid immigration and *Soybean mosaic virus* incidence. M. E. LEE, N. C. Kurtzweil, C. Boerboom, J. Gaska, and C. R. Grau. University of Wisconsin-Madison, 53706. *Phytopathology* 92:S46. Publication no. P-2002-0329-AMA.

The soybean aphid, *Aphis glycines*, causes yield loss and can transmit viruses such as *Soybean mosaic virus* (SMV). Field experiments at Madison, WI determined the impact of insecticides (Warrior® at V3, R1, and Lorsban® at R4) and herbicides (Raptor® and Roundup Ultra® at V3) on the immigration of *A. glycines* and transmission of SMV. A split plot design was used with insecticide as the main plot and herbicide as the sub-plot. Soybean cv. AG2101 was planted 30 May, 2001. Aphids were collected daily from horizontal green mosaic pan traps. The major peak in aphid flight occurred between 30 July and 10 August. Incidence of SMV was 0.3 – 2.4% (V3 – R2), but after the flight began, incidence increased to 80% (R4). Incidence was 100% by R5. Landing rates were greater in insecticide treated plots ($P \leq 0.07$), which were taller and had higher leaf area index. Insecticide did not increase yield ($P \leq 0.95$) or decrease seed mottling ($P \leq 0.97$). Herbicides did not have an effect on canopy during the peak aphid flights, and no significant differences in aphid landing rate was found ($P \leq 0.99$). Raptor® did significantly decrease yield ($P \leq 0.01$) and increased seed mottling ($P \leq 0.001$).

Association of candidate defense response and resistance genes with quantitative blast resistance loci in rice. S. W. LEE (1,2), S. S. Han (2), C. Y. Soon (2), S. H. Choi (2), C. H. Kim (2), and J. E. Leach (1). (1) Plant Pathology, Kansas State University, Manhattan, KS 66506; (2) Plant Pathology, NIAST, Suwon, Korea. *Phytopathology* 92:S46. Publication no. P-2002-0330-AMA.

A candidate gene approach was applied to a population of 164 recombinant inbred (RI) lines derived from a cross between a *japonica/indica* hybrid derivative (Milyang 23) and a *japonica* variety (Gihobyeo) to determine association between defense response genes and resistance gene analogs (RGA) with quantitative trait loci (QTL) for rice blast resistance. Of 157 genes tested with five restriction enzyme digests, 96 were polymorphic. RGA (29) and defense response genes (2) from rice, barley and maize were mapped on the rice chromosomes and analyzed for their association with blast QTL. All markers produced single loci and were well distributed among all the chromosomes except on 12, where no markers were observed. Based on diseased leaf area, and lesion density, size and number using Korean and Philippine blast fungal isolates, a total of nine putative QTL were identified on chromosomes 1, 2, 4, 6, 7, 10 and 12. Five RGA markers were associated with three different QTL.

The amino-terminal encoding portion of CaMV Gene VI controls resistance-breakage in *Arabidopsis thaliana* ecotype Tsu-0. S. LEISNER (1), Y. Li (1), M. Hapiak (1), J. Schoelz (2), and K. Agama (1). (1) Dept. of Biological Sciences, University of Toledo, Toledo, OH 43606; (2) Plant Science Unit, College of Agriculture, Food and Natural Resources, University of Missouri-Columbia, Columbia, MO 65211. *Phytopathology* 92:S46. Publication no. P-2002-0331-AMA.

Arabidopsis thaliana ecotype Tsu-0 is resistant to *Cauliflower mosaic virus* (CaMV) isolate CM1841 but susceptible to the W260 isolate. Analysis of viral chimeras constructed from CM1841 and W260 indicated that gene VI was responsible for resistance-breakage. Furthermore, the critical region of gene VI was localized to the portion encoding the amino-terminal one hundred and ten amino acids of the gene (P6). Resistance is overcome by W260 via a passive mechanism i.e., the virus is not recognized by Tsu-0 host defenses. To test a possible mechanism by which resistance could be overcome, self-association of P6 was examined by yeast two-hybrid analysis.

P6 was found to specifically self-associate. Further analysis of self-association identified several domains that bind to full-length P6, one of which was the resistance-breaking region.

Pathogens of *Prosopis* species in Mexico and Texas. C. L. LENNOX and M. Serdani. ARC-PPRI, Weeds Pathology Unit, P. Bag X5017, Stellenbosch 7599, South Africa. *Phytopathology* 92:S46. Publication no. P-2002-0332-AMA.

Prosopis glandulosa var. *torreyana* and *P. velutina*, native to North and South America, are declared invaders in South Africa. In September/October 2001, a survey was made of pathogens on *prosopis* in Mexico and Texas. Pod anthracnose, caused by an as yet unidentified fungus and characterized by black/grey acervuli, flattening of the pods and seed decay, was found in both Mexico and Texas. Gall rust (*Ravenelia arizonica*) was recorded on young and mature trees in both countries. Leaflet rust (*Ravenelia holwayi*) was regularly found in Texas. Signs of this rust are black telia and an associated leaflet drop. *Diplodia*, *Phoma* and *Sphaeropsis* species were predominantly isolated from material collected in both countries. In November 2001, a decision was taken by a national workshop on the status and long-term management of *prosopis* in South Africa to limit biological control agents to those targeting flowers, pods and seeds. Based on this, and preliminary tests, the potential of the pod anthracnose causal organism as a biological control agent of *prosopis* will be further investigated.

Integrated strategy to control blue mold and bitter rot of 'Golden Delicious' apples. B. LEVERENTZ (1), W. J. Janisiewicz (2), W. S. Conway (1), R. A. Saftner (1), and A. N. Reed (3). (1) USDA-ARS, PQSL, Beltsville, MD 20705; (2) USDA-ARS, AFRS, Kearneysville, WV 25430; (3) Penn State Univ., Fruit Res. & Extension Center, Biglerville, PA 17307. *Phytopathology* 92:S46. Publication no. P-2002-0333-AMA.

'Golden Delicious' apples were treated with heat (38C) for 4 days, MCP, and/or a heat tolerant yeast biocontrol. The treatment was applied at 0 or 12 h after wound inoculation with either *Penicillium expansum* (blue mold) or with *Colletotrichum acutatum* (bitter rot). After treatment, the apples were moved to controlled atmosphere storage for up to four months. Following storage the apples were left at room temperature for 2 weeks. The yeast populations were stable throughout the experiment. Heat alone, or the antagonist plus heat treatment without MCP effectively reduced *P. expansum*. The antagonist alone, or in combination with heat and without MCP effectively reduced *C. acutatum*. This was true for the lesion diameter and the lesion incidence caused by either fungus. The highest lesion incidence occurred on the control treated with or without MCP. In general, MCP treated apples had a higher lesion incidence, and slightly larger lesion diameters compared to non-MCP treated apples.

A new soil assay for *Synchytrium endobioticum*, the causal agent of potato wart. C. A. LEVESQUE (1), L. J. Ward (2), S. N. de Jong (1), S. H. De Boer (2), P. H. J. F. van den Boogert (3), and R. P. Baayen (4). (1) Agric & Agri-Food Canada, ECORC, 960 Carling Ave, Ottawa ON K1A 0C6; (2) Canadian Food Inspect Agency, 93 Mt Edward Rd, Charlottetown PE C1A 5T1; (3) Plant Res Intl, PO Box 16; (4) Plant Prot Service, PO Box 9102, 6700 HC Wageningen, Netherlands. *Phytopathology* 92:S46. Publication no. P-2002-0334-AMA.

No prior molecular data were available for the obligate parasites, *Synchytrium* spp. By developing chytrid specific primers from 18S Genbank data and using them with other universal rDNA primers, sequences of *Synchytrium* spp were obtained for the complete ITS region and most of the 18S gene. Phylogenetic analyses of *Synchytrium* spp and related genera were performed. Sequences obtained from symptomatic tubers, infested soil and herbarium specimens of *S. endobioticum* were identical. The ITS sequence of *S. endobioticum* was distinct, differing from other *Synchytrium* spp, including the ubiquitous *S. aureum*. PCR primers and DNA probes were developed and have been used successfully in Canada and the Netherlands for the sensitive and specific detection of *S. endobioticum* in infested and spiked soil samples.

Systemic infection of plants with bipartite *Tobacco mosaic virus* transient expression vectors. D. J. LEWANDOWSKI, E. Knapp, and G. Danyluk. Dept. Plant Pathology, CREC, University of Florida, Lake Alfred, FL 33850. *Phytopathology* 92:S46. Publication no. P-2002-0335-AMA.

To make transient viral expression vectors more versatile, multiple component vectors have been constructed from the monopartite genome of *Tobacco mosaic virus* (TMV). We artificially divided the TMV genome and built expression vectors based upon a helper virus that expresses the larger

replicase protein and efficiently replicating defective RNAs (dRNAs) that express the smaller (126-kDa) replicase protein. To express foreign genes from the dRNA, the TMV coat protein subgenomic mRNA (sgRNA) promoter was inserted behind the 126-kDa protein ORF, followed by the ORF for the green fluorescent protein (GFP). Replication and movement of these 126-kDa protein-based GFP-replicons were supported *in trans* by the replication-competent helper virus. Accumulation of a novel GFP sgRNA and GFP protein were detected in cells co-infected with helper virus plus GFP-replicon. Bipartite TMV vectors moved systemically and GFP was expressed in upper leaves of *Nicotiana benthamiana*. Dividing sequences (functions) between multiple RNAs may allow for the simultaneous expression of multiple and perhaps larger proteins.

Luteovirus-binding proteins associated with aphid transmission specificity. C. Li (1), F. E. GILDOW (2), and D. Cox-Foster (1). (1) Dept. Entomology; (2) Dept. Plant Pathology, Pennsylvania State University, University Park, PA 16802. Phytopathology 92:S47. Publication no. P-2002-0336-AMA.

Aphid proteins specifically binding barley yellow dwarf luteovirus (BYDV) and potentially functioning in vector-specific transmission were identified using virus-overlay assays on blots containing proteins extracted from aphid heads and separated by 2-dimensional urea SDS-PAGE. Three proteins (MW/pI = 35/4.35, 49/4.56, 50/4.51) from the efficient vector, *Sitobion avenae*, reacted with BYDV-MAV and PAV; and a fourth protein (50/4.41) reacted only to PAV. Three similar proteins (35/4.35, 50/4.51, 50/4.41) were identified in *Metopolophium dirhodum* (transmitting MAV, not PAV). Two proteins (33/4.26, 49/4.56) from *Rhopalosiphum padi* (transmitting PAV, not MAV) also bound to MAV and PAV. No proteins from the non-vector, *Rhopalosiphum maidis*, bound to either of these viruses. These proteins also were identified using antiidiotypic MAV-4 antibody which mimics MAV capsid epitopes. The presence of specific proteins corresponded to the ability to vector MAV or PAV. Results suggest that several aphid proteins are involved in movement of luteoviruses through vector tissues.

for in planta expressed genes in *Magnaporthe grisea*. L. LI, C. Y. Xue, and J. R. Xu. Dept. Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907. Phytopathology 92:S47. Publication no. P-2002-0337-AMA.

The rice blast disease caused by *Magnaporthe grisea* is the most destructive disease of rice worldwide. In the past decade, signaling pathways regulating the infection process have been extensively investigated in *M. grisea*. However, there is only limited knowledge about the molecular events taking place after penetration. The objective of this study is to identify genes specifically expressed after penetration. A promoterless GFP gene with the TrpC terminator was constructed and transformed into wild type strain Guy11. Over 900 hygromycin-resistant transformants were isolated. After screening 150 transformants, we identified one transformant L329 with strong green fluorescence in the infectious hyphae formed inside onion epidermal cells. No green fluorescence was observed in conidia, germ tubes or appressoria, indicating that GFP was specifically expressed after penetration. We are currently isolating the promoter responsible for GFP expression in L329 and characterizing the corresponding gene. Efforts to generate and identify additional transformants expressing GFP after penetration are also under way.

Polygalacturonases of *Sclerotinia sclerotiorum*: expression during saprophytic and parasitic modes of growth. R. LI, R. Rimmer, A. Sharpe, G. Seguin-Swartz, and D. Hegedus. Agriculture Canada, 107 Science Pl. Saskatoon, SK, Canada, S7N 0X2. Phytopathology 92:S47. Publication no. P-2002-0338-AMA.

Polygalacturonases (PGs) are important determinants for *S. sclerotiorum* infection, which under conducive conditions leads to severe yield losses. Using isoelectric focusing we identified numerous PG isoforms and undertook to examine the roles specific PGs play during the infection. Three distinct cDNAs (SSPG4, SSPG5, and SSPG6) were isolated using degenerate PCR or Expressed Sequence Tags. Expression of *Sspg4* was highly influenced by the developmental stage and physical environment in contact with the mycelia. Only actively growing mycelia at the expanding colony edge, either in agar plates or infected plants, expressed *Sspg4*. In liquid culture its expression rapidly declined. Conversely, expression of *Sspg5* was induced by citrus pectin, galacturonic acid, but was subject to glucose catabolite repression. *Sspg5* was expressed only weakly within infected tissue. The expression of *Sspg6* was minimal under all conditions tested. These results indicate that *Sspg* genes are subject to different regulatory mechanisms that reflect their specific roles under saprophytic and parasitic modes.

***Brassica napus* polygalacturonase inhibitors are differentially regulated by abiotic stress, wounding and fungal infection.** R. LI, R. Rimmer, M. Yu, M. Gruber, A. Sharpe, G. Seguin-Swartz, and D. Hegedus. Agriculture and Agri-Food Canada, 107 Science Place, Saskatoon, SK, Canada, S7N 0X2. Phytopathology 92:S47. Publication no. P-2002-0339-AMA.

Plant pathogenic fungi produce several forms of polygalacturonase (PG) to promote tissue invasion and provision of nutrients. In turn, plants express a distinct set of polygalacturonase inhibitory proteins (PGIPs) to interfere with this. To improve resistance of *B. napus* to necrotic pathogens we are studying the interaction between *S. sclerotiorum* PGs and *B. napus* PGIPs. Two cDNAs encoding *B. napus* PGIPs (*Bnpgip1* and *Bnpgip2*) were isolated. *Bnpgip1* expression was highly responsive to flea beetle and mechanical wounding, weakly to *S. sclerotiorum* infection, cold shock and not to dehydration. Conversely, *Bnpgip2* expression was strongly induced by *S. sclerotiorum* infection and to a lesser degree by wounding. *Bnpgip2* expression was localized to tissues adjacent to the infection and was not systemically produced. Jasmonic, but not salicylic acid, induced *Bnpgip1* expression, whereas *Bnpgip2* did not respond to either. Divergent systems appear to govern PGIP expression in response to environment cues and they likely play different roles in plant defense.

and molecular variability of *Fusarium solani* f. sp. *glycines*. S. LI (1), G. Hartman (1,2), and W. Pedersen (1). (1) Dept. Crop Sciences, University of Illinois, Urbana, IL 61801; (2) USDA-ARS. Phytopathology 92:S47. Publication no. P-2002-0340-AMA.

Sudden death syndrome of soybean is caused by *Fusarium solani* f. sp. *glycines* (FSG). A total of 78 FSG isolates were collected from different geographic locations. These isolates were used to inoculate a susceptible soybean cultivar, Great Lakes 3202, in a growth chamber using FSG-infested sorghum, which was placed below seeds at planting. Foliar symptoms, root and root lesion lengths, and shoot and root dry weights were recorded 21 days after planting. There was a significant difference ($P < 0.05$) among isolates for foliar disease severity ranging from 2 to 5 using a 1 to 5 disease severity scale where 1=no disease and 5=severely infected or dead plants. Root lesion lengths varied from 12 to 110 mm. Significant differences also were observed in shoot and root dry weights. Amplified fragment length polymorphism (AFLP) analysis detected DNA polymorphisms using eight combinations of fluorescence-labeled primer sets in the selective amplification. In this study, FSG isolates varied in aggressiveness on soybean and in DNA polymorphisms based on AFLP analysis.

Response of hairy roots of different soybean genotypes to *Fusarium solani* f. sp. *glycines*. S. LI, A. Lygin, O. Zernova, V. Lozovaya (1), G. Hartman (1,2), and J. Widholm (1). (1) Dept. Crop Sciences, University of Illinois, Urbana, IL 61801; (2) USDA-ARS. Phytopathology 92:S47. Publication no. P-2002-0341-AMA.

To test the response of soybean to *Fusarium solani* f. sp. *glycines* (FSG), cotyledon explants of 14 genotypes were incubated with *Agrobacterium rhizogenes* strain K599. Hairy roots were produced from the wounded surface of the cotyledon explants on MXB medium at 25°C in the dark for 3-4 weeks. A mycelial plug, 4-mm-diameter, from the margin of 2-week-old cultures of the FSG isolate Mont-1 was placed mycelial side down directly on top of the hairy roots, as well as potato dextrose agar (PDA), and MXB medium. FSG growth diameters were measured 10 days after inoculation. There were significant differences ($P < 0.05$) among genotypes for fungal growth with mean values ranging from 17 to 40 mm. FSG grew faster on the hairy roots of Spencer, Peking and Essex than on Ripley, and PI 520.733. PDA favored FSG growth, but not the MXB medium. FSG growth differences were also found among genotypes inoculated with 10 microliters of FSG macroconidial suspension. FSG colonies were detected on surface disinfected ground hairy roots 4 days after inoculation indicating FSG had colonized hairy roots.

Relative virulence of *Phytophthora* species, including the sudden oak death pathogen *P. ramorum*, on leaves of several ornamentals. R. G. LINDERMAN (1), J. L. Parke (2), and E. M. Hansen (2). (1) USDA-ARS Horticultural Crops Research Laboratory, Corvallis, OR 97330; (2) Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331. Phytopathology 92:S47. Publication no. P-2002-0342-AMA.

Several *Phytophthora* species cause leaf and shoot dieback diseases of ornamentals similar to that caused by the Sudden Oak Death pathogen, *P. ramorum*. Detached leaves of several landscape plants were inoculated with *P. ramorum*, *P. cactorum*, *P. syringae*, *P. citricola*, *P. hevea*, *P. parasitica*, *P. citrophthora*, and *P. cinnamomi*. Rhododendron, Pieris, and Laurel were the most susceptible to the most pathogens, based on lesion-severity ratings,

with variation depending on the host-pathogen combination. Few hosts were not susceptible to some *Phytophthora* species, and most pathogens infected some hosts. Necrotic lesions were initially similar, but generally with most pathogens subsequent spread was limited. In contrast, *P. ramorum* and *P. citrophthora* lesions spread throughout the entire leaf, suggesting greater virulence and thus underscoring the risk to nursery and landscape plants should the quarantined *P. ramorum* become more widespread.

Molecular characterization and phylogenetic analysis of *Colletotrichum* species using a 1 kb intron of glutamine synthetase gene (GS) and 200 bp intron of glyceraldehyde phosphate dehydrogenase gene (GPDH). B. Liu, J. C. Guerber, and J. C. CORRELL. Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701. Phytopathology 92:S48. Publication no. P-2002-0343-AMA.

The genus *Colletotrichum* is exceptionally diverse and includes saprophytic and plant pathogenic species. The demarcation of species within the genus is based on morphological characters and host range, and often does not reflect the wide genetic variation within a species nor the phylogenetic relationships among species. Introns from two independent genes, a 1 kb GS intron and a 200 bp GPDH intron, were used to characterize a range of isolates representing 15 species. Both introns were amplified by specific primers and examined for RFLP and sequence variation. RFLPs were particularly diagnostic for qualitatively distinguishing the various species. Quantitative, or phylogenetic, relationships based on sequence alignment showed clear differentiation among the various species. The phylogenetic dendrograms of the two introns were congruent. The RFLP and sequence data from this study suggest that taxonomic relationships within and between species of *Colletotrichum* could reliably be assessed using both the 1 kb GS and the 200 bp GPDH introns.

Visualizing PhcA-regulated virulence gene expression of *Ralstonia solanacearum* using transcriptional fusions expressing green fluorescent protein (GFP). H. LIU, Y. Kang, and T. P. Denny. Dept. Plant Pathology, Univ. of Georgia, Athens, GA 30602. Phytopathology 92:S48. Publication no. P-2002-0344-AMA.

To visualize expression of *R. solanacearum* virulence genes regulated by PhcA, the central component of a confinement-sensing regulatory network, we created *cis*-merodiploid strains containing *gfp-mut3a* (with or without a C-terminal destabilization peptide) driven by *eps* or *pilA* promoters. Microscopic colonies of the *Peps-gfp* strains were nonmucooid and nonfluorescent for 30 h after streaking. Fluorescence was observed in the center of colonies starting about 32 h, and gradually increased until by 72 h the whole colony fluoresced. Visible mucoidy increased along with fluorescence intensity. In contrast, colonies of *PpilA-gfp* strains that made unstable GFP fluoresced brightly for up to 40 h. Fluorescence then began to decrease, starting in the colony center, until by 72 h only the margin remained fluorescent. Thus, as PhcA became functional in response to high cell density within the colonies, it positively regulated transcription of *eps* and negatively regulated *pilA*.

Effect of pH on growth and sporulation of *Hirsutella minnesotensis* and *Hirsutella rhossiliensis* in vitro. S. F. LIU and S. Y. Chen. University of Minnesota Southern Research and Outreach Center. Phytopathology 92:S48. Publication no. P-2002-0345-AMA.

The fungi *Hirsutella minnesotensis* and *Hirsutella rhossiliensis* occur naturally in agricultural fields and parasitize a wide range of nematodes. The objective of this study was to determine the effect of pH on their growth and sporulation. Cornmeal agar plates were prepared and their pH was adjusted to 4, 5, 6, 7, 8, 9, and 10 by adding 1N HCl or 1N NaOH. The fungi *H. minnesotensis* isolate WA23-1 and *H. rhossiliensis* isolate OWVT-1 were transferred to the culture media and incubated at room temperature (22-24°C). Colony diameters were recorded at days 3, 7, 14, 21, 28, 35, 42, 49, and 56 after transferring, and the number of spores produced per cm² colony was measured at day 35. The two species grew very well on the media at pH 5 to 8. The optimum pH was approximately 5.5 for *H. minnesotensis* growth and approximately 6 for *H. rhossiliensis*. The growth of both species was reduced drastically when pH was reduced to 4. Optimum pH for *H. minnesotensis* and *H. rhossiliensis* sporulation was between 6 and 7, and the sporulation was greatly inhibited by either low pH at 4 or high pH at 9 and 10. This data shows that the growth and sporulation of the two fungi are not limited between pH 5 and 8, which encompasses the normal range of soil pH for Minnesota soybean production.

CP-RTD genes of BYDV-PAV isolates affect aphid transmission efficiency and symptom severity. S. LIU and W. A. Miller. Dept. of Plant

Pathology, Iowa State University, Ames, IA 50011. Phytopathology 92:S48. Publication no. P-2002-0346-AMA.

Barley yellow dwarf virus (BYDV) encodes two capsid proteins: a major coat protein (CP) and a minor translational readthrough domain (RTD), which are necessary for aphid transmission. We constructed four chimeric infectious transcripts with the CP-RTD derived from an Australian isolate (PAV6), a similar Illinois isolate, or a very divergent, severe New York isolate (PAV-129). All transcripts were infectious in oat protoplasts, but the PAV6-129 chimera accumulated 10 times less RNA than PAV6. Aphid transmissibility and ability to infect whole plants was tested by feeding *Rhopalosiphum padi* on purified virus through Parafilm, and then transferring the aphids to oat seedlings. All four chimeric viruses infected oat plants, with varying transmission efficiencies. Significantly, PAV6 had the lowest transmission efficiency and PAV6-129 had the highest. Yet PAV6 virus accumulated to higher levels than PAV6-129. Symptoms induced by PAV6-129 were much more severe than PAV6 and similar to the wild type PAV129. We conclude that the CP-RTD determine both transmission efficiency and symptomology, and that these were unaffected by virus titer.

Control of monosporascus root rot of melon using *Trichoderma* spp. in Taiwan. C. T. LO and J.-H. Huang. Department of Plant Pathology, Taiwan Agricultural Research Institute, Taiwan. Phytopathology 92:S48. Publication no. P-2002-0347-AMA.

Vine decline and root rot of melons caused by *Monosporascus cannonballus* pollack & Uecker has been reported in several countries in the world including Taiwan. Recently, the disease has persisted to be a limiting factor to melon production in some major commercial areas in Taiwan. The pathogen usually caused yellowing and death of leaves and gradual decline of the vines as the plant is going to maturity. In taproot and some lateral roots, the diseased plants showed necrotic discrete lesions and lack of secondary and tertiary roots. The pathogen produced the black perithecia in both host roots and culture, and usually formed only one large ascospore per ascus. *Trichoderma* spp. have been used as a biocontrol agent to protect plants against soil-borne and foliar diseases in several crops. Consequently, the purpose of this study was to determine whether selected strains of *Trichoderma* spp. in Taiwan are able to reduce the disease caused by *M. cannonballus*. In dual tests, most of isolates of *Trichoderma* spp could inhibit mycelial growth of *M. cannonballus* and then degrade the mycelia, when the biocontrol agents began to contact with the pathogen on malt extract media. For field trials in 1999 and 2000, seeds of cho-hwa cultivar of *Cucumis melo* L. were grown in soilless mixture previously separately inoculated with strain 1295-22 of *T. harzianum* and strain R42 of *T. virens*. After two weeks, the seedlings were then transplanted to field. The results indicated that both strains of *Trichoderma* spp. could promote plant growth of melon and reduce the disease incidence as compared with untreated plots.

Effect of three alternative fungicide materials on the viability and germinability of sclerotia of the soil pathogens *Sclerotinia sclerotiorum* and *Sclerotium rolfii*. J. C. Locke. USDA, ARS, USNA, FNPRU. Phytopathology 92:S48. Publication no. P-2002-0348-AMA.

The fungal soilborne pathogens *Sclerotinia sclerotiorum* and *Sclerotium rolfii* produce numerous sclerotia which serve to allow long-term survival and dissemination. In order to reduce diseases caused by these pathogens, it is important to consider control practices which will greatly reduce the viability and/or germinability of these survival propagules. Uniform sclerotia, produced in the laboratory, were exposed in liquid shake culture to a botanical material and two fungicides of novel chemistry at different concentrations and times of exposure. Sclerotia were recovered, washed of exogenous test solution and transferred to agar plates to evaluate viability and vigor over time. Sclerotia of *S. rolfii*, although larger in size than those of *S. sclerotiorum*, were more sensitive to two of the three test materials, becoming inactive after less exposure time. In some cases, sclerotial germination was delayed by sub-lethal exposure but viability was not completely lost indicating that some cells of the sclerotia were destroyed but some survived to germinate after longer periods of time.

Mutation of an *sdhA* homologue in *Enterobacter cloacae* results in reduced colonization of cucumber but does not affect biocontrol of damping-off. S. M. LOHRKE (1), L. McKenna (1), C. J. Baker (2), S. Liu (3), P. D. Dery (1), J. S. Buyer (1), and D. P. Roberts (1). (1) Sustainable Agricultural Systems Laboratory; (2) Molecular Plant Pathology Laboratory, USDA-ARS, Beltsville, MD 20705; (3) Oil Crops Research Institute, Chinese Academy of Agricultural Science, Wuhan, People's Republic of China. Phytopathology 92:S48. Publication no. P-2002-0349-AMA.

Identification of genes involved in seed and root colonization by biocontrol bacteria may allow for strategies for improved ecological fitness, survival, and performance by these strains. *Enterobacter cloacae* strain M2, containing a single transposon insertion, was reduced in colonization of cucumber roots relative to wild-type strain 501R3 but unaffected in control of *Pythium ultimum* damping-off on cucumber. DNA sequence analysis indicated that the transposon was inserted in *sdhA*, which encodes a subunit of succinate dehydrogenase. The growth profile of strain M2 was consistent with an *sdhA* mutant. Succinate dehydrogenase is involved in aerobic respiration and is responsible for a key metabolic step in the TCA cycle, catalyzing the conversion of succinate to fumarate.

Construction of pMEKm12 for expressing Syr proteins in *Pseudomonas syringae* required for syringomycin production. S.-E. LU (1), B. K. Scholz-Schroeder (2), and D. C. Gross (1). (1) Dept. Plant Pathology & Microbiology, Texas A&M Univ., College Station, TX 77843; (2) Dept. Plant Pathology, Washington State Univ., Pullman, WA 99164. Phytopathology 92:S49. Publication no. P-2002-0350-AMA.

Syringomycin is synthesized by a nonribosomal peptide synthetase system in *P. syringae* pv. *syringae*. Overexpression of genes encoding Syr proteins in the native bacterium can be critical to the characterization of enzymatic functions. Vector pMEKm12 was constructed to overproduce Syr proteins that may require posttranslational modification not performed by *E. coli*. The vector pMEKm12 is composed of the pRO1600 replication origin, the maltose-binding protein (MBP) gene, the *ntpIII* gene for selection, and an inducible *tac* promoter. The *syrB1* gene was PCR amplified and cloned into pMEKm12 to generate a *malE-syrB1* in-frame fusion. The MBP-SyrB1 protein was produced in *P. s.* pv. *syringae* and purified to a final yield of 5 mg/liter. Expression of the *malE-syrB1* and *malE-syrB2* fusion genes *in trans* restored syringomycin production to their respective *syrB1* and *syrB2* mutants. The pMEKm12 vector is a convenient system for expressing high quantities of enzymatically active proteins in *Pseudomonas*.

Pathogen colonization and chitinase accumulation in soybeans with tolerance to Phytophthora root rot. K. LUCE, A. Karr, and J. English. Univ. MO., Dept. Plant Microbiology and Pathology. Phytopathology 92:S49. Publication no. P-2002-0351-AMA.

Tolerance to Phytophthora root rot, caused by *Phytophthora sojae*, is a highly desirable trait in soybean lines. However, specific events in infection and plant defense responses are not well understood. To characterize the timing of early events in the expression of tolerance in soybeans, taproots of 14-day-old soybean plants, designated as tolerant or susceptible, were inoculated with *P. sojae* zoospores. Lateral roots of the same plants were inoculated 48 hr later. Chitinase accumulation, as an indicator of plant defense response, and root colonization were compared after 48 hr additional incubation. At that time little difference in linear root colonization or chitinase accumulation was observed between the tolerant and susceptible cultivars. Chitinase activity was highest in colonized root tissue. However, chitinase accumulation in non-colonized roots of infected plants was higher than in non-inoculated control plants. These results suggest a defense response may be activated in root tissue of ahead of pathogen colonization; future work will characterize the timing of defense response activation in tolerant plants.

Dynamics of inoculum potential of *Monilinia fruticicola* in relation to cultural practices in prune orchards. Y. LUO (1), T. J. Michailides (1), D. P. Morgan (1), W. H. Krueger (2), and R. P. Buchner (3). (1) Dept. Plant Pathology, University of California, Davis, Kearney Agricultural Center, Parlier, CA 93648; (2) University of California, Cooperative Extension Center, Glenn County, Orland, CA 95963; (3) University of California, Cooperative Extension Center. Phytopathology 92:S49. Publication no. P-2002-0352-AMA.

Spore density (ascospores and conidia) of *Monilinia fruticicola* in the air was investigated daily using spore traps in two prune orchards in California during the growing season. Effects of fruit thinning, irrigation and fungicide application on sporulation of thinned fruit were studied. The spore densities in the air were at a low level in early bloom, increased to a high level at full bloom, and decreased to a lowest level at the end of bloom. Spore density in the air increased tremendously during a 7 - 10 day period just after each irrigation that created favorable conditions for sporulation on thinned fruit on the ground. Prolonging the drying of thinned fruit significantly decreased fruit infection. Fungicide treatment on thinned fruit on the ground significantly reduced latent infections of fruit on trees.

Threshold conditions leading latent infection to prune fruit rot caused by *Monilinia fruticicola*. Y. LUO and T. J. Michailides. Dept. Plant Pathology,

University of California, Davis, Kearney Agricultural Center, Parlier, CA 93648. Phytopathology 92:S49. Publication no. P-2002-0353-AMA.

Inoculations were performed in 10 prune orchards in California eight times during the growing season. Branches with blossoms or fruit were sprayed with 5,000, 20,000, and 50,000 conidia/ml of *Monilinia fruticicola*. Each inoculated branch was covered with a plastic bag to keep high humidity for about 14-16 h. The incidence of latent infection (ILI) on the fruit and the percentage of branches with fruit rot (PBFR) were determined 2 weeks before harvest. A linear correlation between ILI and PBFR was obtained. Conditions leading latent infection to fruit rot included level of latent infection, fruit development stage, inoculum concentration, total hours of RH greater than 90%, and hours of dew from mid-July to mid-August. Three levels of PBFR, 1, 5, and 10%, were assigned and threshold conditions leading to these levels of PBFR were determined. The relative possibility of latent infection becoming fruit rot (r_{PBFR}) and PBFR were used in a preliminary decision support model and recommendations on fungicide application could be provided based on input information supplied by the grower.

Mycotoxigenic *Fusarium* and deoxynivalenol production influence chitinase gene expression in *Trichoderma atroviridae*. M. P. LUTZ (1), G. Feichtinger (1), G. Defago (1), and B. Duffy (2). (1) Phytopathology group, Swiss Federal Institute of Technology, Zurich, Switzerland 8092; (2) USDA-ARS, Albany, CA 94710. Phytopathology 92:S49. Publication no. P-2002-0354-AMA.

Fusarium head blight of wheat caused by mycotoxigenic *Fusarium* is an increasing threat worldwide to crop, animal and human health. The fungi survive and sporulate in crop residues. *Trichoderma atroviridae* is a potential biocontrol agent for *Fusarium*. We tested the impact deoxynivalenol (DON) producing *Fusarium* on expression of two *Trichoderma* chitinase genes, *ech42* and *nag1*, which are important for biocontrol activity. *Trichoderma* gene expression was monitored in assays *in vitro* and on maize crop residues using *goxA* reporter gene fusions. We found that DON-producing *Fusarium* strains repressed expression of *nag1* but not *ech42*. DON-negative *Fusarium* or a non-producing *tri5* mutant had no effect on biocontrol gene expression. Synthetic DON added in assays with non-producing *Fusarium* resulted in repression of *nag1*. This is the first demonstration of a target pathogen down-regulating genes in a fungal biocontrol agent, and suggests a novel ecological function for mycotoxins as a factor in *Fusarium* competitiveness.

Composted amendments alter soil microbial communities and suppress southern blight. M. T. LYONS (1), A. D. Peacock (2), D. C. White (2), and C. E. Sams (1). (1) Dept. of Plant Sciences and Landscape Systems, The University of Tennessee, Knoxville, TN 37996; (2) Center for Biomarker Analysis, Knoxville, TN 37932. Phytopathology 92:S49. Publication no. P-2002-0355-AMA.

Composts are effective for the prevention of southern blight (*Sclerotium rolfsii*) in plasticulture tomato production. Extraction of Phospholipid fatty acids (PLFAs) from soil samples allows for the quantitative evaluation of microbial profiles and the characterization of disease suppression. The mean microbial biomass of composted plots (1.2×10^{-5} pmol/g dry wt.) was significantly higher ($P < 0.05$) than the controls (77×10^{-5} pmol/g dry wt.). Branched monounsaturates and monounsaturates were also higher. The latter indicates an increase in gram-negative bacteria. The relative proportion of polyunsaturated PLFA was lower in the composted plots. These signature lipid biomarkers generally indicate the presence of microeukaryotes. The compost treatment had 59% higher tomato yields in 2000 than the control. In 2001 the compost treatment yield was increased by 91%. Disease ratings were 84% and 67% lower ($P < 0.05$) in the compost treatments than in controls in 2000 and 2001, respectively.

Resistance to bacterial angular leafspot disease, caused by *Xanthomonas fragariae*, in strawberry. J. L. MAAS, J. S. Hartung, C. T. Gouin, K. Lewers, and S. Hokanson. Fruit Laboratory, Agricultural Research Service, U.S. Department of Agriculture, Beltsville, MD 20705. Phytopathology 92:S49. Publication no. P-2002-0356-AMA.

Bacterial angular leafspot disease of strawberry has become an increasingly important problem for strawberry producers and nurserymen worldwide. No suitable chemical control strategies are available, nor until now have any sources of resistance to this disease been made available to breeders. We screened numerous *Fragaria* genotypes against two of the four genotypic strain groups of *X. fragariae* and determined that *F. virginiana* SG-89 and 08-4-38 (*F. virginiana* × *F. × ananassa* 'Earliglow') were resistant. In subsequent tests against all four genotypic strain groups, these strawberry clones were highly resistant. The resistance exhibited was not of the hyper-

sensitive host reaction type. Crosses were made between these clones and a susceptible parent, 'Sweet Charlie', and the progeny screened against all four genotypic strains. Transmission of resistance to progeny populations varied from 4-18%, depending on the challenge strain used. Resistance apparently involves two recessive genes, according to Chi-square analyses of data. These two parental clones are being released to strawberry breeders as US 4808 (SG-89) and US 4809 (80-4-38). Plants of each clone in tissue culture may be obtained from the senior author (maasj@ba.ars.usda.gov).

A uniformity trial investigating variation in yield and the incidence of Botrytis fruit rot in annual strawberry. S. J. MACKENZIE, C. L. Xiao, J. C. Mertely, and D. E. Legard. Univ. of Florida, GCREC, Dover, FL 33527. Phytopathology 92:S50. Publication no. P-2002-0357-AMA.

Marketable yield and incidence of Botrytis fruit rot for individual strawberry plants was collected over two seasons for cultivar Sweet Charlie and one season for Camarosa. A nested ANOVA model was used to determine plot edge effects and obtain variance components to describe the relationship between plot size and variance. Plants grown on the edge of plots had higher yields for both cultivars, but there was no edge effect for Botrytis. Smith's index, a measure of field heterogeneity, was relatively high for yield (0.92 and 0.95) and Botrytis (0.91 and 0.69) for 'Sweet Charlie' and 'Camarosa' respectively. This suggests an even distribution of these variables among plants. Power analysis of treatments with four replicates of 32, 16 or eight plants/plot suggest that yield effects from 13 to 24% of the average yield for 'Sweet Charlie' and from 10 to 19% of the average for 'Camarosa' can be detected with a power of 0.80. Treatment differences of 28 to 49% of the mean incidence of Botrytis fruit rot for 'Sweet Charlie' and 49 to 71% for 'Camarosa' should be detected with a power of 0.80.

Use of adjuvants to enhance biocontrol efficacy. W. F. MAHAFFEE, T. M. Sechler, and S. J. Scheuerell. USDA-ARS, Horticulture Crops Research Laboratory, Corvallis, OR 97330. Phytopathology 92:S50. Publication no. P-2002-0358-AMA.

Several commercial surfactants and food-grade polymers (similar to those found in biofilms) were tested as spray adjuvants for enhancing the biocontrol efficacy of fungal, yeast, and bacterial biocontrol agents against *Botrytis cinerea* on geranium. Adjuvants were added to spray solution of the biocontrol agent just prior to application. Plants were maintained in the greenhouse for 7-10 days, then challenged with *B. cinerea* and incubated at 25°C with >80% humidity for 5-7 days. Surfactants consisting of yucca extracts or oils significantly enhanced disease control by bacterial biocontrol agents. Karaya and xanthan gum significantly enhanced disease control by bacterial and yeast biocontrol agents. Fungal biocontrol agents were marginally enhanced by karaya. Microscopic observations of leaf surfaces indicated that the polymers increased microbial cell aggregation and resulted in films of microorganisms across epidermal leaf cells that were similar to biofilms. Addition of the adjuvants appears to enhance establishment and longevity of biocontrol agents on the leaf surface.

Diversity among *Xanthomonas axonopodis* pv. *phaseoli* strains infecting common bean from different geographical areas. G. MAHUKU, C. Jara, M. A. Henriquez, and J. Cuasquer. Centro Internacional de Agricultura Tropical, A. A. 6713, Cali, Colombia, South America. Phytopathology 92:S50. Publication no. P-2002-0359-AMA.

Xanthomonas axonopodis pv. *phaseoli* (Xap) is the causal agent of bacterial blight of common bean, *Phaseolus vulgaris* L. There are two forms of Xap, the non-pigment producing strain (Xap) and the brown-pigment producing fuscans strain (Xapf), but both have the same host range and cause similar symptoms. We used Rep-PCR and RFLP of the 16S rDNA to assess the amount of genetic variation existing within and between Xap and Xapf. A total of 425 strains from 28 countries were tested for pathogenicity on the highly susceptible bean genotype BAT 41. Molecular analysis of 343 pathogenic isolates showed that Xap and Xapf could be differentiated on the basis of polymorphisms of both RFLP of 16S rDNA and Rep-PCR patterns, and the two were very distinct from non-pathogenic xanthomonads. The average genetic similarity between Xap and Xapf was 0.45, while both strains were very distinct (average similarity coefficient of 0.23 and 0.27) between non-pathogenic xanthomonads and Xap or Xapf, respectively. No geographical differentiation was evident within and between Xap and Xapf although there was more variation within Xap (average genetic similarity 0.80) than Xapf (0.89). These results show that Xap and Xapf are genetically distinct.

Barley yellow dwarf viruses in California native perennial grasses. C. M. Malmstrom. Dept. Plant Biology, Michigan State University, East Lansing, MI 48824. Phytopathology 92:S50. Publication no. P-2002-0360-AMA.

In California, hosts of the BYDVs include many native perennial grass species, as well as most of the exotic annual grasses (e.g., *Avena* spp., *Bromus* spp.) that have largely replaced them and now dominate the rangelands. In surveys of native grass populations in the state, I have found BYDV incidence to be as great as 40%. Could the BYDVs be contributing to the decline of remnant native grass populations in California? To investigate this question, I grew individuals from different populations of several native grass species (*Elymus glaucus*, *E. multisetus*, *Hordeum brachyantherum*, *Koeleria macrantha*, and *Nassella pulchra*) and infected them as seedlings with a locally collected BYDV-PAV. The plants were grown in the field in Davis, California, for a complete growing season. Symptoms were evaluated, and biomass and seeds were harvested and weighed. Species and populations differed in response to infection, but biomass and seed production were reduced in many cases, in some by 50%. This finding demonstrates that BYDV-PAV can reduce fecundity and may decrease recruitment in some California native grasses.

Genotype distribution and cultivar preference of *Phialophora gregata*. D. MALVICK (1), W. Chen (2), and C. Grau (3). (1) Dept. of Crop Sciences; (2) Illinois Nat. Hist. Surv., Univ. of Illinois at Urbana-Champaign, Urbana, IL 61801; (3) Dept. of Plant Pathology, Univ. of Wisconsin-Madison, Madison, WI 53706. Phytopathology 92:S50. Publication no. P-2002-0361-AMA.

Brown stem rot of soybean is caused by genotypes A and B of *P. gregata*. Genotype A causes both stem and leaf symptoms, and genotype B typically causes stem symptoms only. An objective was to determine genotype distribution in the field. Plots were established at four sites in Illinois and two sites in Wisconsin in 2001. Six soybean cvs., Bell, Dwight, Sturdy, Williams 82, LN92-12033, and LN92-12054 were sown with four replications at each site. Five mature stems were harvested arbitrarily per plot and assayed by PCR to detect the A and B genotypes. Bell and LN92-12033 were preferentially infected by B, with 98 and 87% of infected stems positive for B, respectively. Sturdy, LN92-12054, and Williams 82 were preferentially infected by A with 60, 68, and 70% of infected stems positive for A, respectively. Dwight was equally infected by A and B. Genotype A was predominant in BSR susceptible cvs. and B was predominant in two of three resistant cvs. Both genotypes were detected at all locations except one in Illinois where only A was detected.

Automation of Tomato spotted wilt virus inoculation. B. Mandal (1), A. S. CSINOS (1), H. R. Pappu (2), N. Martinez-Ochoa (1), and A. K. Culbreath (1). (1) Department of Plant Pathology, University of Georgia, Tifton, GA 31793; (2) USDA-APHIS-PPQ, Riverdale, MD 20737. Phytopathology 92:S50. Publication no. P-2002-0362-AMA.

TSWV inoculation experiments were conducted in the greenhouse using a pressure spray-gun, which was devised by connecting an atomizer with a carbon dioxide powered sprayer. An isolate of TSWV maintained by hand inoculation on tobacco cv. K326 was used as a source of inoculum. Inoculum was prepared by grinding infected leaves at the rate of 1g tissue with 10 ml of 0.1M phosphate buffer containing 0.2% sodium sulfite and 0.01M mercaptoethanol. The extract was filtered through cheese cloth and 1% each of Celite and Carborundum 320 grit were added to the inoculum. Forty day old K326 seedlings were used for the inoculation studies. Inoculation was accomplished by spraying the inoculum at a height of 10 to 12 cm above the plants. Plants produced systemic symptoms at 10 to 14 days post inoculation. Hundred percent infection was obtained by using 1.1 ml inoculum per plant at a pressure of 3.44 or 4.13 bar. This approach offers a rapid and efficient method for inoculating a large number of tobacco seedlings.

Susceptibility of peanut and sunflower to *Impatiens necrotic spot virus*. B. Mandal (1), H. R. Pappu (2), and A. K. CULBREATH (1). (1) Department of Plant Pathology, University of Georgia, Tifton, GA 31793; (2) USDA-APHIS-PPQ, Riverdale, MD 20737. Phytopathology 92:S50. Publication no. P-2002-0363-AMA.

The susceptibility of peanut and sunflower to INSV was evaluated by mechanical inoculation and infection was confirmed by enzyme linked immunosorbent assay. Percent transmission in the five peanut cultivars (Georgia Runner, Georgia Green, C-99R, MDR and Tamrun 96) varied from 20 to 55%. Inoculated leaves of peanut showed yellow patches but new leaves did not show any symptoms. Distribution of INSV in the peanut plant was limited to the inoculated leaves and roots but not in the non-inoculated foliage. Hybrid sunflower varieties, 776CRT, 54SA and 652 and ornamental sunflower varieties, Gyganteus and Sunbeam were infected by INSV. Percent transmission in the hybrid and ornamental sunflower varied from 21 to 45.4% and 60 to 82.4% respectively. Symptoms in all these sunflower varieties were yellow spots and yellowing of lower leaves; only Sunbeam showed necrotic spot symptoms.

Feeding preference of *Frankliniella fusca* for selected peanut cultivars and a breeding line. B. Mandal (1), H. R. Pappu (2), L. Wells (1), A. K. CULBREATH, and J. W. Todd (3). (1) Department of Plant Pathology; (3) Entomology, University of Georgia, Tifton, GA 31793; (2) USDA-APHIS-PPQ, Riverdale, MD 20737. Phytopathology 92:S51. Publication no. P-2002-0364-AMA.

The feeding preference of *Tomato spotted wilt virus* (TSWV) vector, *F. fusca* for three peanut cultivars, Georgia Green, Georgia Runner and C-99R, and one breeding line, C11-2-39, was examined under choice and non-choice feeding conditions. While Georgia Runner is highly susceptible to TSWV, the other three genotypes display varying degrees of field resistance to TSWV. In the choice feeding experiment, thrips had access to the leaves of all the genotypes whereas in the non-choice layout, thrips had access to the leaves of only one of the four genotypes. Thrips were allowed to feed either on a detached leaf or on whole potted plants. Feeding preference was compared by scoring the feeding scars on the leaves. In all the feeding experiments, *F. fusca* showed similar feeding preference for all the peanut genotypes except in the case of the potted plants under macro-cage, where the breeding line, C11-2-39 was preferred more than the three cultivars.

Systematic replacement of amino acid residues in conserved regions of Ptr ToxA. V. A. Manning, R. M. ANDRIE, and L. M. Ciuffetti. Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331. Phytopathology 92:S51. Publication no. P-2002-0365-AMA.

Ptr ToxA is a unique host-selective toxin of *Pyrenophora tritici-repentis*. Amino acid (aa) analysis has identified residues potentially important to ToxA activity, including 2 myristoylation sites, 7 tyrosine phosphorylation sites, and a conserved RGD cell attachment site. Heterologous expression of ToxA with a RGD to AGA mutation yields a protein with decreased activity. The aa residues surrounding the putative cell attachment motif show strong similarities to the same region in the mammalian integrin-binding protein, vitronectin, which suggests that ToxA may interact with an integrin-like receptor. Integrins have been implicated in cell adhesion, signaling, and morphogenesis. Site-directed mutagenesis has been used to mutate single aa residues of the vitronectin-like motif to determine the importance of these residues on the activity of ToxA. The effect of each mutation on activity was assessed by a leaf infiltration bioassay and quantified via a chlorophyll assay. Competition assays of mutants with native ToxA will be used to evaluate the importance of this region to ToxA binding to a putative receptor.

A race for a novel host-selective toxin. V. A. Manning, I. Pandelova, and L. M. CIUFFETTI. Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331 U.S.A. Phytopathology 92:S51. Publication no. P-2002-0366-AMA.

Isolates of *Pyrenophora tritici-repentis* were tested for pathogenicity on differential wheat lines (Katepwa, Glenlea, 6B365 and Salamouni). Inoculation of one of these isolates induced pronounced disease symptoms on Katepwa and Glenlea only. Southern blot analysis determined this isolate does not contain either the *ToxA* or *ToxB* genes. Differences in the severity of disease on Katepwa and Glenlea reflect differences in sensitivity to a partially purified necrosis toxin present in crude culture filtrate. Toxic activity was abolished when treated with Proteinase K and decreased after boiling. Data indicate that we have identified a new race of *P. tritici-repentis* that produces a previously undescribed proteinaceous host-selective toxin, designated Ptr ToxD. Further work on purification and characterization of this protein is in progress.

Bud union disorder in navel associated with a graft transmissible agent. L. J. MARAIS (1) and N. V. O'Connell (2). (1) University of California Riverside; (2) UCCE, Tulare County. Phytopathology 92:S51. Publication no. P-2002-0367-AMA.

A bud union disorder was observed in 9-year-old navel on Carrizo citrange rootstocks in Tulare County. The foliage of affected trees was a dull green, with veins and leaves turning chlorotic with time. Excessive leaf drop occurs which leads to the formation of sparse canopies. Symptoms first appear in sectors of the canopy, eventually enveloping the entire canopy. Leaf drop is followed by twig die-back and affected trees are also stunted. Examination of the bud unions of affected trees revealed a distinct crease several millimeters deep. Removal of a window of bark over the bud union showed the crease to be impregnated with gum. Frequently only sectors of the crease are impregnated with gum and it is these sectors which are associated with sectors exhibiting symptoms in the canopy. It takes an affected tree 3-6 months to collapse following the onset of symptoms. To identify the causal agent, affected trees were indexed for citrus tristeza virus (CTV), citrus tatterleaf virus (CTLV) and citrus viroids (CVds) using standard biological

indicators Mexican lime, cowpea and Citrus excelsa, and Etrog citron Arizona 86-S1, respectively. The ELISA test was also used to screen for CTV. Trees propagated from shoot-tip grafted budwood, which did not exhibit these symptoms were also indexed. Indexing results for the shoot-tip grafted trees were all negative, however the trees exhibiting the bud union disorder were found to be infected with CVds. Tests for CTV and CTLV were negative.

A novel method for *in vitro* quantification of biofilm and planktonic populations of the plant pathogenic bacterium *Xylella fastidiosa*. L. L. R. MARQUES (1), H. Ceri (1), G. P. Manfio (2), and M. E. Olson (1). (1) Biofilm Research Group, University of Calgary, AB, Canada T2K 1N4; (2) CPQBA - UNICAMP, CP 6171, Campinas, SP, 13081-970, Brazil. Phytopathology 92:S51. Publication no. P-2002-0368-AMA.

Xylella fastidiosa are able to colonize the xylem of a range of host plants and can cause economically important diseases, such as Pierce's disease (PD) in grapevines and citrus variegated chlorosis (CVC) in sweet oranges. In a previous study, we described a simple and relatively rapid method for the study of *X. fastidiosa* biofilms *in vitro*, using pine wood sticks. In the present study, an enhancement of that method is presented, using balsa wood (*Ochroma* sp., angiosperm). Remarkable increase in colonization efficiency of the balsa wood surfaces was observed on SEM analyses, particularly for elm and grape strains. Balsa wood immersed in the culture media also enhanced the growth of planktonic populations of the *X. fastidiosa* strains tested, as did soluble extracts derived from balsa. In addition, the soft nature of balsa wood enabled the development of a method for viable counting and direct comparison of biofilm and planktonic populations of *X. fastidiosa* grown under the same conditions.

***Pseudomonas chlororaphis* O6 *gacS* gene is involved in biofilm formation.** L. L. R. MARQUES (1), M. E. Olson (1), H. Ceri (1), Y. C. Kim (2), M. Spencer (3), and A. J. Anderson (3). (1) Biofilm Research Group, Univ. of Calgary, AB, Canada, T2K1N4; (2) Chonnam National University, Korea; (3) Utah State University, Logan, UT 84322-5305. Phytopathology 92:S51. Publication no. P-2002-0369-AMA.

The bacterium *Pseudomonas chlororaphis* O6 (PcO6) is an aggressive colonizer of plant roots under competitive soil conditions. Root colonization by PcO6 induces foliar resistance to *Pseudomonas syringae* pv. *tabaci* in tobacco. To understand the genes involved in root colonization, mutations were generated in O6 by Tn-5 insertion. One mutant was complemented in phenotype by the *gacS* gene. The *gacS* knock-out mutant was deficient in phenazine, acyl-homoserine lactones and extracellular protease production. The ability of wild type and mutant strains to form biofilms was evaluated *in vitro* using the MBEC device. Biofilm formation by the *gacS* mutant, as evaluated by colony counts and SEM, was greatly reduced, but it was restored by complementation with the wild type *gacS* gene. The results demonstrate that the regulatory *gacS* gene plays an important role in biofilm formation and structure in PcO6, which may influence its biocontrol capability.

The heat-shock gene *groEL* as a phylogenetic marker for *Xanthomonas* spp. L. L. R. MARQUES (1), Y. B. Rosato (2), and G. P. Manfio (3). (1) BRG, Univ. Calgary, AB, Canada, T2K 1N4; (2) CBMEG; (3) CPQBA, CP 6171, UNICAMP, SP, 13081-970, Brazil. Phytopathology 92:S51. Publication no. P-2002-0370-AMA.

The genus *Xanthomonas* was subjected to extensive taxonomic revision, resulting in a major reclassification that allocated these organisms into 20 genomic species. 16S rDNA sequence analysis has been investigated as a phylogenetic marker for *Xanthomonas* species, but it showed rather poor resolution due to the high sequence conservation of the gene in these organisms. There is still a need for alternative molecular markers for the differentiation and phylogenetic comparisons of *Xanthomonas* spp. In this study, RFLP analyses of PCR-amplified *groEL* gene fragments (1.0 kb) of 19 type strains of *Xanthomonas* allowed the differentiation of all strains, demonstrating its potential as a molecular marker for these organisms. Partial sequence analysis of *groEL* fragments (360-600 bp) of 15 species yielded similar clustering results as observed with 16S data, with improved resolution. In addition, polymorphism of *groEL* allowed the differentiation between species and subgroups of xanthomonads associated with important diseases in tomato and pepper.

The use of remote sensing to monitor efficacy of control of soilborne pathogens of strawberry. F. N. Martin. USDA-ARS, 1636 East Alisal St., Salinas, CA 93905. Phytopathology 92:S51. Publication no. P-2002-0371-AMA.

Strawberry is sensitive to a range of soilborne pathogens, some of which are lethal while others can significantly reduce yield without killing the plant. Nonlethal pathogens such as *Pythium*, *Cylindrocarpon*, and binucleate *Rhizoctonia* spp. will affect the plant by pruning back the roots and stunting growth, which will reduce subsequent yield. When grown in non- or poorly fumigated soil, strawberry canopy development is thinner and overall vigor is less than observed for plants grown in methyl bromide + chloropicrin fumigated soils. Remote sensing is being used in an effort to quantify this effect on plant growth on a field wide basis. A normalized distribution vegetation index (NDVI) is calculated from red/infrared data collected from aerial pictures and compared to plant growth data (plant biomass, leaf area index, canopy coverage) and NDVI calculated from reflectance data collected on the ground. The relationship between this data and yield at different times during the season will be discussed.

Survey for viruses of grapevine in Oregon and Washington. R. R. MARTIN (1), K. Eastwell (2), A. Wagner (3), I. E. Tzanetakis (1) and S. Lamprecht (1). (1) USDA-ARS-HCRIL, Corvallis, OR; (2) Washington State Univ. Prosser, WA; (3) WSDA, Olympia, WA and Oregon State Univ., Corvallis, OR. Phytopathology 92:S52. Publication no. P-2002-0372-AMA.

We surveyed grapevines for viruses including: *Rupestris stem pitting associated virus* (RpSP), and *Grapevine leafroll associated viruses -1, -2, -3* (GLRaV -1, -2, -3), tested by RT-PCR, and *Arabidopsis mosaic virus* (ArMV), *Grapevine fanleaf virus* (GFLV) and *Tomato ringspot virus* (ToRSV), tested by ELISA. In Washington, 1522 samples were collected at random to represent the total acreage of juice and wine grapes and tested for each of the viruses with the exception of GLRaV-1 and -2 for which only 420 samples were tested. In Oregon, 240 samples were collected at random for testing for each of these viruses. In addition, 3000 samples were collected from 40 vineyards in Oregon known to have high populations of *Xiphinema americanum* nematodes. These were tested for ArMV, ToRSV and GFLV. All samples from Oregon tested negative for ToRSV, ArMV and GFLV. However, 3.3%, 3.3%, 0.4% and 2.9% samples tested positive for RpSP, GLRaV-1, GLRaV-2 and GLRaV-3, respectively. In Washington, 4.6%, 2.6%, 0.2% and 6.5% tested positive for RpSP GLRaV -1, -2 and -3, respectively.

Rapid blight: A new disease of cool season turfgrasses. S. B. MARTIN (1), L. J. Stowell (2), W. D. Gelernter (2), and S. C. Alderman (3). (1) Pee Dee Res. & Ed. Ctr., Florence, SC 29506; (2) PACE Turfgrass Res. Inst., San Diego, CA 92109; (3) USDA ARS NFSPRC, Corvallis, OR 97331. Phytopathology 92:S52. Publication no. P-2002-0373-AMA.

A new disease of cool season turf was first diagnosed in California in 1995 from *Poa annua* putting greens. Since that time, the disease has been observed in nine states and in addition to *Poa annua*, has caused serious damage to large expanses of *Poa trivialis* and *Lolium perenne*. The disease has also been observed associated with salt-stressed *Agrostis palustris*. Symptoms include severe yellowing of turf and a water soaked appearance to the diseased foliage. Football-shaped structures (4-6 um x 10-12 um) similar in appearance to sporangia of chytridiomycete fungi can be observed inside leaf cells of symptomatic plants. All attempts to isolate a fungus from infected tissues have failed. However, the disease was reproduced in *Poa trivialis* seedlings grown in pots containing soil from a golf course green that contained diseased *Poa annua* plants. Preliminary field trial results indicate that the fungicides mancozeb, trifloxystrobin or pyraclostrobin provide some measure of prevention or control.

Analysis of the multiple copy host-selective toxin gene, *ToxB*, from *Pyrenophora tritici-repentis*. J. P. MARTINEZ, N. W. Oesch, and L. M. Ciuffetti. Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331. Phytopathology 92:S52. Publication no. P-2002-0374-AMA.

Race 5 isolates of *Pyrenophora tritici-repentis* are known to produce a 6.6 kDa host-selective toxin (Ptr ToxB) encoded by the multiple copy gene, *ToxB*. Southern blot data have shown there are at least six *ToxB* loci in race 5 isolates. A related gene *toxh* (whose function is unknown), has been found only in nonpathogenic race 4 isolates. To learn more about the *ToxB* loci, we cloned and characterized five of these loci and their flanking DNA sequences. All five *ToxB* loci have identical open reading frames (ORFs), thus only one form of Ptr ToxB appears to be produced. Analysis of the *ToxB* flanking regions revealed a truncated retrotransposon in the upstream region of all five loci. Three loci have 36-bp inverted repeats flanked by 6-bp direct repeats, which are hallmarks of Class II transposons. Truncated retrotransposon-like sequences were also found downstream of the *ToxB* ORF for two loci. The association of these sequences with the cloned *ToxB* loci indicate that transposable elements may be involved in duplication of some of these loci.

Molecular based methods for the detection of *Ralstonia solanacearum* (race3/biovar2) and for biovar differentiation. M. MARTINI (1), I.-M. Lee (1), and E. Stefani (2). (1) Molecular Plant Pathology Lab., USDA, ARS, Beltsville, MD 20705; (2) Dept. Patologia Vegetale, Univ. of Bologna, 40126 Bologna, Italy. Phytopathology 92:S52. Publication no. P-2002-0375-AMA.

Ralstonia solanacearum can be separated into 4 races and 5 biovars. *R. solanacearum* race3/biovar2 infects primarily potato, causing brown rot disease, and has never been reported to infect potato in the US. Brown rot disease is spread by latently infected seed potatoes; therefore a highly sensitive diagnostic method is required for quarantine purposes. In the present study, primer pairs designed from a novel repetitive insertion sequence were developed for PCR detection of *R. solanacearum* race3/biovar2. This PCR method provided a reliable and sensitive tool for the specific detection of all the *R. solanacearum* race3/biovar2 strains tested, including strains isolated from geranium in the US. Southern hybridization and rep-PCR analyses showed that the geranium strains are very closely related to the potato strains. A PCR/RFLP method based on the ribosomal protein operon was also developed for the molecular differentiation of 5 biovars, which are currently distinguished according to different biochemical properties.

Plant parasitic nematodes identified from North Dakota potato fields. P. A. MASON. North Dakota Department of Agriculture, North Dakota State University, Fargo, ND 58105. Phytopathology 92:S52. Publication no. P-2002-0376-AMA.

This survey was undertaken to support the phytosanitary certification of North Dakota grown potatoes intended for export. Field soil surveys were conducted from 1996 through 2001. A total of 859 fields in 23 North Dakota counties, including Benson, Burleigh, Cass, Cavalier, Emmons, Foster, Golden Valley, Grand Forks, Griggs, Kidder, McHenry, Morton, Pembina, Pierce, Ramsey, Ransom, Richland, Rolette, Sargent, Towner, Traill, Walsh, and Williams, were sampled. All fields surveyed had been in potatoes at least 3 of the past 10 years. Soil cores, collected to a depth of 15 cm, were bulked and washed through 20 and 60 mesh sieves for cysts, and a 325 mesh sieve for nematodes. No nematodes of regulatory importance were recovered; however, 8 genera of plant parasitic nematodes, including *Cactodera*, *Helicotylenchus*, *Hoplolaimus*, *Paratylenchus*, *Pratylenchus*, *Quinislucius*, *Tylenchorhynchus*, and *Xiphinema* were identified from these fields.

***Nodulisporium* sp. implicated in death of branches on lemon trees in Arizona.** M. E. MATHERON and M. Porchas. Yuma Agricultural Center, University of Arizona. Phytopathology 92:S52. Publication no. P-2002-0377-AMA.

A white wood rot has been associated with dieback of branches on lemon trees during the summer in Yuma, AZ. A *Nodulisporium* sp. was isolated from decayed wood tissue. Two other wood decay fungi, *Antrrodia sinuosa* and *Coniophora eremophila*, cause a brown heartwood rot on lemon trees in Arizona. To compare the rate of wood decay caused by the three different fungi, Lisbon lemon trees were inoculated in July by placing a small wooden dowel (5-mm-diam. x 13-mm-long) colonized by one of the pathogens into a similar-sized hole drilled into healthy branches. Six months after inoculation, the mean length of wood decay columns in branches inoculated with *A. sinuosa*, *C. eremophila* and *Nodulisporium* sp. was 159, 45 and 148 mm, respectively. In another study, the mean length of wood decay columns 6 months after inoculation of branches on Lisbon lemon, Marsh grapefruit, Valencia orange and Orlando tangelo trees with *Nodulisporium* sp. was 233, 113, 116 and 70 mm, respectively. Wood decay columns produced by *Nodulisporium* sp. on Lisbon lemon branches from late spring to early autumn were at least four times longer than those that developed from late autumn to early spring.

Bacterial pathogens on cereals in the Russian Federation. E. V. Matveeva (1), V. A. Polityko (1), A. N. Ignatov (2), E. V. NIKOLAEVA (1), E. Sh Pekhtereva (2), and N. W. Schaad (3). (1) Russian Research Institute of Phytopathology, Vyasemy, Moscow; (2) Centre "Bioengineering" RAS, Moscow; (3) USDA ARS Foreign Disease-Weed Science Research Unit, Fort Detrick, MD. Phytopathology 92:S52. Publication no. P-2002-0378-AMA.

Little is known about bacterial pathogens of cereals in Russia. To learn more about cereal bacteria, a survey of cereal crops in Russia was conducted in 2001. Over 300 samples of cereal plants with bacterial disease symptoms were collected for characterization of phenotypic and molecular traits and pathogenicity. Results revealed widespread occurrence of bacterial diseases of wheat, barley, and rye in different climatic zones of Russia. Basal glume rot of wheat (*Pseudomonas atrofaciens*) and black-chaff of wheat, barley,

and rye (*Xanthomonas translucens*) were most common. Bacterial mosaic of wheat (*Clavibacter michiganense* subsp. *tessellarius*) was observed less frequently.

Evaluation of inoculation methods for *Aspergillus* ear rot and aflatoxin contamination of corn. L. M. MAUPIN (1), D. G. White (1), and J. M. Perkins (2). (1) Dept. of Crop Sciences, University of Illinois, Urbana, IL 61801; (2) Monsanto Company, Waterman, IL 60556. *Phytopathology* 92:S53. Publication no. P-2002-0379-AMA.

Our goal was to evaluate an inoculation technique to be used for *Aspergillus* ear rot. Inoculum was made by mixing 8 kg of corn with sporulating *Aspergillus flavus* growing on it with 600 ml of cottonseed oil to suspend conidia in the oil. Four kg of the oil and infested corn mixture was then added to 20 kg autoclaved corn and mixed with 350 gm diatomaceous earth to adhere the oil and conidia onto the corn kernels. Two inoculation methods and a control were evaluated in three replicates using 12 hybrids in four locations in Texas (central, upper gulf, lower gulf and deep south) in 2001. The inoculation methods were placing 5 g inoculum in the whorl of each plant and 700 g inoculum per 25 m row on the soil surface in the row at the V9-V10 growth stage. Over all locations and all hybrids aflatoxin levels for the whorl inoculation (395ng/g) were significantly higher ($P = 0.05$) than soil surface inoculation (249ng/g) or noninoculated (94ng/g).

Genetic basis for the unique root-colonizing activity of *Pseudomonas fluorescens* Q8r1-96. O. V. MAVRODI (1), D. V. Mavrodi (1,2), D. M. Weller (1,2), and L. S. Thomashow (1,2). (1) Dept. Plant Pathology, Washington State University, Pullman, WA 99164-6430; (2) USDA Department of Agriculture, Agricultural Research Service, Root Disease and Biological Control Research Unit, Washington State University, Pullman, WA 99164-6430. *Phytopathology* 92:S53. Publication no. P-2002-0380-AMA.

Fluorescent *Pseudomonas* spp. that produce 2,4-diacetylphloroglucinol (DAPG) have biocontrol activity against diseases caused by *Gaeumannomyces graminis* var. *tritici*. Certain DAPG producers colonize roots and suppress soilborne pathogens more effectively than related DAPG producers from which they are otherwise phenotypically very similar. "Premier" strain Q8r1-96 is able to establish and maintain a large population size and to aggressively colonize both wheat and pea. To determine the genetic basis underlying the unique competitiveness of this strain we have constructed an ordered library of Q8r1-96 genomic DNA which consists of 1,536 clones with inserts averaging 30 kb in size. Genes potentially responsible for the premier phenotype of strain Q8r1-96 have been identified by screening the library and are being evaluated for their role in root colonization.

Sensitive, high-throughput, real-time PCR for field diagnosis of citrus canker. V. MAVRODIEVA (1), L. Levy (1), and D. Gabriel (2). (1) USDA-APHIS, NPGQC, Beltsville, MD 20705; (2) Plant Pathology Dept., University of Florida, Gainesville, FL 32611. *Phytopathology* 92:S53. Publication no. P-2002-0381-AMA.

Citrus canker disease is one of the most damaging diseases of citrus and subject to state and federal quarantines and under eradication in Florida. Accurate, fast and reliable detection and diagnosis of the causal agent *Xanthomonas citri* is of great regulatory importance. However, citrus bacterial canker is caused by at least three phylogenetically distinct groups of strains. We developed a high-throughput, real-time PCR assay using a portable field-hardened R.A.P.I.D. system (Idaho Technology) and primers designed to detect all canker causing strains based on the presence of gene *pthA*. Sampling required minimal handling; no DNA extractions and results were obtained in ca. four hours. The following parameters, listed in order of declining importance, were judged to affect sensitivity/reproducibility: primer design, Chelex resin, buffered saline, CaCO₃ and Silwet L77. The above system routinely detected ca. 10 cfu.

The mitochondrial DNA haplotype of *Phytophthora infestans* in 19th century herbarium specimens revealed. K. J. MAY and J. B. Ristaino. North Carolina State University. *Phytopathology* 92:S53. Publication no. P-2002-0382-AMA.

PCR was used to determine the mtDNA haplotype present in 19th and 20th century herbarium specimens infected with *P. infestans*. DNA was extracted from lesions from specimens (n=187) from 7 herbaria. A 100 bp fragment of rDNA was successfully amplified using the PINF/HERB1 primers from 87% of the samples indicating that the leaves were infected with *P. infestans*. Mitochondrial DNA (mtDNA) primers P2F4/R4 amplify a variable *Msp I* site that distinguishes Ib haplotypes from Ia, IIa, and IIb haplotypes. A PCR product was amplified for 70 specimens and sequence analysis indicated that

only 1 specimen from Ecuador was the Ib haplotype. Primers P3F1/R1 and P3F2/R3 amplify a variable region that contains an *Eco RI* site that distinguishes type I from type II mtDNA haplotypes. PCR product using the P3 primers was amplified for 50 and 70 specimens, respectively. The sequence and *EcoRI* digestion data from the P3 region indicates that 47 specimens were the Ia haplotype. Some of the oldest samples infected with *P. infestans* from the US, Britain, Ireland and France were the Ia haplotype and this is the predominant haplotype.

Wheat genotype-specific induction of soil suppressiveness to *Rhizoctonia solani*. M. MAZZOLA and Y.-H. Gu. USDA-ARS, Wenatchee, WA 98801. *Phytopathology* 92:S53. Publication no. P-2002-0383-AMA.

The role of host genotype in eliciting essential transformations in microbial community structure that lead to induction of disease suppressive soils has not been fully recognized. Although wheat cultivars examined possessed uniform susceptibility to *R. solani* AG 8, three successive plantings of specific wheat genotypes induced soil suppressiveness to *Rhizoctonia* root rot of apple caused by an introduced isolate of *R. solani* AG 5 as well as to the wheat pathogen *R. solani* AG 8. Wheat genotypes that induced disease suppression enhanced populations of specific fluorescent pseudomonad genotypes with antagonistic activity toward *R. solani* AG 5 and AG 8, but wheat genotypes that did not elicit a disease suppressive soil did not alter composition of this bacterial community. The same cultivar specific response in terms of transformation of the fluorescent pseudomonad community was obtained through application of wheat root exudates to soil. These results demonstrate the importance of host genotype in induction of soil suppressiveness through modification of the saprophytic soil microflora, and suggest an important role for host genotype in the success of biological control.

Management of late blight tuber rot of potatoes with zoxamide fungicide. A. G. MCFADDEN, A. E. Duttler, R. L. Smith, G. M. Kemmitt, B. D. Olson, J. Edmonds, and D. H. Young. Dow AgroSciences, 9330 Zionsville Road, Indianapolis, IN 46268. *Phytopathology* 92:S53. Publication no. P-2002-0384-AMA.

Infection of potato tubers by the late blight fungus (*Phytophthora infestans*) occurs from spores that develop on the plant foliage. Gavel (TM) fungicide contains a new active ingredient, zoxamide, plus mancozeb in a 1:8 ratio. This fungicide has been evaluated in Europe and North America for the control of late blight. Along with excellent control of foliar symptoms, significant reductions in the level of tuber rot have been observed. Laboratory data suggest the suppression of tuber rot may be related to the effect of zoxamide on reduced production of viable motile zoospores. Gavel provides a key new tool for the management of late blight tuber rot of potatoes.

Efforts to develop *Agrobacterium*-mediated germ-line transformation of soybean. M. A. MCGILL (1), S. Clough (2), C. Desfeux (2), K. Schroeder (1), and A. F. Bent (1,2). (1) Dept. of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706; (2) Dept. of Crop Sciences, University of Illinois, Urbana, IL 61801. *Phytopathology* 92:S53. Publication no. P-2002-0385-AMA.

Floral-dip transformation has dramatically changed and accelerated progress in Arabidopsis research, and it would be advantageous if the method was available for crop species such as soybean. Extensive efforts to adapt our findings regarding Arabidopsis transformation mechanisms to soybean have met with little success. We have explored *Agrobacterium* strain - soybean cultivar combinations known to perform well in tissue culture-based transformation, as well as inoculation methods and plant growth conditions that could optimize transformation of germ-line tissues. We observe frequent flower and seed abortion, and hypothesize that soybean defense responses inhibit transformation. We are presently exploring methods to disarm these defenses. We are also documenting *Agrobacterium* location and persistence on host tissues. These assays will help to identify the limiting step(s) in soybean floral dip transformation.

Differential growth on veinal and non-veinal sites by *Aureobasidium pullulans* on apple leaves. M. J. MCGRATH, R. N. Spear, and J. A. Andrews. Dept. of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706. *Phytopathology* 92:S53. Publication no. P-2002-0386-AMA.

Our field data show that *Aureobasidium pullulans* (Ap) populations occur more commonly and at higher densities over veinal regions of the apple phylloplane. To determine the reason, leaf disks from growth chamber-grown

apple seedlings were inoculated with single GFP-tagged *Ap* blastospores and growth was followed over time. Disks were placed inside micro-humidity chambers mounted on slides where they remained fixed in position. For observations of cells by epifluorescence microscopy, chambers were transferred periodically to the microscope, where individual inoculation sites were relocated using the X-Y stage coordinates. Preliminary data indicate that the mean population sizes along the mid-vein, other major veins and non-veinal regions were 99 plus or minus (+/-) 40, 79 +/-51 and 10 +/-6 cells, respectively, after 6 days. Veinal colonies tended to expand longitudinally, remaining on the veins. Colonies on non-veinal sites usually expanded radially. Evidently, factors on veinal sites support higher per capita *Ap* growth and/or lower death rates.

Web blight caused by *Rhizoctonia solani* on New Guinea impatiens. R. T. MCMILLAN, JR., S. V. Johnson, and W. R. Graves. TREC, University of Florida. Phytopathology 92:S54. Publication no. P-2002-0387-AMA.

An outbreak of a foliar disease on New Guinea impatiens (*Impatiens hawkeri* Bull) was observed in a nursery in Dade County, FL. Symptoms on leaves were water-soaked spots that increased rapidly in size and became light to dark brown necrotic areas. A *Rhizoctonia*-like fungus was isolated from infected leaves and stems on corn meal agar subcultured on potato-dextrose agar and identified as *Thanatephorus cucumeris* (Frank) Donk (anamorph *Rhizoctonia solani* Kuehn). Six plants were randomly inoculated with agar blocks containing the isolate. Control plants received agar blocks without the pathogen. Plants were placed in a modified humidity chamber (polyethylene bags) in a greenhouse at 27°C for three days. The symptoms appeared as water soaked spots 10mm in diameter that enlarged to 25mm or more and turned dark brown. A mycelium web grew over the leaves, killing them, and spreading from leaf to leaf. Small brown sclerotia and mycelium were found on leaves and stems, typical of that found on nursery plants. *Thanatephorus cucumeris* was consistently re-isolated from inoculated plants with no symptoms observed on uninoculated plants, fulfilling Koch's postulates.

Rapid, realtime PCR with fully integrated specimen preparation. W. A. McMillan. Cepheid, Sunnyvale, CA 94089. Phytopathology 92:S54. Publication no. P-2002-0388-AMA.

The Cepheid I-CORE thermally-controlled fluorometer module has true 4-color detection capability and is integrated into both the Smart Cycler systems, and the GeneXpert, a system that processes cartridges which integrate sample preparation and PCR. PCR analysis time is 25 minutes or less. The GeneXpert system is a benchtop 4-site instrument that processes 100 µl to 5 mL volume specimens. Any bacteria present are concentrated down to 40 µL, washed, rapidly lysed using a novel ultrasonics technique, mixed with on-board dried-down PCR reagent beads, and transferred to an integrated PCR tube. Sample preparation typically requires 5 minutes or less. Demonstrated applications include anthrax spores from wet aerosols, Group B *Streptococcus* from swab extracts, and *Mycobacteria tuberculosis* in buffer, and total time for processing is 30 minutes or less. Advanced technology development is focusing on increasing the optical channels to 8-10 colors and on-board DNA or RNA purification.

Antibiotic-producing *Pseudomonas* spp. in Ohio soils: Potential for root disease suppression. B. B. MCSPADDEN GARDENER and E. Lutton. Dept. of Plant Pathology, The Ohio State University-OARDC, Wooster, OH 44691. Phytopathology 92:S54. Publication no. P-2002-0389-AMA.

Microbial antagonism of soilborne plant pathogens has been shown to be mediated by antibiotics produced by various taxa. *Pseudomonas* spp. that produce antibiotics such as 2,4-diacetylphloroglucinol (DAPG) have been identified as effective biological control agents of diverse root pathogens, and they are believed to contribute substantially to natural soil suppressiveness. To further elucidate the microbial ecology of these functionally important bacteria, we characterized the abundance and diversity of antibiotic-producing pseudomonads on corn and soybean plants grown throughout Ohio. DAPG producers were found at 93% of the field sites examined in 2001. While over 90% of the corn plants examined were colonized, less than 50% of the soybean plants were colonized by pHLD+ bacteria. In contrast, pyoluteorin- and pyrrolnitrin-producing pseudomonads were detected on < 20% of plants tested. Four distinct genotypes were detected, including two newly discovered genotypes. Isolates of the dominant genotypes inhibited the growth of several root pathogens *in vitro*, indicating their potential for contributing to soil suppressiveness in corn and soybean cropping systems.

cificity, population dynamics, and biocontrol of take-all by DAPG-producing *Pseudomonas* spp. in a long-term wheat field. B. B. McSpadden Gardener (1,2) and D. M. WELLER (2). (1) Dept. of Plant

Pathology, The Ohio State University-OARDC, Wooster, OH 44691; (2) USDA-ARS Root Disease and Biological Control Research Unit, Washington State University, Pullman, WA 99164. Phytopathology 92:S54. Publication no. P-2002-0390-AMA.

Pseudomonas spp. that produce the antibiotic 2,4-diacetylphloroglucinol (DAPG) are key contributors to take-all suppression in long-term wheat monoculture soils of WA state. Here we report on the population dynamics of indigenous DAPG-producing populations colonizing the rhizosphere of four different field crops (wheat, peas, oats, and barley) grown at Lind, WA. Two genotypes, D and L, were detected at the site and found to differ in their colonization and biocontrol potential. Both genotypes were found in the rhizosphere of wheat and pea but only the L genotype colonized oats and barley. The biological control properties of each genotype were assessed *in vitro* and in the field. In each setting, the D genotype displayed a superior biocontrol capacity. We conclude that certain crop rotations may select for more active biocontrol populations, and that indigenous populations of the D genotype at this site are constrained by some unknown environmental factors.

Sudden oak death surveys in Oregon - 2001. M. G. McWilliams (1), A. Kanaskie (1), N. Osterbauer (2), E. M. Goheen (3), E. M. HANSEN (4), W. Sutton (4), and J. Mair (1). (1) Oregon Dept. Forestry; (2) Oregon Dept. Agriculture; (3) U.S. Forest Service; (4) Oregon State University. Phytopathology 92:S54. Publication no. P-2002-0391-AMA.

Two aerial surveys in SW Oregon, in July and October 2001, detected 33 sites with recently killed tanoak trees. Eight more sites with dead tanoaks were identified during ground checking. Of these 41 sites, 9 were positive for *Phytophthora ramorum*. The infested sites all are within a 9-square mile area in Curry County, and range in size from 0.5 to 12 acres. They occur on industrial and non-industrial forest land and federal Bureau of Land Management lands. The primary species killed was tanoak, but rhododendron and evergreen huckleberry also are infected and occasionally killed. Other causes of tanoak mortality included *Armillaria* species (9 sites), herbicide or mechanical damage (7 sites), an unidentified *Phytophthora* (5 sites), and unknown agents (11 sites). *Armillaria* sp. was the most frequently encountered pathogen associated with single trees or small clumps of 2 or 3 dead tanoaks, while larger groups of dead tanoak were more likely to be SOD. Ground surveys not associated with the aerial surveys failed to detect Sudden Oak Death.

Field evaluation of tomato experimental lines and hybrids for resistance to begomoviruses in Guatemala. L. MEJIA, R. E. Teni (1), H. Czosnek, F. Vidavski (2), A. Bettilyon, M. K. Nakhla, and D. P. Maxwell (3). (1) Universidad de San Carlos, Guatemala; (2) The Hebrew University of Jerusalem, Israel; (3) University of Wisconsin-Madison 53706. Phytopathology 92:S54. Publication no. P-2002-0392-AMA.

The incidence of begomovirus infection of tomatoes in Guatemala is commonly 100%. Tomato lines were evaluated in an area where four begomoviruses occur. Resistant F6 lines, F6-2211 and F6-5221, were selected from the F1 hybrid, FAVI 9 (Phytopathology 88:910), with resistance genes for *Tomato yellow leaf curl virus* (TYLCV) from *L. hirsutum*. Resistant F6 line P2-1132 was selected from a segregating line with resistance genes to TYLCV from *L. pimpinellifolium* and *L. peruvianum* (H. Laterrot, pers. com.). HC7880 was selected in Cuba by O. Gomez, and it is high yielding line but susceptible to begomoviruses. The experimental hybrids H1 (F6-2211 x HC7880) and H2 (F6-5221 x HC7880) had mild viral symptoms, which indicated that the resistance gene(s) are dominant. These hybrids yielded about three times the susceptible cv. Marina. The experimental hybrid H3 (F6-5221 x P2-1132) had the least viral symptoms, and thus, the highest level of resistance. H1 and H2 will be evaluated in commercial fields.

Summary of efficacy tests on Agry-Gent™, a new agricultural antibiotic formulation for control of fire blight in apple and pear. G. L. MELCHIOR and P. J. David. Gowan Company. Phytopathology 92:S54. Publication no. P-2002-0393-AMA.

Agry-Gent is a 10% WP formulation of gentamicin sulphate, an aminoglycoside antibiotic active to important genera of phytopathogenic bacteria. Gentamicin has registered commercial use outside the United States against bacterial diseases in pear, potato, tomato, and pepper. It is highly active to *Erwinia amylovora*, the causal agent of the most serious and economically damaging bacterial disease of pome fruit in North America, fire blight. Studies were initiated with Agry-Gent in 1997 in eastern and western fruit growing regions of the U.S. to determine commercial utility, with emphasis on streptomycin-resistant *E. amylovora*. In general, suspensions of Agry-

Gent plus surfactant were applied in dilute or full coverage sprays to apple or pear blossoms, then challenged with inoculations of either streptomycin-susceptible or -resistant isolates of *E. amylovora*. Data were collected on bacterial cell survival, blossom infection, or infection of new shoots. Composite results indicate equivalent efficacy to streptomycin susceptible and resistant *E. amylovora* strains, and excellent commercial potential.

Use of bentamicin to control fire blight in apples: Need, efficacy, and human health implications. G. L. MELCHIOR (1), P. J. David (1), V. W. Winkler (1), and R. A. Spitko (2). (1) Gowan Company; (2) New England Fruit Consultants. *Phytopathology* 92:S55. Publication no. P-2002-0394-AMA.

Fire blight, caused by *Erwinia amylovora*, is the most serious bacterial disease of pome fruit. Disease pressure is now at catastrophic proportions due to market forces demanding highly susceptible cultivars and horticultural practices favoring disease development worldwide. Selection of streptomycin-resistant *Erwinia amylovora* populations due to lack of effective alternative control measures is increasing this risk. Gowan Company is developing the commercial use of gentamicin to control fire blight in apples. Results indicate equivalent efficacy in streptomycin-susceptible and -resistant *Erwinia amylovora* strains, and excellent potential for commercial application. An analysis of risks associated with 1) occupational exposure to gentamicin, 2) dietary exposure to gentamicin, and 3) potential dietary exposure to gentamicin-resistant bacteria show that the proposed use meets the reasonable certainty of no harm for FFDC, and exceeds the bar of risk versus benefit requirements of FIFRA.

Effect of post inoculation relative humidity on reaction of peanut to *Sclerotinia minor*. H. A. MELOUK (1) and K. E. Jackson (2). (1) USDA-ARS, PSWCRL, Oklahoma State University; (2) Oklahoma State University. *Phytopathology* 92:S55. Publication no. P-2002-0395-AMA.

Detached shoots (DS) from 8-wk-old plants of 'Okrun' (OK), and 'Southwest runner' (SW), a *Sclerotinia*-susceptible and -resistant peanut cultivars, respectively, were inoculated with *Sclerotinia minor* (Peanut Sci. 19:58-62). At 24 hours post inoculation (PI), one-half of the DS were subjected to a lower relative humidity (RH), and the other half remained at 100% RH. Lesion length (LL) was measured daily from 2 to 6 days PI. Disease incidence (DI) under the 100% RH for OK and SW was 96 and 98%, respectively. DI in both genotypes was reduced by about 40% under the low RH regime. LL under 100% RH at day 6 PI for SW and OK was 47 and 64mm, respectively. Reduction of LL at day 6 under low RH was 66 and 76% for OK and SW, respectively. Correlation coefficient between LL at day 6 PI and area under disease progress curve was 0.96 ($P = 0.0001$). These data support field observations that occurrence of low RH immediately following onset of *Sclerotinia* blight infection results in a lower disease severity.

Quantifying the acquisition and transmission periods of the corn flea beetle for effective management of Stewart's disease. B. MENELAS and F. W. Nutter, Jr. Iowa State University. *Phytopathology* 92:S55. Publication no. P-2002-0396-AMA.

The corn flea beetle (CFB) *Chaetocnema pulicaria*, is the primary vector of *Erwinia stewartii*, yet little is known about acquisition and transmission of *E. stewartii* by the CFB. Field-collected CFBs were allowed to feed for 12, 24, 36, 48, and 72 hours on corn seedlings inoculated with a rifampicin-resistant strain of *E. stewartii* (isolate ES Rif-9A) to determine acquisition time. The beetles were then ground in 300 microliters of PBST buffer and *E. stewartii* was isolated on nutrient broth yeast extract agar amended with cycloheximide, rifampicin, and nalidixic acid (NBY-CRN), indicating that acquisition of the bacterium had occurred. Preliminary results indicated that acquisition was highest when beetles fed for 48 hours. These time periods are being evaluated in subsequent experiments to determine the time required for CFB to transmit the bacterium to healthy plants. Information gained from this study will be used to more effectively manage the CFB and Stewart's disease of corn.

Population genetics of the carrot blight pathogens, *Alternaria dauci* and *Xanthomonas campestris* pv. *carotae*. X. MENG, R. L. Gilbertson, R. M. Davis, and J. Nunez. Dept. Plant Pathology, University of California, Davis, CA 95616. *Phytopathology* 92:S55. Publication no. P-2002-0397-AMA.

Knowledge of genetic diversity in *Alternaria dauci* and *Xanthomonas campestris* pv. *carotae* (Xcc) causing carrot blight can be used to identify sources of inoculum. PCR amplification of the intergenic spacer (IGS) regions of *A. dauci* isolates revealed fragment length polymorphisms. IGS regions with the greatest size polymorphisms were cloned and sequenced,

and a *A. dauci* IGS primer pair was designed. Using this primer pair and rapid sonication of mycelia and conidia, IGS fragment length polymorphisms among isolates were revealed and evaluated as a tool to study population genetics of *A. dauci*. Genetic diversity among Xcc strains was evaluated by rep-PCR with BOX, REP and ERIC primers. Xcc strains were divided into two genotypes by BOX- and REP-PCR, and into three genotypes by ERIC-PCR. Using a combination of Xcc-specific PCR and rep-PCR, Xcc genotypes from California, Oregon and Washington have been determined. Results of these analyses are being used to gain insight into Xcc inoculum sources.

Control of postharvest diseases of apple by the volatile-producing fungus *Muscodor albus*. J. MERCIER and D. Manker. AgraQuest Inc., 1530 Drew Avenue, Davis, CA 95616. *Phytopathology* 92:S55. Publication no. P-2002-0398-AMA.

Fumigation for controlling blue mold and gray mold of apple was attempted using *Muscodor albus*, a fungus which produces a number of antimicrobial volatiles. Volatiles produced by *M. albus* on PDA plates inhibited and killed *Penicillium expansum* and *Botrytis cinerea*. To test for disease control, inoculated apples, cv. Gala, were fumigated by placing them in closed plastic boxes containing autoclaved rye grain colonized by *M. albus*. There was no physical contact between the fruits and *M. albus*. The fruits were wound-inoculated with *P. expansum* or *B. cinerea* conidia (10^4 /ml), either 24 h or immediately before exposure to *M. albus* volatiles and incubated at room temperature for 7 days, after which the colonized grain was removed. Blue mold and gray mold were completely inhibited by the volatiles, even when inoculation took place 24 h before treatment. Very few fruits developed infections after the *M. albus* preparation was removed. Fumigation with *M. albus*, a sterile mycelium, could be an attractive approach for the control of storage diseases.

Non-specific yield reductions caused by *Colletotrichum acutatum* and *C. gloeosporioides* on strawberry. J. C. MERTELY, S. J. Mackenzie, C. K. Chandler, and D. E. Legard. Univ. of Florida, GCREC-Dover, FL 33527. *Phytopathology* 92:S55. Publication no. P-2002-0399-AMA.

To investigate yield reductions caused by *Colletotrichum* spp. on strawberry, cultivars Camarosa and Sweet Charlie were grown in 14-plant plots during the 2000-01 and 2001-02 seasons. Plants were inoculated by dipping roots at planting or spraying crowns after establishment with 1×10^6 conidia/ml of *C. acutatum* (isolate 98-324) or *C. gloeosporioides* (98-264). Each treatment was replicated four times in a split-plot design with cultivar main plots and inoculation method/isolate subplots. *C. acutatum* reduced marketable yields 10-64% in 'Camarosa' and 8-38% in 'Sweet Charlie'. Inoculated plants of both cultivars were stunted, yet remained alive. Root dip inoculations typically caused higher yield loss than sprayed crowns. *C. gloeosporioides* reduced yields 26% in 'Camarosa' and 14% in Sweet Charlie in 2000-01. Plant mortality accounted for most of the yield decline in Camarosa, but not in 'Sweet Charlie'. The incidence of anthracnose fruit rot was low (<5%) in both experiments due to weekly applications of Captan.

Testing biological control and induced systemic resistance for the control of Aphanomyces root rot of sugarbeet. M. S. METZGER (1) and J. J. Weiland (1,2). (1) Department of Plant Pathology, North Dakota State University, Fargo, ND 58105; (2) USDA-Agricultural Research Service, Red River Valley Agricultural Research Center, Fargo, ND 58105. *Phytopathology* 92:S55. Publication no. P-2002-0400-AMA.

Seedling damping off and chronic root rot of sugarbeet caused by *Aphanomyces cochlioides* has caused increasing losses to U.S. producers. Lack of effective control measures for Aphanomyces root rot prompted the initiation of a program aimed at the discovery of new, safe components for disease control. A biological control bacterium and a known inducer of systemic resistance were tested for their ability to control Aphanomyces root rot at two locations in the Red River Valley of the north central U.S. during the 2001 growing season. At both field locations, sugarbeet yield was increased where seed was treated with the bacterium *Burkholderia cepacia* AMMDR1. At one location, treatment with formulated harpin protein (Messenger™) also resulted in increased sugarbeet yield. Future testing will aid in determining new approaches to be implemented alone or in conjunction with current disease control measures to reduce losses caused by this serious pathogen of sugarbeet.

Detection of root-associated microbes that produce compounds active against plant-parasitic nematodes. S. L. F. MEYER (1), D. P. Roberts (2), J. K. Nitao (1), and D. J. Chitwood (1). BARC-West, 10300 Baltimore Ave., Beltsville, MD 20705, (1) USDA, ARS Nematology Laboratory; (2) USDA, ARS Sustainable Agriculture Systems Laboratory. *Phytopathology* 92:S55. Publication no. P-2002-0401-AMA.

Rhizosphere-inhabiting bacteria and fungi isolated from soil and plant roots, and known to be active against plant-pathogenic fungi, were assayed in vitro for production of compounds antagonistic to root-knot nematode (*Meloidogyne incognita*). In addition, culture filtrates of fungi isolated from eggs of soybean cyst nematode (*Heterodera glycines*) were tested for activity against *M. incognita* and *H. glycines*. Several culture filtrates from the microbes isolated from soil and plant roots inhibited *M. incognita* egg hatch and/or mobility of second-stage juveniles. Filtrates of ca. 10% of the fungi collected from *H. glycines* suppressed egg hatch of the source nematode by more than 60% or inhibited juvenile mobility, and ca. 19% similarly affected *M. incognita*. Identification of agents with potential to act against plant-pathogenic fungi and plant-parasitic nematodes may be useful for suppression of multiple plant diseases.

Dactylella pseudoclavata, a new nematode-trapping fungus from China. Z. Q. Miao (1), S. D. Li (1), M. X. He (1), and X. Z. LIU (2). (1) Biocontrol Inst., Chinese Acad. of Agri. Sci., Beijing 100081, China; (2) Inst. of Microbiol., Chinese Acad. of Sci., Beijing 100080, China. Phytopathology 92:S56. Publication no. P-2002-0402-AMA.

While surveying parasite of *Meloidogyne incognita* in China, a new hyphomycete, *D. pseudoclavata* Miao & Liu, was isolated from greenhouse soil in Beijing suburb and described. Colonies of the fungus on CMA are effuse, slightly brown. Mycelium composed of hyaline, septate, branched, prostrate hyphae. Conidiophores erect, simple, occasionally branched, hyaline, septate, 150-300 µm long, 6-7.5 µm wide at the base, tapering to 4-5.5 µm at apex. Conidiogenous cell produces holoblastically 1-4 conidia at apex. Conidia ob-clavate, round to blunt in distal end, constricted into bottleneck in basal part, 0-1 septate, constricted at septum. Chlamydospores yellowish to brown, globose or subglobose, warted on surface, abundant in aged culture. Trapping nematode by adhesive networks. The fungus closely resembles *D. arcuata*, *D. clavata*, *D. crassa*, *D. cylindrospora*, *D. huisuniana* and *D. submersa*, but can be distinguished from them by morphology of conidiophore, conidium and predatory device.

Isolation of fungi from unprocessed Pinus radiata chips exported from Chile to the United States. J. A. MICALES and H. H. Burdsall, Jr. USDA-FS, Forest Products Laboratory, One Gifford Pinchot Dr., Madison, WI 53705. Phytopathology 92:S56. Publication no. P-2002-0403-AMA.

International trade in unprocessed wood chips for the pulp and paper industry is a potential source for the introduction of non-native wood-inhabiting pests and pathogens that could pose a severe hazard to U.S. forestry resources. The Animal and Plant Health Inspection Service allowed the importation of a small quantity of unprocessed *Pinus radiata* chips from Chile to an isolated processing plant on the pier at Bellingham, WA to evaluate the danger of introducing non-native fungal pathogens. Sixteen different shipments were sampled. Six fungal genera were consistently recovered and represented nearly 90% of the isolates. These genera included *Geotrichum*, *Gloeocladium*, *Paecilomyces*, *Penicillium*, *Phanerochaete*, and *Trichoderma*. The *Trichoderma* species accounted for nearly half of the total species isolated. This number would have been even higher if the specimens had not been initially surface disinfested. Isolates of *Graphium*, potential bluestain or vascular wilt pathogens, were recovered from only 0.32% of specimens. Species of *Trichoderma* appear to be acting as biological control agents in wood chip shipments.

Ability of an ELISA-based seed health test to detect Erwinia stewartii in maize seed treated with fungicides and insecticides. P. M. MICHENER (1), J. K. Pataky (1), C. C. Block (2), L. M. Shepherd (3), and D. C. McGee (3). (1) Univ. of Illinois, Urbana, IL; (2) USDA-ARS NCRPIS; (3) Seed Science Center, Iowa State Univ., Ames, IA. Phytopathology 92:S56. Publication no. P-2002-0404-AMA.

An ELISA-based seed health test detected *E. stewartii* in infected maize seed treated with 11 combinations of captan, carboxin, clothianidin, imidacloprid, thiamethoxam, fludioxonil, mfenoxam, or thiram. Treated, healthy seeds were blended with treated, *E. stewartii*-infected seeds of Jubilee or A632 to produce 100-kernel samples with a 96% or 88% chance, respectively, that a randomly-selected sample would contain infected kernels. Forty-six samples were tested per seed treatment and a non-treated control. Based on Type I errors near 5%, >20 of 23 samples per treatment were expected to be positive for samples with Jubilee, and >17 of 23 samples per treatment were expected to be positive for samples with A632. For Jubilee, all treatments had >20 positive samples. For A632, all treatments had >17 positive samples except for seed treated with clothianidin or captan/carboxin which had 17 and 15 positive samples, respectively. Mean absorbance values of positive samples did not differ among seed treatments.

Effects of soil temperature fluctuations on sclerotia germination and apothecia production of Sclerotinia sclerotiorum, causal agent of soybean stem rot. A. L. MILA and X. B. Yang. Dept. of Plant Pathology, Iowa State Univ., Ames, IA 50011. Phytopathology 92:S56. Publication no. P-2002-0405-AMA.

It has been observed that apothecia production of *Sclerotinia sclerotiorum* differs among different soil depths and tillage systems, where observed soil temperature has similar daily mean but differs in levels of fluctuation. Possible effect of temperature fluctuations on sclerotia germination and apothecia production was examined in five soil temperature fluctuation treatments using natural sclerotia collected from a field in Ackley, IA: constant 20°C, 18-22°C, 16-24°C, 14-26°C, and 12-28°C. Daily mean was 20°C for all treatments. Our results showed that the number of germinated sclerotia and apothecia production were significantly reduced under the high temperature fluctuation treatments. Significant differences were also found in the proportion of fungi isolated from the sclerotia surface in the different treatments. *Fusarium lateritium*, *Trichoderma* spp., *Gliocladium* sp. are some of the species recovered from the sclerotia.

The effect of soil pH on soybean cyst nematode reproduction. D. R. MILLER and S. Y. Chen. University of Minnesota, Southern Research and Outreach Center. Phytopathology 92:S56. Publication no. P-2002-0406-AMA.

pH has been previously reported to influence reproduction of the soybean cyst nematode, *Heterodera glycines*. During the 2001 growing season, we detected a strong positive correlation between soil pH and *H. glycines* egg population density in a field located in Waseca County, Minnesota. This study was conducted to confirm the pH/*H. glycines* relationship under greenhouse conditions. Soils representing pH levels of 5.5, 6.4, 7.3, and 8.1 were collected from the field and treated with microwave heating to kill existing *H. glycines* eggs. The soils were inoculated with approximately 2,000 eggs/100 cm³ soil at planting and 1,500 eggs/100 cm³ soil at 21 days after planting. The susceptible cultivar Sturdy was used to promote *H. glycines* reproduction. After 62 days, the soils were harvested and processed to determine final egg population densities. The results showed a strong positive linear relationship ($R^2 = 0.92$) between pH and final egg population density. Reproduction was 2.0 times greater in the 8.1 pH soil compared to the 5.5 pH soil. Further research is planned to investigate the effects of pH on the *H. glycines* life cycle.

Characterization of the complete internal transcribed spacer region of the nuclear ribosomal DNA of Typhula spp. S. M. MILLETT (1) and D. P. Maxwell (2). (1) Wisconsin Dept. of Agriculture; (2) University of Wisconsin. Phytopathology 92:S56. Publication no. P-2002-0407-AMA.

Rapid and accurate identification of *Typhula* spp. can be difficult using *in vitro* methods. Isolates of the *Typhula ishikariensis* complex (TISH), *T. incarnata* and *T. phacorrhiza* were characterized by sequence analysis of the complete internal transcribed spacer region (CITS) of the nuclear ribosomal DNA. The hypotheses tested were that the CITS could be used to differentiate *Typhula* spp., especially the biological species of the TISH complex (BSI and BSII). DNA was extracted from the isolates, amplified, transformed and then sequenced. Percentage of sequence identity for the 44 sequences from the 30 *Typhula* spp. isolates ranged from 59 to 100% with a 0 to 30% divergence. The 26 sequences from the 20 TISH isolates were 87 to 100% identical to each other with a 0 to 8% divergence. These results indicate that the CITS region can be used to identify the *Typhula* spp. "Signature sequences" were discovered that differentiate BSI and BSII of the TISH complex, however, sequence analyses infer they are the same species.

Factors affecting take-all of wheat and its management in Arkansas. E. MILUS, R. Cartwright, C. Rothrock, M. Anders, P. Rohman, and C. Parsons. Dept. of Plant Pathology, University of Arkansas, Fayetteville, AR 72701. Phytopathology 92:S56. Publication no. P-2002-0408-AMA.

Based on plant and soil samples from 13 commercial fields over three growing seasons, take-all of wheat was associated with multiple years in a wheat / soybean double-cropping system, soil pH > 6.5, and in certain fields, deficiencies of sulfur, copper, or manganese. In irrigated fields, a one-year rotation out of wheat, regardless of the rotation crop, allowed the next wheat crop to be grown with minimal take-all. Rotations with rice were the most beneficial for reducing take-all. Cool-season grassy weeds, especially rescuegrass, prairie wedgegrass, cheatgrass, and ryegrass, promoted survival of the take-all fungus and need to be controlled during the seasons out of wheat. In non-irrigated fields, rotation options are limited, and continuous wheat with summer fallow was the best option for managing take-all. In a separate 2-year study, survival of the take-all fungus over

summer was much less in rice fields because of anaerobic conditions and in fallow fields because of high soil temperatures than in irrigated corn or soybean fields.

Ultrastructural characterization of infection and colonization of maize leaves by *Colletotrichum graminicola*, and by a *C. graminicola* pathogenicity mutant. C. W. Mims (1) and L. J. VAILLANCOURT (2). (1) Dept. Plant Pathology, University of Georgia, Athens, GA 30602; (2) Dept. Plant Pathology, University of Kentucky, Lexington, KY 40546. Phytopathology 92:S57. Publication no. P-2002-0409-AMA.

We observed the ultrastructure of infection of leaves of maize by *Colletotrichum graminicola* and by a *C. graminicola* pathogenicity mutant. The mutant, which is deficient in signal peptidase, causes no symptoms. There was no difference in infection or colonization by the mutant or wild-type up to 48 hours after inoculation. Both strains produced appressoria within 24 hours, and by 36 hours, both had penetrated host epidermal cells. Host cells formed papillae, but these did not often prevent penetration by either strain. For both strains, penetration was followed by formation of swollen infection hyphae which grew biotrophically for a short time. Both strains grew cell-to-cell, initiating new biotrophic interactions in each cell, between 36 and 48 hours after inoculation. A dramatic difference finally occurred when the mutant failed to transition to necrotrophy, while the wild-type made that switch between 48 and 72 hours after inoculation.

Quorum sensing modulates biofilm formation in *Pantoea stewartii* subsp. *stewartii*. T. MINOGUE (1), M. Koutsoudis (2), and S. Beck von Bodman (1,2). (1) Dept. of the Plant Science, University of Connecticut; (2) Dept. of the Molecular and Cell Biology, University of Connecticut. Phytopathology 92:S57. Publication no. P-2002-0410-AMA.

Quorum sensing is essential for the cell density-dependent expression of a capsular polysaccharide (CPS), which is an essential virulence factor in the cause of *Pantoea stewartii* subsp. *stewartii* (*Ps*) induced Stewart's wilt. EsaR is the quorum sensing regulator that represses CPS biosynthesis at low cell density, and derepression requires inducing levels of the *N*-(3-oxo-hexanoyl)-L-homoserine lactone signal, which is synthesized by the EsaI enzyme. We have recently determined that the cell density-dependent, quorum sensing regulated synthesis of CPS is required for biofilm development and maturation. Specifically, we analyzed the wild type strain DC283 and the quorum sensing mutants ESN51, *esal*(CPS⁻), and ESdeltaIR, *esal esaR*(CPS⁺), for their ability to attach to an artificial substrate. These studies found that the wild type strain attaches at low levels during the early growth phase, while ESN51 attaches strongly and ESdeltaIR is essentially attachment deficient. Epifluorescence microscopy analyses using the same strains, constitutively expressing GFP, show differential microcolony formation, colony morphology, and biofilm development. Moreover, the *In planta* dissemination of strains lacking CPS, or expressing CPS constitutively, is significantly attenuated. Confocal microscopy studies show biofilm depth and maturation is dependent upon the production of CPS. We present evidence that the regulation of CPS is critical for the development and maturation of biofilm, and, that this developmental process is critical for the overall disease biology of the bacterium.

Generation of non-pathogenic mutants of *Acidovorax avenae* subsp. *citricola* by chemical and insertional mutagenesis. G. V. Minsavage and J. B. JONES. Dept. of Plant Pathology, University of Florida, Gainesville, FL 32611. Phytopathology 92:S57. Publication no. P-2002-0411-AMA.

Acidovorax avenae subsp. *citricola* (Aac) causes a serious disease of both seedlings and mature fruit in watermelon production fields. Chemical mutagenesis of a wild-type strain (AacTX19) with *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine (NTG) and subsequent inoculation of pepper leaves and watermelon seedlings with 400 individual colonies of NTG-treated bacteria resulted in the identification of nine non-pathogenic mutants. All nine mutants failed to cause disease in watermelon seedlings and to elicit a wild-type non-host hypersensitive response (HR) when infiltrated into leaves of pepper. In a second experiment, 1200 Tn5-insertion mutants of a wild-type strain (Aac37) were individually tested for HR in pepper leaves and one mutant (Aac248) was negative. In population studies, this mutant failed to grow in susceptible watermelon seedlings. Cosmid libraries were generated from DNA of AacTX19 and Aac248 for use in complementation tests and sequence analysis of the region flanking the Tn5 insertion.

Controlling powdery mildew in flowering dogwood with a bicarbonate salt and household soaps. M. T. Mmbaga. Tennessee State University. Phytopathology 92:S57. Publication no. P-2002-0412-AMA.

Four biorational products were compared with four synthetic fungicides in controlling powdery mildew in field-grown seedlings, and two cultivars of flowering dogwood between 2000 and 2001. The biorational products were: ArmicarbTM (potassium bicarbonate), Ajax[®], Equate[®] and Palmolive[®] (household soaps). The fungicides were: Banner[®] (propiconazole), Clearys 3336[®] (thiophanate methyl), Heritage[®] (azoxystrobin) and Phyton 27[®] (copper sulfate pentahydrate). While biweekly applications of Clearys 3336 and Banner were clearly better than Heritage and Phyton 27 in suppressing powdery mildew, weekly applications of the biorational products provided the best control. Biweekly applications of the biorational products were highly effective compared to the non-treated control. Spray regimes that included biorational products and a Banner rotation gave better disease control and enhanced plant growth over the biorational products alone. Biweekly applications of the biorational products with a Banner rotation were as effective as biweekly applications of Banner or Clearys 3336 alone.

Environmental influences on development of pseudothecia and the production and dispersal of ascospores of *Mycosphaerella citri*, the cause of citrus greasy spot. S. N. MONDAL and L. W. Timmer. University of Florida, CREC, Lake Alfred, FL 33850. Phytopathology 92:S57. Publication no. P-2002-0413-AMA.

Development of pseudothecia and the production and dispersal of ascospores of *Mycosphaerella citri* under different environmental conditions were evaluated on infected decomposing grapefruit leaves. The pathogen required intermittent moisture for pseudothecial development and ascospore formation. The most pseudothecia and ascospores were produced when leaves were soaked 2 h per day 3 times per week at 28°C. The pathogen produced most pseudothecia and ascospores when infected leaves were incubated at 100% RH for 3 days, then dried for one day. In the field, pseudothecia were produced quickly on leaves in the summer rainy season from June to September, but total pseudothecia and ascospores produced were few. From October to May, pseudothecia developed and matured more slowly, but were produced in much larger numbers. The number of pseudothecia, but not ascospores, were negatively related to average temperature and total rainfall.

Pseudothecial development and ascospore production of *Mycosphaerella citri*, the cause of citrus greasy spot as affected by CaCO₃, Dolomite or Urea. S. N. MONDAL and L. W. Timmer. Citrus Research and Education Center, University of Florida, Lake Alfred, FL 33850. Phytopathology 92:S57. Publication no. P-2002-0414-AMA.

Mycosphaerella citri produces ascospores in the leaf litter that serve as the major source of inoculum for greasy spot. The effect of CaCO₃, dolomite or urea on development of pseudothecia and production of ascospores was evaluated in the greasy spot infected matured grapefruit leaves. In laboratory experiments, one urea application reduced production of pseudothecia and ascospores by up to 90%, but did not affect leaf decomposition. Application of CaCO₃ or dolomite accelerated the development of pseudothecia and ascospore release, but reduced the total numbers produced. Immature pseudothecia on leaves treated with CaCO₃, dolomite or urea degenerated rapidly and produced few ascospores. Similar results were observed in the microplot systems under field conditions. Applications of CaCO₃, dolomite or urea can reduce ascospore inoculum and be useful in an integrated program for greasy spot control.

Development of a sorghum ergot (*Claviceps africana*) prediction model for hybrids in northern Mexico. N. MONTES (1), G. Odvody (2), H. Williams (3), and T. Isakeit (1). (1) Dept. Plant Pathology, Texas A&M University, College Station, TX 77843; (2) Texas Agricultural Experiment Station, Corpus Christi, TX 78410; (3) INIFAP, Campo Experimental Rio Bravo, Tam., Mexico. Phytopathology 92:S57. Publication no. P-2002-0415-AMA.

Sorghum ergot, caused by the fungus *Claviceps africana*, can develop in hybrids damaged by low temperatures 2 to 3 weeks before flowering. Hybrids (36) were planted in Rio Bravo, Tam., Mexico at six dates between May and September, 1997, to observe the relationship between weather variables and natural ergot incidence, and to develop an ergot prediction model for hybrids. Ergot developed when low temperatures occurred 10 to 12 days before half bloom and followed by high relative humidity during flowering. A three-factor surface response regression model was developed that accounted for 87% of the variation in disease incidence ($P < 0.001$). The factors were: triad values of maximum and minimum temperatures 10 to 12 days before half bloom, and the minimum relative humidity during flowering. The model predicted that the disease could have the greatest impact if hybrids are planted during the fall, winter and early spring months of the year.

Propamocarb resistance in species of *Pythium*. G. W. MOORMAN (1), N. Arora (1), and S. H. Kim (2). (1) Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802; (2) Bureau of Plant Industry, Pennsylvania Dept. of Agriculture, 2301 N. Cameron St., Harrisburg, PA 17110. Phytopathology 92:S58. Publication no. P-2002-0416-AMA.

During 1996-2002, *Pythium* was found to be involved in 120 samples from commercial greenhouses in Pennsylvania. Of these isolates, 28 were found to be resistant to propamocarb. One or more isolates of *Pythium aphanidermatum*, *P. cylindrosporium*, *P. dissotocum*, *P. heterothallicum*, group F, *P. irregulare*, *P. splendens*, and *P. ultimum* had EC50s exceeding 4000 ug of propamocarb per ml of corn meal agar. The EC50s of all other isolates were near or well below 100 ug/ml. Resistant isolates overcame the presence of propamocarb and killed geranium seedlings. A discriminatory concentration of 1000 ug/ml for the routine *in vitro* screening of isolates for resistance is proposed. This is the first report of *Pythium* resistance to propamocarb.

Time and number of applications of the mycoherbicide *Phomopsis amaranthicola* affect *Amaranthus lividus* interference with pepper. J. P. MORALES-PAYAN (1), R. Charudattan (2), W. M. Stall (1), and J. DeValerio (2). (1) Hort. Sci. and (2) Plant Pathol. Dept., IFAS, Univ. of Florida, Gainesville, FL 32611. Phytopathology 92:S58. Publication no. P-2002-0417-AMA.

Phomopsis amaranthicola was applied (1×10^6 conidia/ml) to a bell pepper (*Capsicum annuum*, cv. Camelot) crop artificially infested with *Amaranthus lividus* (livid amaranth) (3 plants/m²). The fungus was applied 0, 1, 2, 3, or 4 times to the weed-crop canopy, with the first application at 10, 20, 30, or 40 days after weed emergence (DAE). Uncontrolled, *A. lividus* interference reduced pepper yield by 33 percent (%). *A. lividus* was partially controlled when *P. amaranthicola* was applied once at 10 or 20 DAE, but pepper yield loss was greater than 23%. One application at 30 or 40 DAE was less effective (>30% yield loss). Two applications at 10 and 20 DAE caused 35% weed mortality, and the yield loss was about 13%. No further yield improvement was achieved with 3 or 4 applications. Thus, one or two early applications of *P. amaranthicola* can significantly reduce *A. lividus* interference with pepper and improve crop yield. Best yields were obtained with two early applications of the mycoherbicide.

Estimating green leaf area index (GLAI) of soybean canopies using a radiometer. A. J. A. MOREIRA, J. Guan, and F. W. Nutter, Jr. Iowa State University. Phytopathology 92:S58. Publication no. P-2002-0418-AMA.

Accurate estimates of green leaf area index (GLAI) have potential to be used to estimate soybean yields. A preliminary step toward this goal is to estimate GLAI. Most methods for GLAI estimation are destructive and costly. Accuracy, precision, and costs must be taken into consideration when measuring GLAI. A multispectral, hand-held radiometer may make it possible to measure GLAI accurately and precisely in a non-destructive manner. Percentage reflectance from soybean canopies in eight narrow-wavelength bands (460, 510, 560, 610, 660, 710, 760, 810 nm) was related to GLAI by measuring area of harvested green leaf tissue using a leaf area meter. At 760 nm ($Y = 1.46 + 0.11 X$) and 810 nm ($Y = 1.56 + 0.10 X$), percentage reflectance explained 82% and 81%, respectively, of the variation in GLAI. Based on this information, percentage reflectance could be used to non-destructively estimate soybean GLAI during the growing season. There was a close linear relationship between percentage reflectance at 760 and 810 nm ($Y = 1.28 + 1.05 X$, $R^2 = 99.8\%$).

Negative signaling among rhizobacteria. J. E. MORELLO, E. A. Pierson, and L. S. Pierson III. Dept. Plant Pathology, The University of Arizona, Tucson, AZ 85721. Phytopathology 92:S58. Publication no. P-2002-0419-AMA.

Pseudomonas aureofaciens 30-84 is a model system for the study of the influence of the rhizosphere microbial community on gene expression patterns. *P. aureofaciens* 30-84 suppresses take-all disease of wheat caused by *Gaeumannomyces graminis* var. *tritici* (Ggt). Phenazine antibiotic production is responsible for pathogen inhibition and rhizosphere persistence by 30-84. Previously, ca. 6% of a wheat rhizosphere community library was shown to block phenazine production in 30-84, several by interfering with expression of the *phz* biosynthetic operon via the production of extracellular signals. These signals are not extractable using conditions favorable for other known negative compounds. A genomic library of one negative signaling bacterium was constructed and is being screened for negative signal production. *In vitro* analyses show these negative cross-communicating bacteria interfere with the ability of *P. aureofaciens* to inhibit the growth of Ggt. Thus, negative cross-communication among the rhizobacteria community may reduce the ability of a biological agent to control plant disease in the field.

Effects of spring burning and fungicide applications on big bluestem and its pathogens in a tallgrass prairie and a monoculture. G. W. MORGAN, K. A. Garrett, T. C. Todd, and N. A. Tisserat. Department of Plant Pathology, Kansas State University, Manhattan, KS 66506. Phytopathology 92:S58. Publication no. P-2002-0420-AMA.

We studied the effects of spring burning and fungicide applications on pathogen development and biomass of big bluestem (*Andropogon gerardii*) in a natural ecosystem at the Konza Prairie Biological Station and a monoculture planting at the USDA Plant Materials Center (PMC) near Manhattan, KS. In June, *Burkholderia andropogonis* was the most common pathogen at both locations. Rust caused by *Puccinia andropogonis* was the most prominent disease observed from late June through September. The incidence of both diseases was low (<10%) in 2001 due to hot, dry weather. Severity of *B. andropogonis* was reduced in spring-burned plots at Konza, but not at PMC. Rust severity was not affected by burning at either location, but was reduced ($P < 0.05$) with biweekly applications of the fungicide zoxytrobilin. Biomass accumulation at Konza and seed yield at PMC were not affected by fungicide application. However, in a greenhouse study four azoxystrobin applications at two-week intervals reduced ($P < 0.05$) the biomass of big bluestem in the absence of rust.

Effect of temperature on the daylily rust pathogen, *Puccinia hemerocallidis*. D. S. MUELLER (1), J. L. Williams-Woodward (2), and J. W. Buck (1). Dept. Plant Pathology, University of Georgia, (1) Griffin, GA 30223; (2) Athens, GA 30602. Phytopathology 92:S58. Publication no. P-2002-0421-AMA.

A serious new rust disease on daylilies (*Hemerocallis* spp.) caused by the fungus *Puccinia hemerocallidis* has become an increasing problem for daylily growers. At present little is known about the biology of *P. hemerocallidis*. The objectives of this study were to determine the effect of temperature on *P. hemerocallidis* urediniospore germination and infection of daylily. Rust urediniospores germinated between 7°C and 32°C on PDA, but did not germinate at 4, 34 and 36°C. Urediniospore incubation at 36°C for 6 h followed by 8 h at 22°C lowered germination 95%. One week of incubation at 4°C followed by 8 h at 22°C lowered germination 54%. Infection of daylily 'Pardon Me' was reduced from 8.6 lesions/cm of leaf at 22°C to 0.7 lesions/cm of leaf at 10°C and 0.2 lesions/cm leaf at 30°C. There were fewer than 0.1 lesions/cm of leaf at 4 and 36°C. These data suggest that daylily infection will be reduced during extended periods of hot weather associated with the summer growing season in the SE United States.

Summary of quinoxifen performance for grape powdery mildew control in the USA and Europe. J. P. MUELLER (1) and E. A. Green (2). (1) Dow AgroSciences LLC, 175 Mesquite Ct. Brentwood, CA 94513; (2) Dow AgroSciences, Letcombe Regis, Wantage, Oxon, OX12 9JT, UK. Phytopathology 92:S58. Publication no. P-2002-0422-AMA.

Quinoxifen fungicide (Quintec™, Arius™), registered in Europe, and in development in the USA, has a unique mode of action for powdery mildew control. University and Dow AgroSciences researchers in vine growing regions worldwide have tested it for *Uncinula necator* control in grapes. 27 U.S. trials conducted from 1997-2001, and more than 100 European trials, were evaluated for consistency and efficacy under a variety of environmental conditions. Season-long quinoxifen, and alternations with other modes of action, were tested at spray intervals ranging from 7 to 21 days. Analysis shows that quinoxifen performance was consistently among the best of all products tested. In a wide range of spray volumes, 50 to 75 grams active ingredient (g.a.i./hectare) were very effective at 14 day intervals, and in risk index model timings. At 110 g.a.i./hectare, it is one of very few products which provide control when used on 21 day intervals. This flexibility leads to a variety of options for use in growers' mildew control programs, and as a resistance management tool.

Diversity in the RNA-2 genome of pecluviruses causing peanut clump disease in West Africa and India. R. A. NAIDU (1,3), S. A. Sawyer (2), and C. M. Deom (1). (1) Dept. Plant Pathology, University of Georgia, Athens, GA 30602; (2) Dept. Mathematics and Genetics, Washington University, St. Louis, MO 63130; (3) previously at ICRISAT, Patancheru (PO), India 502 324. Phytopathology 92:S58. Publication no. P-2002-0423-AMA.

The complete sequence of RNA-2 of four isolates of *Peanut clump virus* (PCV) and two isolates of *Indian peanut clump virus* (IPCV) were determined. The organization of their genomes was similar to the two previously published sequences of PCV and IPCV. The size of RNA-2 ranged between 4290 and 4652 nucleotides with sequence identities between 58 and 89%. Among the five open reading frames (ORFs), the protein encoded by ORF 4 is highly conserved whereas that encoded by ORF 2 is least conserved. The

coat protein of these eight isolates showed amino acid identities between 37 and 89% and contained several conserved residues found in rod-shaped viruses like *Tobacco mosaic* and *Tobacco rattle viruses*. Phylogenetic trees based on the entire RNA-2 revealed three clusters. Two of the African PCVs appear to cluster more strongly with the Indian IPCVs than with the other African isolates. The data indicates that there is substantial divergence among the RNA-2 genomes of PCV and IPCV isolates.

Cloning and sequence analysis of the genome of beet mosaic potyvirus. L. G. NEMCHINOV and R. W. HAMMOND. USDA-ARS, Molecular Plant Pathology Laboratory, Beltsville, MD 20705. Phytopathology 92:S59. Publication no. P-2002-0424-AMA.

Beet mosaic virus (BtMV), a member of the economically important Potyvirus group, was first reported in *Beta vulgaris* in Germany in the late 1950s. It is distributed worldwide in all major beet growing areas. To better understand the molecular biology of the virus, we have determined the nucleotide sequence of cloned viral cDNA. BtMV has a single open reading frame typical for *Potyriviruses*. Amino acid sequences of the CI RNA helicase, nuclear inclusion (NIa) protein, RNA-dependent RNA polymerase (RdRp), and capsid protein (CP) show significant similarities to homologous sequences from other potyviruses. Putative proteolytic cleavage sites for the NIa protease and the group-specific signature at the NIa-NIB junction were predicted by analogy to consensus sequences and genome arrangements of potyviruses. The RdRp identifiers common for positive-strand RNA viruses are also conserved in BtMV. The DAG motif, important for aphid transmission in *Potyriviruses*, is present near the N terminus of the CP. BtMV appears to be a distinct species of genus *Potyrivirus* with the most closely related species being *Peanut Mottle Virus* (approximately 60% amino acid identity).

Role of *Clonostachys rosea* in preventing the overland transmission of *Ceratocystis fagacearum*. M. F. NEUMAN and J. JUZWIK. Dept. Plant Pathology, Univ. of Minnesota and USDA Forest Service, St. Paul, MN 55108. Phytopathology 92:S59. Publication no. P-2002-0425-AMA.

The overland spread of oak wilt results from acquisition and successful transmission of *Ceratocystis fagacearum* spores from fungal mats to fresh wounds on healthy oaks by insects. Secondary microbes, such as *Clonostachys rosea*, frequently colonize mats. In our previous *in vitro* studies, overgrowth of *C. fagacearum* colonies by *C. rosea* prevented recovery of viable *C. fagacearum* propagules. Two field trials were conducted to determine the effect of *C. rosea* on *C. fagacearum* propagule viability and recovery from naturally occurring mats. Augmentation sprays with *C. rosea* were used to achieve different amounts of mat colonization by that fungus. A higher frequency of subsamples (19% of 1332) from treated mats yielded *C. rosea* than those from non-treated mats (5% of 516). Conversely, a lower frequency (49% of 1332) of subsamples from treated mats yielded *C. fagacearum* compared to subsamples from non-treated mats (68% of 516). Therefore, *C. rosea* is likely contributing to natural control of overland spread of *C. fagacearum* by reducing inoculum availability.

Isolation, identification, and detection of undescribed sweetpotato viruses. S. L. NEW, J. A. ABAD, and J. W. MOYER. Dept. Plant Pathology, North Carolina State University. Phytopathology 92:S59. Publication no. P-2002-0426-AMA.

A study was initiated to detect the presence of undescribed viruses affecting sweetpotatoes. Novel sweetpotato viruses are difficult to isolate due to the presence of SPFMV; therefore, an alternative strategy was developed. Double stranded (ds) RNA was extracted from two sweetpotato cvs, 'Beaugard' (GWB) and 'White Bunch' (WB), and three additional cvs with 'russet crack' disease symptoms. Using dsRNA as a template, RT-PCR was performed with universal degenerate primers for potyviruses. The amplicons were cloned into pGemT vector (Promega, Madison, WI). After screening the library, clones were sequenced and compared with known sequences. From 18 clones obtained from GWB, six were closely related to SPFMV-RC, five were homologous with SPFMV-C and seven were related to sweetpotato virus G. These results show not only the efficacy of our system but also demonstrate that strains of the same virus (SPFMV RC and C) are coinfecting the same host, suggesting that they may be different viruses. The isolation of each virus in a selected host range and the identification of undescribed viruses in WB and plants with 'russet crack' is underway.

Implication of stigmatic exudate in infection of blueberry flowers by *Monilinia vaccinii-corymbosi*. H. K. NGUGI and H. SCHERM. Dept. of Plant Pathology, University of Georgia, Athens 30602. Phytopathology 92:S59. Publication no. P-2002-0427-AMA.

Infection of blueberry flowers by *Monilinia vaccinii-corymbosi*, which causes mummy berry disease, occurs when conidia germinate on the stigmatic surface, followed by hyphal ingress into the stylar canal and subsequent colonization of the ovary. We have previously shown that while no germination occurs in deionized water (dH₂O), the exudate produced on the wet stigmata of blueberry flowers strongly enhances germination of *M. vaccinii-corymbosi* conidia *in vitro*. In further experiments, germination increased up to 77.9% as exudate concentration in dH₂O was increased. Germination also occurred in tap water and in 0.2% sucrose in dH₂O but at levels significantly lower than in dH₂O containing exudate. The exudate also enhanced germination or hyphal growth rates in seven fungal species non-pathogenic to blueberry, but the effect was not as pronounced as in *M. vaccinii-corymbosi*. On detached flowers, removal of the exudate by washing reduced hyphal growth and style necrosis following inoculation with *M. vaccinii-corymbosi*. Exudate utilization and potential implications for directional guidance of hyphal growth within the stylar canal will be discussed.

Further characterization of the HG type classification test for soybean cyst nematode field populations. T. L. NIBLACK (1), R. D. RIGGS (2), J. WANG (3), and R. D. HEINZ (3). (1) Dept. Crop Sciences, University of Illinois, Urbana, IL 61801; (2) Dept. Plant Pathology, University of Arkansas, Fayetteville, AR 72701; (3) Dept. Plant Microbiology and Pathology, University of Missouri, Columbia, MO 65211. Phytopathology 92:S59. Publication no. P-2002-0428-AMA.

The HG Type classification system is a tool for characterizing genetically diverse populations of *Heterodera glycines*, the soybean cyst nematode (SCN). HG Type classification is based on a bioassay very similar to that used for race classification except in two important aspects: 1) 'Pickett' is not included in the list of soybean lines used for HG Type designation; and 2) HG Types are defined by SCN development on seven soybean lines rather than four. Both HG Types and races are determined on the basis of a Female Index (FI), calculated as follows: (mean number of females on test soybean line / mean number of females on standard susceptible) × 100. We compared HG Type and race classification on over 100 SCN populations to determine their relative utility for making cultivar recommendations to farmers. We tested the use of alternative susceptible cultivars and found that designation of one standard, Lee 74, is essential for repeatability of HG Type tests. We also tested the use of eggs per cyst and cysts per gram root as an alternative to the FI and found that neither is generally worth the additional effort involved in obtaining such numbers.

Weaponry revealed: secreted proteins of *Erwinia amylovora*. R. M. NISSINEN (1), K. J. VAN WIJK (2), J. A. YTTERBERG (2), T. THANNHAUSER (3), and S. V. BEER (1). (1) Dept. Plant Pathology; (2) Dept. Plant Biology; (3) Biotechnology Center, Cornell University, Ithaca, NY 14853. Phytopathology 92:S59. Publication no. P-2002-0429-AMA.

The pathogenicity of *Erwinia amylovora*, the causal agent of fire blight, depends on the chromosomal *hrp/dsp* gene cluster, located on a pathogenicity island of 66 kbp. We aimed to isolate and identify proteins secreted via the Hrp pathway by a microproteomics approach. We isolated secreted proteins from culture supernatants of several *E. amylovora* strains and mutants grown under *hrp*-inducing conditions. The secreted proteins were resolved by 2-D-PAGE, and proteins were identified by MALDI-TOF-MS. Clear differences were seen between protein maps of wild-type strain Ea273 and mutants impaired in Hrp-secretion or in *hrp*-gene induction indicating that at least 15 proteins are secreted via the *E. amylovora* Hrp pathway under apoplast-mimicking conditions. A promoter trap library was developed to isolate genes that are induced in apoplast-mimicking conditions, including the genes encoding the secreted proteins.

Comparison of the spatial pattern of two foliar diseases of strawberry. M. NITA and L. V. MADDEN. Department of Plant Pathology, The Ohio State University, OARDC Wooster, OH 44691. Phytopathology 92:S59. Publication no. P-2002-0430-AMA.

Spatial distributions were determined over 3 yr for two foliar diseases, Phomopsis leaf blight (causal agent: *Phomopsis obscurans*), and powdery mildew (causal agent: *Sphaerotheca macularis*), occurring in the same commercial strawberry fields. Incidence of each disease was recorded from n = 15 leaflets in each of N = 100 (10 by 10 grid) evenly spaced sampling units. Degree of heterogeneity was measured with the *theta* parameter of the beta-binomial distribution (an indication of small-scale pattern), and degree of spatial correlation among sampling units was measured with the SADIE *Ia* index (an indication of larger scale patterns). Tests of *Ia* showed significant aggregation for all powdery mildew data sets, but for less than half of the Phomopsis data sets. Both *theta* and *Ia* were higher for powdery mildew than for Phomopsis in all data sets. Combined statistical results indicated that:

powdery mildew was aggregated at both small and large scales, with disease patches extending over multiple sampling units; and Phomopsis leaf blight was aggregated mostly at small scales, with patches being generally less than the size of a sampling unit.

Identification of Myrothecium leaf spot resistance in *Syngonium* species and cultivars. D. J. NORMAN, R. J. Henny, J. M. F. Yuen, and T. A. Mellich. Mid-Florida Research and Education Center, University of Florida, Apopka, FL 32703. *Phytopathology* 92:S60. Publication no. P-2002-0431-AMA.

All widely-grown cultivars of *Syngonium podophyllum* (Araceae) are susceptible to Myrothecium leaf spot (*Myrothecium roridum*). Therefore, commercial production of *Syngonium* requires rigorous sanitation programs and frequent applications of fungicides to control this disease. The goal of this research was to identify species and noncultivated accessions of *Syngonium* that are resistant to *Myrothecium*. Five commercial cultivars and 30 accessions, comprising 16 different *Syngonium* species, were screened for resistance. All five commercial cultivars of *S. podophyllum* were susceptible to *M. roridum*. However, seven species (*S. neglectum*, *S. wendlandii*, *S. dodsonianum*, *S. erythrophyllum*, *S. chiapense*, *S. dodsonianum*, and *S. angustatum*) showed useful levels of resistance, as did two noncultivated accessions of *S. podophyllum*. DNA profiles of resistant species and accessions were compared to susceptible ones with AFLP. Unique polymorphisms were identified in resistant plants which allowed comparisons of disease resistance and genetic markers that can be useful in future breeding work.

Relationship between Botryosphaeria blight severity and changes in carbohydrate content in pistachio. N. NTAHIMPERA, D. G. Felts, and T. J. Michailides. Department of Plant Pathology, University of California, Davis, Kearney Agricultural Center, Parlier, CA 93648. *Phytopathology* 92:S60. Publication no. P-2002-0432-AMA.

Effect of changes in carbohydrate content of pistachio fruit on infection and symptom development of Botryosphaeria blight caused by *Botryosphaeria dothidea* has not been studied. To determine the relationship between Botryosphaeria blight and changes in carbohydrate content in pistachio hulls under field conditions, wounded and non-wounded pistachio fruit were inoculated with *B. dothidea* periodically in an orchard at the Kearney Agricultural Center in 2001. Disease progression was evaluated weekly and the relative AUDPC were calculated. Fruit characteristics were determined for each inoculation date. Fruit reached its final size in early July when the kernel occupied 90% of the shell cavity. Inoculations performed early May on wounded and non-wounded immature fruit resulted in higher severity than those performed later. The percentage of soluble solids ranged between 12 and 14%. Analysis of corresponding carbohydrate content is under investigation. Results will be discussed in relation to various disease control strategies.

Mapping the prevalence of Moko disease of banana using GPS and GIS. F. W. NUTTER, JR. (1) and R. A. Coelho Netto (2). (1) Iowa State University; (2) INPA, Coordenacao de Pesquisa em Ciencias Agronomicas, Manaus, AM Brazil. *Phytopathology* 92:S60. Publication no. P-2002-0433-AMA.

Moko disease of banana, caused by the bacterium *Ralstonia solanacearum*, causes a lethal disease that is especially devastating to banana growers in the Amazonia region of Brazil. Fifteen municipalities in the Amazonas State were inspected for Moko disease; between 3 and 26 plantations per municipality were assessed for disease prevalence (number of banana plantations with Moko detected/total number of banana plantations assessed) and disease incidence (number of banana plants with Moko/number of banana plants assessed). Disease prevalence and GPS data were entered into a GIS (ArcView) to generate disease prevalence (risk) maps for Moko disease. Moko disease was detected in 5 of 15 municipalities and disease prevalence within these municipalities ranged from 11 to 70%. Disease incidence within plantations ranged from 0.62% to 63.8%, although these values may underestimate the actual incidence of Moko disease within banana plantations because dead banana plants (probably killed by Moko) were not assessed.

Efficacy of biocontrol agents and resistance inducers against tomato bacterial spot in the greenhouse. A. OBRADOVIC (1), J. B. Jones (1), M. T. Momol (2), S. M. Olson (2), P. Pradhanang (2), and B. Balogh (1). (1) Plant Pathology Dept., University of Florida, Gainesville, FL 32611; (2) NFREC, University of Florida, Quincy, FL 32351. *Phytopathology* 92:S60. Publication no. P-2002-0434-AMA.

Bacterial spot incited by *Xanthomonas campestris* pv. *vesicatoria* is a serious threat in tomato commercial production. In an attempt to develop a more sustainable strategy for controlling the disease we investigated the effect of various combinations of plant growth-promoting rhizobacteria, foliar applications of harpin protein (Eden Bioscience), acibenzolar-S-methyl (ASM, Syngenta, Inc.), bacteriophage mixture (AgriPhi, Inc.), and antagonistic bacteria on disease development in the greenhouse. Untreated plants and those treated with copper hydroxide were used as controls. Following treatment with the chemicals and/or biologicals, the plants were inoculated with 10^8 CFU/ml of Xcv. ASM initiated plant defense mechanisms against the pathogen and completely prevented occurrence of the disease. Application of bacteriophage in combination with ASM decreased the intensity of atypical symptom development and provided successful disease control compared to the other treatments.

Management of tomato bacterial spot in the field by foliar applications of bacteriophages and SAR inducers. A. OBRADOVIC (1), J. B. Jones (1), M. T. Momol (2), S. M. Olson (2), P. C. King (2), and B. Balogh (1). (1) Plant Pathology Dept., University of Florida, Gainesville, FL 32611; (2) NFREC, University of Florida, Quincy, FL 32351. *Phytopathology* 92:S60. Publication no. P-2002-0435-AMA.

Due to the devastating effect of bacterial spot caused by *Xanthomonas campestris* pv. *vesicatoria* in tomato commercial production, a replicated field experiment was conducted to develop more efficient disease management strategy. Various combinations of harpin protein (Eden Bioscience), acibenzolar-S-methyl (ASM, Syngenta, Inc.), and bacteriophage mixture (AgriPhi, Inc.), were selected for control of bacterial spot in the field based on results from greenhouse experiments. Prior to setting into the field in the fall 2001, transplants were inoculated with a bacterial suspension. Disease severity was assessed periodically in the field. All ASM treatments applied alone or in combination with bacteriophage and/or harpin significantly reduced bacterial spot compared to copper-mancozeb (CM). Although total yield was not significantly affected, plots receiving phage alone or in combination with ASM or harpin had significantly more medium-size fruit than those treated with CM.

Determining efficacy of streptomycin sulfate for preventing shoot blight following an injury event. S. C. OCKEY and S. V. Thomson. Utah State University. *Phytopathology* 92:S60. Publication no. P-2002-0436-AMA.

An experiment was designed to address control of shoot blight on Jonathan apple trees under simulated climatic conditions including rain dispersal of bacteria and hail injury to foliage. The youngest leaf on actively growing shoots was either injured and inoculated or inoculated without injury. Trees were treated with streptomycin before the injury and inoculation or sprayed with streptomycin at 4 or 24 hours post injury and inoculation. Results showed no statistical difference in control or infection rates between either of the injury and inoculation treatments and were both effectively controlled with an application of streptomycin 4 hours after the injury and inoculation event. Slight control of infections was obtained when streptomycin was applied as a preventive spray before injury and inoculation when compared to the check without streptomycin applications. There was significantly less control when streptomycin was applied 24 hours after injury and inoculation. Streptomycin applied to the non-injured and inoculated treatments prevented all infections whereas there was 4% infection when no streptomycin was applied.

HR induction in tobacco by *Erwinia amylovora* requires DspE in addition to a harpin. C.-S. OH, W.-S. Kim, and S. V. Beer. Dept. Plant Pathology, Cornell University. *Phytopathology* 92:S60. Publication no. P-2002-0437-AMA.

Erwinia amylovora elicits a hypersensitive response (HR) on nonhost plants like tobacco. Previous work has shown that HrpN and HrpW, but not DspE elicit the HR in tobacco plants following infiltration of protein solutions into the intercellular spaces of leaves. We re-examined these effectors using two bacterial means to synthesize and deliver the proteins to plant cells. When expressed transiently with *Agrobacterium*, HrpN with a signal peptide and DspE alone induced clear HRs. The HR resulted also when *Escherichia coli* strain MC4100 carrying genes for Hrp type III secretion machinery of *E. amylovora* was infiltrated into the apoplast of tobacco leaves. Such strains expressing only HrpN or DspE among effectors did not induce HR on tobacco in contrast to the transient expression results. However, when two proteins, HrpN and DspE, or HrpW and DspE, but not HrpN and HrpW together were expressed in the same cell, clear HR was induced, suggesting that DspE is needed together with a harpin to induce an HR on tobacco where these proteins are presented by bacteria. The amount of HrpN and DspE secreted by bacteria is being determined through western blot analysis.

Wheat cultivar-dependent root architecture and root colonization by *P. fluorescens* Q8r1. P. A. OKUBARA (1), B. B. Landa (2), B. Madsen (1), and J. P. Kornoely (1). (1) USDA ARS, Root Disease & Biological Control, Washington State University, Pullman, WA 99164; (2) Dept. of Crop Protection, Institute of Sustainable Agriculture, CSIC, 14080 Córdoba, Spain. Phytopathology 92:S61. Publication no. P-2002-0438-AMA.

Host genes involved in interactions with soil microbes are expected to have important roles in the tolerance of plants to fungal root pathogens, and in the establishment of biological control agents in the rhizosphere. Identifying key host genes will provide genetic strategies and molecular tools for enhanced resistance to root pathogens. Twenty eight Pacific Northwest cultivars of *Triticum aestivum* (hexaploid wheat) representing six market classes of wheat have been selected on the basis of yield, flour quality, and resistance to leaf and stem pathogens. These are being examined for genetic variation in 1) root growth rate and branching patterns, and 2) ability to maintain rhizosphere populations of the aggressive *P. fluorescens* strain Q8r1 as compared to that of Q2-87, a weaker wheat root colonizer. In initial experiments, cvs. Penawawa, Boundary, Edwin, Lambert, Finley and Stephens showed higher root-associated populations of Q8r1 in either soil or seed treatment experiments. In general, roots from plants treated with Q8r1 were reduced in size, suggesting that root colonization had a detrimental effect in the absence of pathogens. Current colonization data will be discussed with respect to root characteristics.

Transplant root dips with biocontrol agents reduce strawberry black root rot. R. O. OLATINWO and A. M. C. Schilder. Dept. Plant Pathology, Michigan State University, East Lansing, MI 48824. Phytopathology 92:S61. Publication no. P-2002-0439-AMA.

Four commercial biocontrol products [Mycostop (*Streptomyces griseoviridis*), Primastop (*Gliocladium catenulatum*), Plantshield (*Trichoderma harzianum*), and Companion (*Bacillus subtilis*)], three *Paenibacillus* isolates (Pm-1, Pm-13s, and Pm-18) and two *Trichoderma* isolates (T2 and T10) were evaluated for their efficacy in controlling strawberry black root rot. The biocontrol agents were applied separately as a root dip to field-dug strawberry transplants (cv. Honeoye). The plants were transplanted into pots containing naturally infested field soil or sterilized soil inoculated with a mixture of *Rhizoctonia* and *Pythium* isolates. In the inoculated soil, Plantshield, Primastop, and Mycostop significantly reduced root lesions and increased root and shoot weight compared to the untreated control. T2 and T10 significantly reduced root lesions and increased shoot weight, whereas Pm-1 and Pm-18 increased dry shoot weight. In the field soil, only T10 significantly reduced root lesions and increased fresh and dry root weights over the untreated control. Promising biocontrol agents will be further evaluated in field trials.

Resistance of *Colletotrichum graminicola* to Qo inhibitors. G. OLAYA (1), C. Avila-Adame (2), and W. Koeller (2). (1) Syngenta Crop Protection, Vero Beach, FL 32967; (2) Department of Plant Pathology, Cornell University, Geneva, NY 14456. Phytopathology 92:S61. Publication no. P-2002-0440-AMA.

Isolates of *Colletotrichum graminicola* exhibiting resistance to Qo inhibitors have been collected from turf in the United States and Japan. The isolates were highly resistant to azoxystrobin, with resistance levels exceeding a factor of 1000. The sensitivity of the isolates was determined *in vitro* and the response was confirmed *in vivo*. Comparing the cytochrome b gene amino acid sequences, the insensitive isolates were distinguished from baseline isolates by a G143A mutation (glycine is replaced by alanine at position 143 in the gene sequence). This mutation is also involved in Qo inhibitor resistance in other plant pathogenic fungi. An extensive resistance monitoring program is in place to determine the extent and impact of the resistance in the control of this disease.

Control of foliar late blight with a premix formulation of zoxamide and mancozeb. B. D. OLSON, G. Kemmitt, R. J. Her, and J. Edmonds. Dow AgroSciences LLC, 9330 Zionsville Rd., Indianapolis, IN 46268. Phytopathology 92:S61. Publication no. P-2002-0441-AMA.

In 2001 Dow AgroSciences registered the premix formulation of zoxamide and mancozeb (1:8 ratio, respectively) under the trade name of Gavel 75DF, for control of early and late blight in potato. Zoxamide is a new active ingredient with fungicidal activity against oomycete species. In 2001, studies were conducted to evaluate the performance of Gavel versus other commercially available fungicides against late blight of potato caused by *Phytophthora infestans*. Trials were conducted in Inchy En Artois, France; Marcham, United Kingdom; and Phelps, New York (USA). At all locations, Gavel, applied at 2.25 kg of formulated product per hectare, provided control

of late blight that consistently ranked it in the top group of fungicides. In a separate study conducted in Phelps, New York, Gavel was compared to Acrobat MZ (dimethomorph / mancozeb), Curzate MZ (cymoxanil / mancozeb), Omega (fluazinam), Dithane 75DF (mancozeb) and Bravo (chlorothalonil) fungicides. Each product was applied at their respective 1, 1/2 and 1/4 X rates. Gavel along with fluazinam provided significantly greater control of late blight than all other fungicides tested.

Coinfection of potato with *V. dahliae* and geographic strains of *P. penetrans*. M. OMER (1), A. MacGuidwin (2), and R. Rowe (1). (1) Dept. Plant Pathology, OSU-OARDC, Wooster, OH 44691; (2) Dept. Plant Pathology, Univ. of Wisconsin, Madison, WI 53706. Phytopathology 92:S61. Publication no. P-2002-0442-AMA.

Two isolates of Vd (VCG 4A & 4B) and eight strains of Pp collected from USA (OH, MN, WA and WI) were used to evaluate the response of potato to single and co-infections. Generally, severity was highest on plants coinfecting with 4A plus any of the Pp strains, followed by co-infection with 4B plus Pp strains. No significant differences in disease severity were detected among most of the Pp strains alone, or in combination with 4A or 4B. Severity was lowest on control plants. Total tuber weight and number tended to be less influenced by co-infection with both pathogens than did disease severity. Tuber yield was very similar among plants treated with Pp alone. All eight geographic isolates of Pp did interact with Vd, but the interactions with 4A were the strongest. Moreover, most of the Pp strains reacted similarly within each of the two subgroups 4A and 4B. These results are consistent with previous work that both 4A and 4B are capable of interacting synergistically with Pp, though 4A is more aggressive. This confirms earlier observations on synergism and adds validity to this pattern of interaction.

Differentiation of rhizosphere microbial communities of tobacco, soybean and corn using fatty acid methyl ester analysis. K. L. ONG, D. A. Kluepfel, B. A. Fortnum, and M. B. Riley. Dept. of Plant Pathology and Physiology, Clemson University, Clemson, SC 29634. Phytopathology 92:S61. Publication no. P-2002-0443-AMA.

An effective method to manage bacterial wilt on tobacco caused by *Ralstonia solanacearum*, has been rotation with soybean and/or corn. It has been hypothesized that rotation crops are responsible for the generation/selection of a microbial community suppressive to *R. solanacearum* or the disease it causes. FAME analysis was used to characterize the effect of soybean and corn rotations on the rhizosphere community. FAME profiles were obtained by direct fatty acid extraction from soil and from extracting the microbial growth on serial dilutions of soil suspensions grown on R2A media. Principal component analysis of the FAME profiles differentiated the two extraction approaches. Further, FAME profiles, obtained by culturing, of the tobacco rhizosphere samples were different from corn and soybean. Fatty acids that differentiated tobacco, soybean and corn revealed a larger proportion of Gram-negative bacteria in the tobacco rhizosphere, while FAME profiles of soybean and corn suggest a larger proportion of Gram-positive bacteria.

Root system characteristics related to the resistance of commercial soybean varieties to sudden death syndrome. L. M. ORTIZ-RIBBING and D. M. Eastburn. Dept. of Crop Sciences, University of Illinois at Urbana-Champaign. Phytopathology 92:S61. Publication no. P-2002-0444-AMA.

Wide variation in resistance exists among commercial soybean varieties to sudden death syndrome, caused by *Fusarium solani* f.sp. *glycines*. Field experiments near Urbana, IL were designed to determine if root characteristics (architecture, root length, root surface area, root volume, average root diameter), time and location of colonization, disease severity and yield varied among 12 resistant and susceptible commercial soybean varieties. Significant differences in foliar symptoms, yield and root characteristics existed among the varieties when inoculated with the pathogen. Resistant varieties had significantly smaller root diameters and lower root volumes than susceptible varieties. Colonization by the fungus on the upper taproot and laterals significantly increased throughout the season. However, colonization on upper taproots, lower taproots and laterals was not significantly different among varieties. Yield and AUDPC were not significantly related. Results suggest that the soybean root system plays an important role in the resistance process.

Identification of the causal agent of fairy ring disease on cranberry. P. V. OUDEMANS (1), C. Constantelos (1), L. Wasilwa (1), J. Polashock (1), F. V. Caruso (2), and L. M. Carris (3). (1) Rutgers University; (2) University of Massachusetts; (3) Washington State University. Phytopathology 92:S61. Publication no. P-2002-0445-AMA.

Fairy ring on cranberry is a disease of uncertain etiology. Earlier literature suggests two possible causal agents, *Psilocybe agrariella* and a *Phialophora* sp. During the growing seasons of 2000 and 2001, a slow-growing coelomycete was isolated from affected vines in New Jersey and Massachusetts. The fungus was tentatively identified using sequence analysis of rDNA including ITS1, 5.8s and ITS2. Homology to published sequences was determined for two isolates of the fairy ring fungus using BLAST (Basic Local Alignment Search Tool). The most homologous (>80%) species were those of the ascomycete genus *Pezizula*. Sequences were aligned using the Megalign module in DNASTar and Clustal V. The tree was generated using PAUP. This analysis suggests that the fairy ring fungus in cranberry is unrelated to *Fusicoccum*, *Phialophora* and *Psilocybe*, but is probably a member of the genus *Pezizula*. High homology was noted for other members of the Dermataceae including *Dermea* and *Neofabrea*.

Detection and quantification of fairy ring disease on cranberry using remote sensing. P. V. OUDEMANS and M. G. Hughes. Department of Plant Biology and Pathology, Rutgers University Blueberry and Cranberry Research and Extension Center, Chatsworth, NJ 08019. Phytopathology 92:S62. Publication no. P-2002-0446-AMA.

Fairy ring on cranberry is a disease of uncertain etiology. Controls are time-consuming and expensive. To evaluate the relative effects of fairy ring, 1:12000 color-IR aerial photography for the time period covering 1987 - 2000 was used. Images were scanned into a digital format and then georeferenced. Diseased areas within each bed were delineated by "heads-up" digitizing in a GIS, using the imagery as a backdrop. Yield estimates were made for diseased and healthy areas and the total loss for the bed was estimated. To estimate yield effects, sampling points were placed both within diseased areas as well as in adjacent healthy areas. Results from the four adjacent cranberry beds of the cultivar Ben Lear show that over thirteen years, fairy rings within a bed generally increased in both size and number. In general, the majority of rings were perennial and increased in area by approximately 20 percent each year. The rings ranged in size from 2 - 20m in diameter. Results indicate that within a ring there was approximately a 52 percent reduction in cranberry yield.

Investigation of the ITS region for pathovar-specific detection of (*Pseudomonas syringae* pv. *lachrymans*) a pathogen of cucurbits, by polymerase chain reaction (PCR). Z. OZDEMIR and T. A. Zitter. Department of Plant Pathology, Cornell University, Ithaca, NY 14853. Phytopathology 92:S62. Publication no. P-2002-0447-AMA.

Pseudomonas syringae pv. *lachrymans* (Psl) is a pathogen of cucurbits that causes angular leaf spot. We investigated a putative PCR-based method for detection of the pathogen. Psl strains from different geographical origins were characterized with LOPAT, BIOLOG, and pathogenicity tests prior to isolating genomic DNA for amplification of 16S-23S ribosomal DNA, which includes internally transcribed spacer (ITS) regions. A universal primer pair was used to amplify the ITS region. Amplified PCR products from five Psl strains were sequenced and a BLAST search was done for similarity of sequences. No significant sequence differences were found in the ITS regions of Psl strains and of *Pseudomonas syringae* pathovars *apii*, *maculicola*, *papulans*, *phaseolicola*, *pisi*, *porri*, *syringae*, *tomato*. Based on these results, we were not able to design a pathovar-specific PCR primer from the ITS region.

Impact of *Meloidogyne graminicola* on yield of lowland rainfed rice in Bangladesh. J. L. PADGHAM (1), G. S. Abawi (2), J. M. Duxbury (1), and M. A. Mazid (3). (1) Dept. Crop and Soil Sciences, Cornell University, Ithaca, NY 14853; (2) Dept. Plant Pathology, Cornell University, Ithaca, NY 14853; (3) Bangladesh Rice Research Institute, Rajshahi, Bangladesh. Phytopathology 92:S62. Publication no. P-2002-0448-AMA.

Replicated plots with and without the application of Furadan 5G (40 kg/ha) were established at two locations in northwestern Bangladesh to assess the impact of *M. graminicola* on lowland rainfed rice in the rice-wheat system. At the first location, rice yields increased by 0.2, 0.7, and 1.0 t/ha where Furadan was applied to the seedbed only, to the field only, and to both the seedbed and field, respectively, compared to a nontreated control. At the second location, rice yield increased by 1.1 t/ha where Furadan was applied to both the seedbed and field compared to the control. At harvest, soil levels of *M. graminicola* in the nontreated plots were more than 3 times greater than those in the treated plots at both locations. Seedling shoot height and dry weight were significantly greater in the treated seedbed plots compared to the nontreated plots. This is the first on-farm study to demonstrate a negative impact of *M. graminicola* on rice growth and yield during the monsoon season in Bangladesh.

Detection of *Xanthomonas arboricola* pv. *pruni* with a PCR primer set and a digoxigenin-labeled DNA probe. C. M. PAGANI (1,2) and D. F. Ritchie (1). (1) Dept. Plant Pathology, NC State Univ., Raleigh, NC 27695-7616; (2) National Agriculture Research Institute, (INIA), Andes 1365, Uruguay. Phytopathology 92:S62. Publication no. P-2002-0449-AMA.

A DNA fragment obtained by random amplified polymorphic DNA (RAPD) analysis, apparently unique to *Xanthomonas arboricola* pv. *pruni* (Xap) was cloned and sequenced. PCR primers specific to this fragment were synthesized and the PCR product evaluated as a diagnostic DNA probe. A set of 120 strains of Xap could be differentiated from 63 strains of non-*Xanthomonas* species and pathovars including saprophytes associated with *Prunus* plants. The sensitivity of the PCR assay was 25 to 50 cells. Xap could be accurately detected and identified by PCR and probe hybridizations on asymptomatic samples from orchard trees in which bacterial spot had occurred the previous year. Thus, these two assays are considered important tools for detection and identification of Xap in overwintering sites on symptomless tissues of peach trees.

Continuous exposure to exogenous ethylene does not affect brown rot development on cold-stored stone fruits. L. PALOU, C. H. Crisosto, and L. M. Basinal. Dept. of Pomology, University of California, Davis, CA 95616. Phytopathology 92:S62. Publication no. P-2002-0450-AMA.

The influence of continuous exposure to exogenous ethylene during long-term cold storage on the development of postharvest brown rot was investigated during three seasons on selected cultivars of climacteric (peach, plum, nectarine, and apricot) and non-climacteric (sweet cherry) stone fruits. Fruit were wound inoculated with *Monilinia fructicola* and placed in plastic cavity trays in 7.8-L plastic containers. The containers were connected to an ethylene flow-through system inside standard cold storage rooms at each desired temperature. Depending on the experiment, climacteric stone fruit were exposed to ethylene concentrations of 0, 0.1, 1, 3, 10, or 100 ppm during storage at 0, 5, or 10°C for up to 28 days; sweet cherries were exposed to 0, 0.01, 0.1, or 1 ppm ethylene during storage at 0 or 5°C for 21 days. In every test, neither brown rot incidence nor severity were significantly affected by ethylene exposure. Therefore, no benefit in decay reduction can be expected from actively removing ethylene from commercial cold storage rooms containing stone fruits.

Biochemical implications of blocking the ergot alkaloid pathway of a grass endophyte. D. G. PANACCIONE (1), B. A. Tapper (2), G. A. Lane (2), C. L. Schardl (3), R. D. Johnson (1), C. Machado (3), E. Davies (2), and K. Fraser (2). (1) West Virginia University, Morgantown, WV 26506; (2) AgResearch, Palmerston North, New Zealand; (3) University of Kentucky, Lexington, KY 40506. Phytopathology 92:S62. Publication no. P-2002-0451-AMA.

Several *Neotyphodium* spp. that are endophytes of grasses produce ergovaline, a peptide associated with livestock toxicoses, and other ergot alkaloids. Disruption of the *lpsA* (lysergyl peptide synthetase) gene in the perennial ryegrass endophyte *Neotyphodium* sp. Lp1 eliminates the accumulation of ergovaline in infected plants. Since *lpsA* controls an intermediate step in the pathway, we are investigating the broad implications of this mutation. Profiles of significant non-ergot alkaloids (lolitrem and peramine) were similar in symbiota containing the wild type or *lpsA* knockout endophyte. Non-peptide ergot alkaloids were at similar or lower levels in the knockout than in wild type, with one exception. A compound with chromatographic and spectral properties of an ergot alkaloid, but that appears to have been previously uncharacterized, accumulated to higher concentrations in *lpsA* knockout-containing symbiota than in wild type-containing symbiota.

A galactosyl diacylglycerol from cucumber leaves inhibits the development of gray mold caused by spores of *Botrytis cinerea*. J.-H. Park, J.-C. KIM, G. J. Choi, H. T. Kim, and K. Y. Cho. Agrochemical Screening Team, Korea Research Institute of Chemical Technology, Taejon 305-600, Korea. Phytopathology 92:S62. Publication no. P-2002-0452-AMA.

When the potted cucumber plants of the 4th leaf stage were artificially inoculated with spores of *Botrytis cinerea*, the disease severities were remarkably different among leaves. Compared to 1st and 2nd leaves, 3rd and 4th leaves were highly resistant to *B. cinerea* infection. However, such a phenomenon did not occur in the cucumber leaves inoculated with mycelial discs of the fungus. Water extracts from individual leaves showed different inhibitory effects on the development of gray mold and the spore germination of *B. cinerea*. An active principle was purified from 4th leaves as white powders by repeated silica gel column chromatography, preparative TLC, and *in vivo* bioassay. Its chemical structure was determined as 2,3-di-(9Z,12Z,15Z-octadecatrienoyl)-2,3-dihydroxypropylgalactopyranoside by

various instrumental analyses. The principle exhibited dose-dependent inhibitory effects on the disease development *in vivo* and the spore germination of *B. cinerea* without any effects on the mycelial growth of the fungus. This substance appears to be at least partially responsible for the disease resistance of young leaves of cucumber plants against *B. cinerea* infection.

Purification and characterization of polygalacturonase from apple tissue infected by *Colletotrichum acutatum*. E. PARK, J. L. McEvoy, and W. S. Conway. USDA, PQSL, 10300 Baltimore Ave. Beltsville, MD 20705. Phytopathology 92:S63. Publication no. P-2002-0453-AMA.

Bitter rot caused by *Colletotrichum acutatum* can be a serious problem for the apple industry in the Eastern United States. To investigate various pathogenicity factors associated with this organism, two polygalacturonase isozymes were purified from apple tissue decayed by *C. acutatum*. These isozymes were purified using preparative isoelectric focusing and anion exchange chromatography. Two peaks were identified by Mono Q ion exchange chromatography, and one of the peaks was further characterized and exhibited three bands on SDS-PAGE. After digestion with N-glycosidase F, only one band remained, suggesting that the three bands represent one isozyme with various degrees of glycosylation. The approximate molecular weight for both isozymes is 35,000 Da. Substrate hydrolysis showed that both isozymes exhibit exo- as well as endopolygalacturonase activity. Optimal pH and heat stability were also determined. Future research is ongoing to determine if any additional polygalacturonase isozymes are produced by this pathogen.

The effect of calcium chloride infiltration into apples on pathogenicity of *Colletotrichum acutatum* during early stages of infection. E. PARK (1), J. L. McEvoy (1), W. S. Conway (1), and C. E. Sams (2). (1) USDA, PQSL, 10300 Baltimore Ave. Beltsville, MD 20705; (2) University of Tennessee, Knoxville, TN 37996. Phytopathology 92:S63. Publication no. P-2002-0454-AMA.

'Golden Delicious' apples were treated postharvest with 0% and 4% solution of calcium chloride and stored at 0°C for two months. Fruits were then inoculated with *Colletotrichum acutatum*, held at 0°C, and the lesion size measured over a 16 day period. Fungal growth and polygalacturonase (PG) expression were determined in Richard's solution supplemented with cell walls extracted from calcium treated or control fruit. During the first 18 hours, in both cases, PG activity was not detected. However, within 24 to 28 hours, the PG activity increased, and was 45% greater in the nontreated control than in the 4% calcium treated cell wall medium. After 36 hours, PG activity in both media was similar. These results correspond with *pg* gene expression studies. The pH was 4.4 in the control media and 5.1 in the 4% calcium medium after 28 hours. The concentration of ammonia, a factor associated with an increase in pH, was three-fold greater in the 4% calcium treated cell wall medium than in the control.

A transcription factor regulating appressorial penetration and infectious hyphae growth in *Magnaporthe grisea*. G. PARK and J. R. Xu. Dept. Botany & Plant Path., Purdue Univ., W. Lafayette, IN 47907. Phytopathology 92:S63. Publication no. P-2002-0455-AMA.

In the rice blast fungus *Magnaporthe grisea*, a MAP kinase *PMK1* is known to regulate appressorium formation and infectious hyphae growth. *MST12* that can weakly interact with *PMK1* in yeast two-hybrid assays was characterized as one of the transcription factors regulated by *PMK1* in this study. Mutants deleted of *MST12* failed to form functional appressoria and failed to penetrate and colonize rice plants. However, *mst12* mutants, in contrast to *pmk1* mutants, still produced normal and melanized appressoria. Appressoria formed by *mst12* mutants had appressorial pores but were unable to develop penetration pegs. These data indicate that transcription factors other than *MST12* must exist for regulating appressorium formation in *M. grisea*. Data on isolating and characterization of these transcription factors will be presented. In addition, we found a putative cAMP-dependent protein kinase phosphorylation site that was well conserved between *MST12* and its homologues from other filamentous fungi. This site may be involved in connecting the *PMK1* MAPK and cAMP signaling pathways and is currently being studied by site directed mutagenesis.

Susceptibility of *Vaccinium* to *Phytophthora ramorum*, cause of sudden oak death. J. L. PARKE (1), R. G. Linderman (2), and E. M. Hansen (1). (1) Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331; (2) USDA-ARS Horticultural Crops Research Laboratory, Corvallis, OR 97330. Phytopathology 92:S63. Publication no. P-2002-0456-AMA.

Phytophthora ramorum, cause of sudden oak death in California and Oregon, also causes foliar infection and dieback on several understory species including evergreen huckleberry (*Vaccinium ovatum*). The susceptibility of other wild *Vaccinium* species and of horticulturally important *Vaccinium* crops such as blueberry, cranberry, and lingonberry was tested in detached leaf assays using mycelial plugs or zoospore inoculum of *P. ramorum*. Disease severity was compared to that resulting from leaf inoculation of known hosts (tanoak, bay, evergreen huckleberry, rhododendron, madrone). A wide range of disease responses was observed among the *Vaccinium* species, from resistant (cranberry) to highly susceptible (lingonberry). The 20 blueberry cultivars tested also differed significantly in their response to the pathogen. Sporulation of the pathogen on excised leaf disks differed among plant species and cultivars, allowing quantification of the reproductive potential on each host.

Direct Inhibition of fungal pathogens by a synergistic mixture of Cutinase and Tween 20. D. M. PARKER and W. K. Köller. Cornell University, NYSAES. Phytopathology 92:S63. Publication no. P-2002-0457-AMA.

A mixture of cutinase with Tween 20 prevented infection of bean and cabbage leaves by *Sclerotinia sclerotiorum*. This correlated well with our previous report in which the same mixture protected bean leaves from infection by *Rhizoctonia solani* (MPMI 11:514-522). The mycelial growth of both pathogens *in vitro* was inhibited by the mixture. In addition, *in vitro* germination of ascospores of *S. sclerotiorum* was inhibited. The mixture was toxic to fungal mycelia resulting from permeabilization of the plasma membrane. Individual components of the mixture did not adversely impact either fungus. In the Tween series of surfactants, one of the polyoxyethylene residues is esterified with a fatty acid. Although the polyester cutin represents the natural substrate of cutinases, these enzymes are also known to be active as nonspecific esterases and lipases. Hydrolysis products of Tween 20 generated by cutinase are likely responsible for the toxic effect of the mixture on these pathogens. A related polyoxyethylene surfactant, Triton X-100, has no ester bonds and did not inhibit pathogen growth.

Seedborne *Phytophthora infestans*: Transmission and growth responses of different potato cultivars. H. M. Partipilo (1), M. L. Powelson (1), and D. A. INGLIS (2). (1) Dept. Bot. and Plant Path., Ore. St. Univ., Corvallis, OR 97331; (2) Research and Extension Unit, Wash. St. Univ., Mount Vernon, WA 98273. Phytopathology 92:S63. Publication no. P-2002-0458-AMA.

Seed piece to plant transmission of *Phytophthora infestans* occurred with isolates of clonal lineages US-8 in OR and with US-11 in WA. Transmission with US-8 was 2.3% for Russet Burbank (RB) and with US-11 was 0.5, 4.9, and 1.4% for RB, Russet Norkotah (RN) and Shepody (SH), respectively. With US-8, final emergence and emergence rate were significantly lower in Kennebec (KE), RB, RN, and SH compared to Bzura (BZ), and with US-11, both responses were significantly lower in KE, RN and SH compared to BZ and RB. Plant vigor (aerial biomass) of BZ and RB was significantly higher compared to KE, RN and SH; results with US-8 were inconsistent. The incidence of seed piece decay was similar for all cultivars with US-8 whereas with US-11 percent seed piece decay was significantly less with RB compared to the other four cultivars. Regardless of clonal lineage, transmission occurred more frequently and growth responses were suppressed more often in cultivars highly susceptible to the foliar phase of late blight.

Responses of potato cultivars to seedborne clonal lineages of *Phytophthora infestans*. H. M. Partipilo (1), M. L. POWELSON (1), and D. A. Inglis (2). (1) Dept. Bot. and Plant Path., Ore. State Univ., Corvallis, OR 97331; (2) Research and Extension Unit, Wash. State Univ., Mount Vernon, WA 98273. Phytopathology 92:S63. Publication no. P-2002-0459-AMA.

Responses of potato cvs Russet Burbank and Russet Norkotah grown from seed pieces infected with isolates representing the US-8 or US-11 clonal lineages of *Phytophthora infestans* were evaluated in the greenhouse. Cultivar*clonal lineage*inoculum density interactions were significant for seed piece decay, emergence rate, final emergence and aerial biomass. Russet Norkotah was equally susceptible to both pathogen genotypes. Russet Burbank was more resistant than Russet Norkotah to seedborne inoculum of US-11. Compared to Russet Norkotah, Russet Burbank showed greater percent final emergence, emergence rate and aerial biomass, and reduced seed piece decay when inoculated with US-11. Increasing inoculum density of US-11 had no effect on any of the measured response variables of Russet Burbank suggesting US-8 and US-11 may act as different races in the tuber infection biology on this cultivar.

Potential for integrated management of Sclerotinia blight on peanut with Fluazinam and the biocontrol agent *Coniothyrium minitans*. D. E. PARTRIDGE, J. E. Bailey, and D. L. Jordan. North Carolina State University, Raleigh, NC 27695. *Phytopathology* 92:S64. Publication no. P-2002-0460-AMA.

Sclerotinia blight of peanut (*Sclerotinia minor*) is an important disease that has spread to all major peanut producing counties in North Carolina. *C. minitans* is capable of colonizing sclerotia of *Sclerotinia* spp. and is currently available as a commercial formulation, ContansWG. A long-term field experiment was initiated in 1999 to test repeated soil applications of *C. minitans* at rates of 2 kg/ha and 4 kg/ha for control of Sclerotinia blight. *C. minitans* was applied in the fall of 1999 and in 2000 in a field that had been planted to cotton and harvested prior to the applications. Peanuts were planted in the spring of 2001 with chemical (fluazinam) and cultivars (susceptible NC-V11 and moderately resistant Perry) subplot treatments. Fluazinam (2.5 pt/ha) was applied according to a weather based sclerotinia advisory warning system and was found to reduce disease across both cultivars. Application of *C. minitans* at 4 kg/ha reduced disease only in the cultivar, Perry. The combination of *C. minitans* and fluazinam on Perry showed greatest control of Sclerotinia blight.

Exploring genetic diversity of *Uromyces appendiculatus* by AFLP analysis. M. A. PASTOR-CORRALES (1), J. M. Eibl (1), N. R. O'Neill (1), P. van Berkum (1), and J. R. Steadman (2). (1) USDA, ARS, Beltsville, MD 20705; (2) Dept. of Plant Pathology, University of Nebraska, Lincoln, NE. *Phytopathology* 92:S64. Publication no. P-2002-0461-AMA.

A conclusion drawn from results of several completed virulence studies is that the bean rust fungus *U. appendiculatus* is one of the most variable phytopathogens. More than 90 races have been identified based on their interaction with 19 standard bean lines. We selected 13 races, maintained at Beltsville, to characterize genome variability by AFLP analysis. Twelve races originated from the USA, two were recovered from snap beans and ten from dry beans. The remaining race was recovered from dry beans in Guatemala. Genome diversity among the 13 races was inferred from variation in fingerprint patterns. The races originating from snap beans were distinguishable from those of dry beans. Three distinct fingerprint patterns were evident among the races recovered from dry beans. The race from Guatemala and one from Florida were distinguished from each other and from the other nine races. An additional 25 races originating from dry beans grown in 12 Latin American countries were distinguished from the original 13 races, but little genetic diversity was evident among them.

Preventing pollination increases yield of cuitlacoche, *Ustilago maydis*. J. K. Pataky. Department of Crop Sciences, University of Illinois, Urbana, IL 61801. *Phytopathology* 92:S64. Publication no. P-2002-0462-AMA.

Cuitlacoche (Syn. huitlacoche) is the name the Aztecs gave to edible galls that form when kernels of *Zea mays* are infected by *Ustilago maydis*. Silk channel inoculation techniques developed to assess sweet corn reactions to common smut were used to produce cuitlacoche. Yield and quality of cuitlacoche were highest when ears were inoculated 4 to 8 days after the mid-silk growth stage and when galls were harvested 16 or 17 days after inoculation. Severity of ear galls and yield of cuitlacoche increased when pollination was prevented by detasseling a sweet corn hybrid or by growing male sterile field corn hybrids. Severity of ear galls averaged about 37% and 49% for pollinated sweet corn and field corn, respectively, and about 52% and 55% for non-pollinated treatments. Yield of cuitlacoche averaged about 37 and 72 g ear⁻¹ for pollinated sweet corn and field corn, respectively, and 63 and 101 g ear⁻¹ for non-pollinated treatments. Percent recovery (cuitlacoche yield/ear weight × 100) was about 14% in pollinated treatments and 22% in non-pollinated treatments.

Estimation of leaf wetness status using a binary logistic regression model. P. A. PAUL and G. P. Munkvold. Dept. Plant Pathology, Iowa State University, Ames, IA 50011. *Phytopathology* 92:S64. Publication no. P-2002-0463-AMA.

Leaf wetness is an important input variable used in models developed to predict plant diseases. However, leaf wetness data are often lost due to equipment failure. To complete a data set used to predict the severity of gray leaf spot of maize in Iowa, empirical models were developed and tested to estimate missing leaf wetness data. Air temperature, relative humidity, and leaf wetness were measured at 30-minute intervals using sensors and data loggers placed outside of a maize field during May – September 1998. Air temperature and relative humidity were used as predictors of leaf wetness (presence = 1 or absence = 0). Seventy-five percent of the data were used for model development and 25% for testing. Binary logistic regression, tree-

based (CART), and linear discriminant analysis (LDA) models were developed using S-Plus and compared for their accuracy in predicting leaf wetness. The logistic regression and CART models performed better than the LDA model, correctly classifying 88% of the test cases compared to 85% correctly classified by the LDA model. With this model, temperature and relative humidity may be used to estimate missing leaf wetness data.

Effect of temperature and relative humidity on sporulation of *Cercospora zae-maydis* on diseased leaves of maize. P. A. PAUL and G. P. Munkvold. Munkvold Dept. Plant Pathology, Iowa State University, Ames, IA 50011. *Phytopathology* 92:S64. Publication no. P-2002-0464-AMA.

The purpose of this study was to determine the effects of temperature and relative humidity on the production of inoculum by *Cercospora zae-maydis*, causal agent of gray leaf spot of maize, on senescent diseased leaf tissue. Incubators were used to achieve temperatures of 15, 20, and 25C, and saturated salt solutions to achieve different relative humidities between 70 and 100%. Five replicates of four relative humidity treatments were randomly assigned to each incubator. Diseased leaf blades were collected from the field and maintained in a leaf-press until they were dry. Lesions were then excised, measured, and incubated under different combinations of temperature and relative humidity. After 3 days, the leaf tissues were vortexed in a 20-ml mixture of distilled water and Tween 20, and spore production was estimated by assessing spore concentrations in the suspension. Spore production varied among and within treatments. At the temperatures used in this study, the effect of relative humidity was not significant; however, more spores were produced at 25 than at 15C.

A new species of *Pythium* from apple and wheat in eastern Washington. T. C. PAULITZ (1), K. Adams (1), M. Mazzola (2), and C. A. Levesque (3). (1) USDA-ARS, Pullman, WA 99164-6430; (2) USDA-ARS, Wenatchee, WA 98801; (3) Agriculture and Agri-Food Canada, Ottawa, ON K1A 0C6. *Phytopathology* 92:S64. Publication no. P-2002-0465-AMA.

In 2000, over 500 isolates of *Pythium* were collected from 96 sites in seven counties in the wheat production areas of eastern Washington. The ITS1 region of nuclear rDNA was sequenced from 230 isolates. An unidentified species had an ITS1 sequence that did not match any sequence from a worldwide collection of over 1200 *Pythium* species, including all the types and neotypes. This species has a unique morphology, with oospores, sporangia, and appressorial swellings formed inside appressoria. The remnants of the appressoria often remained attached to the base of sporangia and oospores. The name *Pythium abappressorium* sp. nov. is proposed. Isolates were pathogenic to wheat, causing damping-off and stunting in pasteurized soil. Isolates are identical to those described by Mazzola et al. (Plant Dis. in press) from apple roots in the Wenatchee, WA area. *P. abappressorium* was not pathogenic to apples, but was competitive in the apple rhizosphere and protected apple roots from infection by *P. ultimum* and *P. sylvaticum*.

Variation in severity of *Monilinia vaccinii-corymbosi* infection among lowbush blueberry clones. L. N. PENMAN and S. L. Annis. Dept. of Biological Sciences, University of Maine, Orono, ME 04469. *Phytopathology* 92:S64. Publication no. P-2002-0466-AMA.

Monilinia vaccinii-corymbosi is a major fungal pathogen of *Vaccinium* species and infects its host by primary ascospore infection of developing tissues and secondary conidial infection of ovaries. Field observations suggested that phenotypically-diverse lowbush blueberry clones exhibit different levels of infection. 14 blueberry clones in 2 commercial fields were evaluated by marking stems (10 with and 10 without primary infection) within each clone and measuring the percent infection of leaves, flowers, and fruit on these stems 3 times during the growing season. We also determined the percent yield ((fruit number/flower number) × 100 percent) and average berry weight of each stem. The severity of both primary and secondary infection was significantly different among clones. We found that secondary infection and percent yield were not correlated with primary infection when comparing all stems. However, average berry weight was inversely correlated with primary infection. We plan to examine clonal morphology and phenology of shoot and floral bud break to evaluate whether disease severity is correlated with these traits.

PFDFAD, an expert system for timing fungicide applications for control of postbloom fruit drop. N. A. R. PERES (1), S. Kim (2), H. W. Beck (2), N. L. Souza (1), and L. W. Timmer (3). (1) Sao Paulo State Univ., Botucatu, Brazil; (2) Univ. Florida, Gainesville; (3) Univ. Florida, CREC, Lake Alfred. *Phytopathology* 92:S64. Publication no. P-2002-0467-AMA.

Postbloom fruit drop (PFD), caused by *Colletotrichum acutatum*, affects flowers and produces abscission of young fruitlets. Since a predictive model developed in Florida to time fungicide applications was not adequate in other regions, an expert system with broader applicability was developed and implemented in Java. The PFD-FAD system considers the previous history of PFD, varietal susceptibility, the bloom stage as well as rainfall, leaf wetness following the rain, and the current inoculum levels in the grove. It predicts the need for a fungicide application based on these factors and the time since the last spray. Several methods for timing of fungicide applications were evaluated in Brazil. Calendar timing, the grower's program, the PFD model, and the PFD-FAD system resulted in 2, 3, 2, and 1 spray, respectively. All programs were similar in effectiveness and reduced disease and increased yields. The PFD-FAD system is easier to use and minimizes the need for scouting and precise weather information compared to the model.

Hop disease surveys in Argentina. B. A. PEREZ (1), D. Barreto (1,2), E. Martinez (3), and A. Leibrecht (4). (1) INTA, IMYZA, 1712 Castelar; (2) Buenos Aires Univ.; (3) INTA, AER El Bolson; (4) Camara Lupulera, Rio Negro. Phytopathology 92:S65. Publication no. P-2002-0468-AMA.

In South America, hop (*Humulus lupulus* L.) is grown in southwestern Argentina. Disease surveys were initiated in winter 2001 due to lower cone yields from cultivars that previously had very good performance. Diseased rootstocks, bines, leaves and cones were studied through the season. In winter, *Fusarium solani*, and *Rhizoctonia solani* were associated with rotted and discolored rootstocks. Through spring and summer, microscopic examinations of the leaves revealed *Pseudoperonospora humuli* in stunted plants. Other fungi were: *Verticillium* in plants with yellow and necrotic leaf patches, and *Alternaria alternata* in leaf, bine and cone spots. In bines with die-back, the only fungi detected were *F. solani* and *A. alternata*. The surveys indicated the imminent need for an integrated disease management.

Genetic differentiation of *Phoma ligulicola* isolates from pyrethrum in Tasmania. S. J. PETHYBRIDGE (1), J. S. Scott (1), K. Harrison (1), F. S. Hay (1), and T. Groom (2). (1) University of Tasmania - North West Centre, Burnie Tas 7320, Australia; (2) Botanical Resources Australia Pty. Ltd., Ulverstone Tas 7315, Australia. Phytopathology 92:S65. Publication no. P-2002-0469-AMA.

Phoma ligulicola is the most severe fungal pathogen affecting pyrethrum crops in Tasmania, Australia. It causes the disease, ray blight, which is associated with necrotic lesions on leaves, at nodes on stems, and a failure of the flower to develop causing a decrease in yield and pyrethrin content in the flowers. The internal transcribed spacer regions (ITS1 and ITS 2) of the ribosomal DNA genes from 11 isolates of *P. ligulicola* were amplified using the polymerase chain reaction (PCR) and sequenced. Phylogenetic trees were produced by combining the sequences of the ITS regions and comparison to the published sequences of other *Phoma* spp. and other closely related fungi. Isolates were differentiated into three subgroups, P1, P2, and P3 through phylogenetic analysis. The closest match to the ITS sequences of other fungi were *Phoma herbarum* (86.5 – 91.5% homology) and *Didymella bryoniae* (88.2 to 93.6% homology). Primers were designed to the ITS regions that could be used to detect all three subgroups. Further work will concentrate on the biological implications of this molecular diversity and the development of a nested PCR test for the simultaneous characterisation of the three subgroups.

Local focus expansion rates in grass stem rust estimated with nonparametric spatial analysis. W. F. PFENDER and G. Whittaker. USDA-ARS NFSPRC. Phytopathology 92:S65. Publication no. P-2002-0470-AMA.

Epidemics of stem rust (*Puccinia graminis* subsp. *graminicola*) in perennial ryegrass (*Lolium perenne*) begin with numerous small foci. The dynamics of early development in foci is important because fungicides typically are applied to this crop when foci are small. We studied focal expansion in 2m x 2m plots, with readings taken in square, 0.15m grids 2 times per week for several weeks. Observations were taken from a suspended platform to reduce artificial dispersal of spores. Data were analyzed with multivariate nonparametric regression, which makes no assumptions about the functional form of the disease gradient. Rates for isopath movement and for increase in area bounded by an isopath were made. Speed of isopath movement ranged from 1 to 15 cm/day, and did not necessarily increase with distance from the focal center in early stages of the focus development. Expansion rate variations were partially explained by weather parameters, including temperature, leaf wetness and wind direction. The approach should be useful in spatial simulation studies of epidemic development.

Functional and sequence comparisons of *Brassica* FLS2 homologues and their elicitors. C. PFUND, F. M. Dunning, and A. F. Bent. Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S65. Publication no. P-2002-0471-AMA.

One means of pathogen recognition is the interaction of pathogen elicitors with plant proteins. We seek to understand the determinants of these interactions. We are studying the specificity determinants for the interaction between FLS2, a leucine-rich repeat (LRR) receptor-like kinase, and its elicitors. A peptide based on a conserved domain of bacterial flagellin proteins, flg22, triggers FLS2-mediated growth/defense responses in Arabidopsis (Gomez-Gomez, 2000). These workers also reported elicitation of Arabidopsis by crude flagellin extracts from many bacteria, but not from extracts of two *Xanthomonas campestris* pathovars. In contrast, we observed both weak and strong responses using extracts from 14 *X. campestris* pv. *campestris* (*Xcc*) strains. We have also identified homologues of FLS2 in several species of *Brassica* and plan to characterize their phenotypic and molecular responses to *Xcc* strains and flg22. We will compare sequences of *Xcc* flagellin genes and the LRRs of FLS2 homologues. These analyses will highlight residues in both the elicitor and putative R protein that determine interaction specificity

RAPD markers for the *Cr* gene for resistance to *Cronartium ribicola* in *Ribes nigrum*. D. D. PICTON (1), K. E. Hummer (2), and J. D. Postman (2). (1) Department of Horticulture, Oregon State University; (2) USDA-ARS National Clonal Germplasm Repository, Corvallis, OR. Phytopathology 92:S65. Publication no. P-2002-0472-AMA.

White pine blister rust, caused by *Cronartium ribicola* Fisch., is an exotic disease from Asia. This disease was introduced into North America about 1890 on infected five-needled pines (*Pinus* subsect. *Strobus*). Legislation in some eastern states restricts *Ribes* L., the currants, which co-host the disease. The dominant *Cr* gene from *R. ussuriense* Jancz. confers immunity and has been used to breed rust-immune black currants. Our objective was to find RAPD markers for this gene. We examined 180 'Ben Lomond' x 'Consort' (susceptible x heterozygous resistant) seedlings. 50 RAPD primers generated an average of 4.7 products with 1.5 polymorphic products (31.9% of polymorphism). The markers are linked to the *Cr* gene with an average genetic distance of 30 cM. The low genetic variability determined with RAPD's has not permitted a highly resolved map at this time. However, this initial map could be developed with additional molecular and morphological markers and used for marker-assisted selection or to screen cultivated or wild currants for rust-immunity.

Epidemiology, detection, and management of tomato ringspot virus and *Xiphinema americanum* in red raspberry. J. N. PINKERTON, J. Kraus, and R. R. Martin. USDA ARS HCRL, 3420 NW Orchard Ave., Corvallis, OR 97330. Phytopathology 92:S65. Publication no. P-2002-0473-AMA.

Population densities of *Xiphinema americanum* were monitored monthly for three years in plots established in an infected red raspberry field. The ability of the nematode to vector tomato ringspot virus (ToRSV) was assayed with cucumber seedlings planted in soil collected from each of 12 blocks. Cucumber leaves were assayed for ToRSV by ELISA. Nematode population densities were greatest in the winter and early spring, with lowest densities observed in the summer. Conversely, virus was not detected in assay plants in the winter while virus infection was greatest in the summer. Crop rotations were evaluated for the control of the nematode and ToRSV. Eighteen months cropping with rapeseed or clean fallow significantly reduced nematode densities as did soil fumigation, while cropping with fescue and raspberry did not. Raspberries were planted in the cover crop plots and tested for ToRSV after 12 months. ToRSV was not detected in raspberry leaves from plants in any treatment. A reverse transcriptase-polymerase chain reaction was developed to detect ToRSV in nematodes.

Functional analysis of the 3'-termini of avirulence genes from two *Xanthomonas* species. G. PONCIANO (1), H. Ishihara (2), S. Tsuyumu (2), and J. E. Leach (1). (1) Dept. Plant Pathology, Kansas State University, Manhattan, KS 66506, USA; (2) Faculty of Agriculture, Shizuoka University, Shizuoka 422-8529, Japan. Phytopathology 92:S65. Publication no. P-2002-0474-AMA.

Many *avr/pth* gene family members in *Xanthomonas* species exhibit both avirulence and virulence functions. Gene structural features important for these functions include a central region of multiple repeated sequences essential for host cultivar specificity, and three nuclear localization signals and an acidic activation domain (AAD) at the C-terminus necessary for avirulence and virulence function. Using gene chimeras constructed with portions of *avrXa10* and *avrXa7* from *Xanthomonas oryzae* pv. *oryzae* and

apl1 and *apl2* from *Xanthomonas axonopodis citri*, we identified a 417 bp *HincII* and *SphI* fragment in the 3' region that contains a putative leucine zipper, and is required for avirulence to rice and virulence to rice and citrus. Together with the requirement for nuclear localization signals and the AAD, our data are consistent with a role for the *avr/pth* gene family members in host plant transcriptional activation.

Influence of soil moisture on severity of common scab of potato (*Streptomyces scabies*). E. C. POOLE (1), D. S. Douches (2), W. W. Kirk (1), and R. Hammerschmidt (1). (1) Dept. Plant Pathology; (2) Dept. Crop and Soil Sciences, Michigan State University, East Lansing, MI 48824. Phytopathology 92:S66. Publication no. P-2002-0475-AMA.

Potato scab, caused by *Streptomyces scabies*, is the fourth most important potato disease. Tubers are infected through stomata and immature lenticels yet to form a protective barrier; therefore, they are most susceptible during the early stages of development. Maintaining adequate soil moisture during tuber development and the use of resistant varieties are the most effective control management strategies. The objective of this study was to investigate the interaction between cultivar resistance and soil moisture on scab incidence and severity. Greenhouse experiments with boxes filled with sand were conducted to evaluate the effects of four moisture regimes and three varieties differing in susceptibility to scab development. The higher soil moisture treatments reduced scab infection on the susceptible cultivar. Quantification of *S. scabies* populations is in progress and may enable determination of the inoculum requirements for potato scab infection.

Bacillus strains that have multiple mechanisms for antagonism of *Xanthomonas*. A. R. POPLAWSKY and W. Chun. Dept. PSES, University of Idaho, Moscow, ID 83844. Phytopathology 92:S66. Publication no. P-2002-0476-AMA.

Bacillus strains isolated from cauliflower leaves induced two zones of inhibition against *Xanthomonas campestris* pv. *campestris* (*X.c.c.*, causal agent of black rot of crucifers) on agar medium. In the smaller zone A, there was no growth of the pathogen. In the larger zone B, *X.c.c.* grew but was reduced in pigment (Pig), and extracellular polysaccharide (EPS), production. However subculturing of these bacteria resulted in colonies that were restored to Pig⁺, EPS⁺. Occasional *X.c.c.* colonies appearing resistant to the zone A inhibition were subcultured, and they were Pig⁺, EPS⁺, and stably resistant. When occasional yellow sectors from zone B were subcultured, they were Pig⁺, EPS⁺, and resistant to zone B inhibition. Since no *X.c.c.* mutants were resistant to both kinds of inhibition, and different *X.c.c.* mutants were generated in each zone, two mechanisms are probably involved. The *X.c.c.* DF signal is required for pigment and EPS production, as well as epiphytic survival and host infection. Since *Bacillus* antagonism and *X.c.c.* resistance appear to involve *X.c.c.* pigment and EPS production, these *Bacillus* strains may inhibit the production or activity of the *X.c.c.* DF signal.

***Xanthomonas campestris* pv. *campestris* extracellular polysaccharide is required for natural host infection as well as virulence.** A. R. POPLAWSKY and W. Chun. Dept. PSES, University of Idaho, Moscow, ID 83844. Phytopathology 92:S66. Publication no. P-2002-0477-AMA.

Xanthomonas campestris pv. *campestris* (*X.c.c.*, causal agent of crucifer black rot) produces a diffusible butyrolactone signal (DF). DF⁻ mutant strains are impaired in xanthomonadin pigment and extracellular polysaccharide (EPS) production, epiphytic survival, and natural host infection via hydathodes. Previous work showed that DF was required for epiphytic survival because the pigment plays a role in survival. However, it was not known whether DF is required for host infection because EPS plays a role in infection, or for some other, unknown reason. Compared to *X.c.c.* parent strain FC2, strain XcD (mutated in the first step specific for EPS biosynthesis) is 50% reduced in virulence using artificial infection (Katzen et al., J. Bacteriol. 180:1607). In the present study, XcD and FC2 were similar in DF and pigment production, as well as epiphytic survival. However XcD was completely deficient in EPS production, and with natural infection averaged 0.0 lesions/plant compared to 2.4 for FC2. Thus, EPS plays an important role in host infection as well as virulence, and this is probably why DF is required for host infection.

Survival of sporangia and zoospores of *Phytophthora infestans* in surface water. L. D. PORTER and D. A. Johnson. Dept. Plant Pathology, Washington State Univ., Pullman, WA 99164. Phytopathology 92:S66. Publication no. P-2002-0478-AMA.

Surface water in potato fields may provide a medium for survival and dispersal of *Phytophthora infestans*. Petri plates with water suspensions

(120mls water/plate) of sporangia and zoospores (100,000 sporangia/plate) were embedded in sandy soil to quantify longevity of spores in water under natural conditions in Eastern WA in July and October, 2001. Effects of light intensity, presence or absence of soil in petri plates (15gm/plate), spore type, and isolate (US-8 or US-11) on survival were investigated. Spores in water survived 8-16 days in direct sunlight and 13-20 days in shade. Addition of soil to the water significantly increased time of spore survival. In July the maximum survival of spores in water exposed to direct sunlight, with no soil, was 1 to 2 days; in October it was 6 to 8 days. Survival was 3-8 days longer in direct sunlight when soil was added to the water. Zoospores and sporangia did not significantly differ in total time of survival but fewer zoospores than sporangia survived for extended periods. Spores of US-11 and US-8 isolates did not differ in time of survival but sporangia of US-11 survived in significantly greater numbers (*P* equals 0.05).

Expressed sequence tags analysis from germinating urediniospores of the plant pathogen *Phakopsora pachyrhizi*. M. L. POSADA and R. D. Frederick. USDA-ARS Foreign Disease-Weed Science Research Unit, 1301 Ditto Ave., Fort Detrick, MD 21702. Phytopathology 92:S66. Publication no. P-2002-0479-AMA.

Rust on soybean is caused by the obligate fungal pathogen *Phakopsora pachyrhizi*. A unidirectional cDNA library was constructed using mRNA isolated from urediniospores germinating on a water surface. Single pass sequencing of 908 clones revealed that 398 sequences displayed significant similarities to sequences deposited in public databases. The remaining 510 sequences showed weak or no similarities to database entries. They were queried against the NCBI dbEST using the BLASTN algorithm and 40 sequences revealed high or moderate similarities to plant and fungal sequences. 488 Unique ESTs were identified of which 108 appeared as multiple copies. Among genes with assigned function, approximately 13.6% were involved in primary metabolism, 7.3% in protein metabolism, 2.5% in RNA metabolism, 6.3% in cell structure, 3.5% in growth and morphology, 2.8% in stress response, and 1.0% in cell division. Approximately 56.5% of the identities found were to hypothetical proteins and proteins with unknown function.

Phylogenetic studies of corn and rice strains of *Acidovorax avenae* subsp. *avenae* by DNA/DNA hybridization. E. Postnikova and N. W. SCHAAD. USDA ARS Foreign Disease-Weed Science Research Unit, Fort Detrick, MD 21702. Phytopathology 92:S66. Publication no. P-2002-0480-AMA.

Acidovorax avenae subsp. *avenae* (Aaa) is the causal agent of diseases of several important economic crops, including bacterial streak of corn (*Zea mays*) and bacterial stripe of rice (*Oryza sativa*). To determine the phylogenetic relationship of these two pathogens, a highly reproducible S1 exonuclease DNA/DNA hybridization technique was used. DNA was purified by a modified Marmur method and only DNA with an 260/280 absorbance ratio of 1.8 or greater was used. DNA of the neopathotype strain from corn (ATCC 19860) and a rice strain (ATCC 19882) were labeled with ³³P. Unlabelled DNA was used from six strains from each of corn and rice of different origins, including the United States, India, Japan, and Nigeria. Homologies between strains originating from the same two hosts were very high; 95-99 percent for corn and 87-97 percent for rice. Comparisons of strains between the two pathogenic groups revealed a mean homology of 45 percent (range 42 - 58). Based on a species requirement of 70 percent homology or greater, these results show that the *A. avenae* pathogens from corn and rice should be reclassified as separate species.

An excised-leaf inoculation technique for evaluating host-pathogen interactions of dematiaceous hyphomycetes on bermudagrass. R. G. Pratt. USDA, ARS. Phytopathology 92:S66. Publication no. P-2002-0481-AMA.

An excised-leaf inoculation technique was developed to evaluate interactions of species of *Exserohilum*, *Bipolaris*, *Curvularia*, and *Drechslera* with bermudagrass. Sections of leaf blades from near growing stem tips were excised, placed on water agar, and inoculated with mycelium of pathogens at distal edges. Symptoms were evaluated according to the extent of necrosis that developed down the leaf sections after 5 days. Symptom severity increased in leaves progressively distal from growing stem tips. Fewer differences in virulence of pathogens were evident in excised leaf tissues than in foliage inoculated with spores. In repeated experiments, significant differences in extent of necrosis were observed among 40 randomly selected genotypes of bermudagrass when excised leaf sections were inoculated with *E. rostratum*. It is suggested from these results that the excised-leaf inoculation technique may be used to identify and compare quantitative host resistance to dematiaceous hyphomycetes in individual genotypes of bermudagrass.

Role of biofilms in biocontrol of *Botrytis cinerea*. C. M. PRESS, W. F. Mahaffee, M. M. Roche, T. M. Sechler, and A. L. Davis. USDA-ARS, HCRL, Corvallis, OR 97330. Phytopathology 92:S67. Publication no. P-2002-0482-AMA.

Microorganisms often inhabit the leaf surface in organized structures termed biofilms. *Burkholderia cepacia*, FP62 is a biocontrol agent of *B. cinerea* in geranium and forms extensive biofilms in the phyllosphere. Scanning electron micrographs demonstrate extensive phyllosphere colonization (60-70% of the leaf surface). FP62 biofilms appeared to be many cells layers thick and enveloped in a polymer-like matrix. The biofilm phenotype of this strain is related to biocontrol. Isolation of transposon mutants that are deficient in biofilm formation in an in vitro biofilm assay, also lacked the capacity to control *B. cinerea* when applied to geranium leaves. The biofilm mutants are less efficient in phyllosphere colonization lacking many of the characteristics of wild-type biofilms. The biofilm mutation and biocontrol capacity could be restored through the addition of exogenous polymer to the biocontrol formulation of the mutants. The addition of polymers to the formulation of several other biocontrol agents also improved their biocontrol capacity suggesting that biofilms contribute to biocontrol efficacy and are an important aspect of phyllosphere competence.

Reconciling meiotically and molecularly defined fumonisin biosynthetic genes in *Gibberella moniliformis*. R. H. Proctor, A. E. DESJARDINS, and R. D. Plattner. USDA-ARS-NCAUR, Peoria, IL 61604. Phytopathology 92:S67. Publication no. P-2002-0483-AMA.

Gibberella moniliformis (anamorph *Fusarium verticillioides*) is one of the most common ear and stalk rot pathogens of maize and can produce carcinogenic mycotoxins called fumonisins. Previous meiotic analyses of *G. moniliformis* identified three tightly linked fumonisin biosynthetic loci: the *Fum1* locus confers ability to produce fumonisins and the *Fum2* and *Fum3* loci confer ability to hydroxylate fumonisins at carbons 10 and 5, respectively. The goal of this study was to determine whether these meiotically defined *Fum* loci correspond to any of the 15 molecularly defined *FUM* genes in a recently identified fumonisin biosynthetic gene cluster. Linkage and transformation-mediated complementation analyses revealed that *Fum1* and *FUM5*, which encodes a polyketide synthase, are the same gene, now designated *FUM1*. In addition, *Fum2*-defective strains and a disruption mutant of *FUM12*, which encodes a putative hydroxylase, have the same fumonisin production phenotype. Therefore, *Fum2* and *FUM12* are most likely the same gene, now designated *FUM2*. These results indicate that the *Fum* loci are located within the fumonisin gene cluster.

Acidification of host tissue by *Penicillium* spp. as a mechanism to increase virulence. D. PRUSKY (1), J. L. McEvoy (2), R. A. Saffner (2), and W. S. Conway (2). (1) Dept. of Postharvest Science of Fresh Produce, ARO, Bet Dagan 50250, Israel; (2) USDA, ARS, PQSL, 10300 Baltimore Ave. Beltsville, MD 20705. Phytopathology 92:S67. Publication no. P-2002-0484-AMA.

The phytopathogenic fungi *Penicillium expansum*, *P. digitatum* and *P. italicum* are among the major postharvest pathogens attacking deciduous and tropical fruits. *Penicillium* spp. acidify the ambient environment of apple and citrus fruits during decay development. They utilize two mechanisms of acidification: fungal organic acid production and fungal NH_4^+ influx/ H^+ efflux. Organic acids were detected when *P. expansum* and *P. digitatum* decayed apples and citrus fruits. *P. expansum* decayed apple tissue of different cultivars associated with a reduction of NH_4^+ content and fruit tissue pH. Transcripts encoding the endopolygalacturonase gene *pepg1* from *P. expansum* accumulated under acidic culture conditions while no accumulation was observed under neutral conditions. Acidification of the tissue by organic acid treatment significantly enhanced pathogenicity. The results indicate the importance of the acidification mechanism as a virulence factor enhancing pathogenicity of *Penicillium* spp.

Effects of Messenger® on disease resistance and plant growth enhancement in strawberry and cucumber. D. QIU, K. Clayton, and Z.-M. Wei. EDEN Bioscience Corporation, Bothell, WA. Phytopathology 92:S67. Publication no. P-2002-0485-AMA.

Messenger is a biopesticide containing 3% active ingredient harpin protein. Harpin is a proteinaceous hypersensitive response elicitor isolated from *Erwinia amylovora*. Previous studies have indicated that when applied to plants, the harpin protein is recognized and triggers a complex set of signaling pathways that contribute to an overall acquired disease resistance in the plant. Plants treated with Messenger also demonstrate an increase in yield and a general plant growth enhancement effect. In recent studies, strawberry plants (Diamonte) were treated with Messenger at rates of 0, 1, 5,

10, 20, 40, 80 and 120 mg/ml. After Messenger treatment, the plants were inoculated with powdery mildew (*Sphaerotheca macularis* f. sp. *Fragariae*) or *Xanthomonas fragariae*. Disease resistance was determined by using a disease severity index. Messenger spray treatments substantially induced resistance in strawberries against strawberry powdery mildew and *Xanthomonas fragariae*. In separate studies, treatments of 10-40 mg/ml Messenger were shown to be sufficient to induce increases, over control plants, of 10 to 13% with respect to cucumber (Park's All Season Burpless) plants height and seedling dry weight; as well as strawberry plants height and leaf number.

Virus induced gene silencing in protoplasts. W. P. QIU and H. S. Hou. Department of Fruit Science, Southwest Missouri State University, Mountain Grove, MO 65711. Phytopathology 92:S67. Publication no. P-2002-0486-AMA.

Virus induced gene silencing (VIGS) is a defense response targeting on RNA in plant cells upon infection by a virus. In the previous study, a green fluorescence protein (GFP) gene was inserted into a *Tomato bushy stunt virus* (TBSV) vector with an inactivated p19 gene (pHST2-14) to create a recombinant virus pTG. The same GFP gene was silenced in the pTG-infected GFP transgenic *Nicotiana benthamiana* plant (NbGFP) while the co-expression of the TBSV p19 protein (P19) suppressed silencing. In this study, we analyzed silencing of the GFP gene in NbGFP protoplasts. Protoplasts were transfected with transcripts derived from pTG, pTG-p19/wt in which the p19 gene was restored, and pHST2-14. Total RNAs were extracted at 22 hours post transfection and RNA blots were performed. The GFP mRNA level was reduced in pTG-transfected protoplasts, and also in protoplasts transfected by pTG-p19/wt that expresses the P19 compared to the pHST2-14- or mock-transfected control. These results suggested that GFP gene silencing was triggered in NbGFP protoplasts by the TBSV-expressed GFP gene, and the P19 does not appear to interfere with this process within 22 hour.

Molecular analysis of European and North American isolates of *Spongopora subterranea* f. sp. *subterranea*, the cause of powdery scab of potatoes. X. S. QU and B. J. Christ. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802. Phytopathology 92:S67. Publication no. P-2002-0487-AMA.

Spongopora subterranea f. sp. *subterranea* is a soil-borne biotrophic pathogen that causes powdery scab of potato. Powdery scab seriously reduces tuber quality and marketability and is a major concern to potato growers worldwide. Genetic diversity among 51 isolates of *S. subterranea* from different geographic regions in Europe and North America was assessed by rDNA sequence analysis. Two distinct genetic groups (I and II) were identified based on the internal transcribed spacers (ITS1 and ITS2) sequence diversity among the isolates. Genetic group I represented 35.3% and genetic group II represented 64.7% of all isolates tested. European isolates occurred in genetic groups I and II, whereas North American isolates belonged only to genetic group II. The European genetic groups of *S. subterranea* are associated with particular potato cultivars. It is possible that *S. subterranea* genetic groups are different pathotypes but this has yet to be established.

Functional analysis of RPS2 interaction with putative partners in disease resistance signal transduction. B. F. Quirino, J. H. Ham, B. B. Bonacci, and A. F. BENT. Dept. of Plant Pathology, University of Wisconsin-Madison, WI 53706. Phytopathology 92:S67. Publication no. P-2002-0488-AMA.

To dissect the molecular basis of defense activation in gene-for-gene resistance, we have studied the interaction between RPS2 of Arabidopsis and *avrRpt2* of *Pseudomonas syringae* pathovars. The RPS2 product belongs to the CC-NBS-LRR class of R gene products that carry a coiled coil (leucine zipper), a nucleotide binding site, and leucine-rich repeats. A modified yeast-two-hybrid system was used to identify proteins that interact with RPS2, and to evaluate those interactions. We identified Arabidopsis proteins that interact with RPS2 in the yeast two-hybrid system in the presence but not in the absence of AvrRpt2. RPS2 domains and full-length RPS2 carrying NBS or LRR mutations are being tested for interaction with these newly identified interactors. Related studies examine direct interaction between AvrRpt2 and RPS2 or the interactors. Disease resistance phenotypes are being tested in plants with disrupted expression of the interactors. These interactor proteins may represent virulence targets of AvrRpt2 and/or proteins that participate in plant defense signal transduction.

Reaction of soybean to a Wisconsin field isolate of *Tobacco streak virus*. P. F. RABEDAUX and C. R. Grau. University of Wisconsin-Madison. Phytopathology 92:S67. Publication no. P-2002-0489-AMA.

Tobacco streak virus (TSV) is increasingly becoming an economic threat to the soybean crop in the North Central Region of the U.S. Twenty different lines of soybean were challenged with a Wisconsin field isolate of TSV (TSV-C) under greenhouse conditions. Fully expanded unifoliolate leaves were mechanically inoculated. Symptoms were rated on a scale of 0-3, with 0 representing an asymptomatic plant, and 3 showing severe symptoms. A split-plot design was used for the study and the data were analyzed using the SAS system. Intraspecific variation of symptoms associated with infection with TSV-C was observed. Significant ($P < 0.0001$) differences were found in symptom severity among soybean lines. Lines showing the highest rating, 3.00, include Colfax, 548-380 and M90-135046; lines with the lowest rating, 0.33, include Parker, 153-282 and M93-149112. This is the first report of putative resistance to TSV in soybean. Crosses will be performed and analyzed for segregation of putative resistance loci to TSV-C.

Phylogeny of hydrogen cyanide synthase biosynthetic genes in biocontrol fluorescent pseudomonads. A. Ramette (1), M. Frapolli (1), Y. MOËNNE-LOCCOZ (2), and G. Defago (1). (1) Phytopathology group, Institute of Plant Sciences, ETH, Zürich, Switzerland; (2) UMR CNRS Ecologie Microbienne, Université Claude Bernard (Lyon 1), 69622 Villeurbanne, France. *Phytopathology* 92:S68. Publication no. P-2002-0490-AMA.

The production of hydrogen cyanide (HCN), which requires a hydrogen cyanide synthase, is implicated in the biocontrol activity of certain root-colonizing pseudomonads against soil-borne plant pathogens. Phylogenetic analysis of HCN biosynthetic genes was done using a worldwide collection of HCN-producing biocontrol fluorescent pseudomonads. Partial *hcnBC* sequences (587 bp) were amplified and sequenced. Cluster analysis of DNA and deduced protein sequences showed the existence of four main Hcn groups, each of them containing isolates originating from different countries or host plants. 16S rDNA and *hcnBC* phylogenies for representative strains revealed the presence of different *Pseudomonas* species within each Hcn group. These Hcn groups could be differentiated based on the quantity of HCN produced in vitro by individual strains, which has implications for biocontrol.

Molecular basis of Verticillium wilt: From Arabidopsis to crop plants. P. RAUYAREE (1), S. Kang (1), M. Ospina-Giraldo (1), R. G. Bhat (2), K. V. Subbarao (2), and K. Dobinson (3). (1) Dept. Plant Pathology, Penn State, University Park, PA 16802; (2) Dept. Plant Pathology, UC-Davis, Salinas, CA 93905; (3) Agriculture & Agri-Food Canada, London N5V 4T3, Canada. *Phytopathology* 92:S68. Publication no. P-2002-0491-AMA.

Verticillium dahliae is a soil-borne fungus that causes vascular wilt in over 250 plant species, many of which are economically important crops. Despite its economic importance, there have been limited studies on the mechanisms of its pathogenicity. The main objective of this project is to provide such knowledge through the use of a new model system, based on *Arabidopsis thaliana* as the principal host. Given the extensive data and resources currently available for *A. thaliana*, we have focused on characterizing the mechanisms of pathogenicity in *V. dahliae* by determining the functions of its genes that are homologous to known or suspected pathogenicity genes of other fungal pathogens. To test the role of these genes in pathogenicity, we have mutagenized them using a technique based on *Agrobacterium tumefaciens*-mediated transformation. The degree of virulence and other phenotypic changes in the resulting mutants will be presented.

Genes expressed during the resistance response to *Mycosphaerella graminicola*. S. Ray (1), S. B. Goodwin (2), and J. M. ANDERSON (2). (1) Purdue University; (2) USDA-ARS. *Phytopathology* 92:S68. Publication no. P-2002-0492-AMA.

Mycosphaerella graminicola is a widespread and important pathogen of wheat. Differential display experiments to compare gene expression in inoculated resistant (*Tadina* harboring *Stb4*) and susceptible (*Yecora Rojo*) wheat lines identified a putative differentially expressed gene with significant homology to protein disulfide isomerase (PDI). The association between PDI expression and the defense response in wheat to *M. graminicola* was examined in another resistant wheat line that contained *Stb3*. Treatments consisted of the resistant line inoculated with water (control) or *M. graminicola* (treated) and leaf tissue was collected at 0, 1, 3, 6, 12, 24 and 96 hrs after inoculation. Real-time quantitative PCR showed that PDI was expressed at very low levels in control tissue at all time points whereas in treated tissue it was highly induced at 3 hrs and 6hrs and by 24 hrs was at the preinduced level. In comparison, known defense response genes (PR-1, beta 1,3-endoglucanase, and thaumatin) showed rapid induction at 6 hrs only in the treated samples, reached a maximum level at 12 hrs and then gradually declined.

Maize necrotic streak virus is most closely related to members of the genus tobusvirus. M. G. REDINBAUGH (1) and K. Scheets (2). (1) USDA-ARS, Wooster, OH; (2) Dept. Botany, Oklahoma State University, Stillwater, OK. *Phytopathology* 92:S68. Publication no. P-2002-0493-AMA.

Initial reports indicated that *Maize necrotic streak virus* (MNeSV) is most closely related to viruses in the family *Tombusviridae*. The two 5'-most open reading frames (ORFs) are most similar to the corresponding tombusvirus proteins, while the 27.4 kDa coat protein (CP) is more closely related to the *Tobacco necrosis virus* CP. In this study, the complete sequence of MNeSV (4094 nt) was shown to be significantly smaller than the typical 4.7 kb tombusvirus genome. The 122 nt 5' untranslated region (UTR) could fold into a stable T-shaped domain. Overlapping ORFs at the genome's 3' end encoded proteins of 21.4 kDa and 19.0 kDa similar to the tombusvirus p21 and p19, respectively. The 3' UTR was 196 nt long, and did not contain the small ORF (pX) found in some tombusviruses. Three sgRNAs of 1607, 781 and 190 nt were found. MNeSV vRNA replicates readily in Black Mexican Sweet maize protoplasts facilitating future studies.

Diseases of shade-grown medicinal herbs. R. D. Reeleder. Agriculture and Agri-Food Canada, London, ON N5V 4T3. *Phytopathology* 92:S68. Publication no. P-2002-0494-AMA.

Goldenseal (*Hydrastis canadensis*) and black cohosh (*Cimicifuga racemosa*) are medicinal herbs that are indigenous to North America. Both are potential companion or rotation crops in shade-cultivated ginseng (*Panax quinquefolius*) production systems. The susceptibility of *H. canadensis* and *C. racemosa* to two serious root diseases of *P. quinquefolius* was determined. Plants of the three species were grown in a shaded greenhouse and inoculated with isolates of *Phytophthora cactorum* or *Cylindrocarpon destructans* known to be virulent to ginseng. *P. quinquefolius* plants were severely diseased or killed by both pathogens. However, neither fungus was pathogenic on *H. canadensis* or *C. racemosa*. In related work, local goldenseal crops were observed to be severely affected by root disease. *Cylindrocladium colhounii* was isolated from diseased roots and shown to be pathogenic on *H. canadensis* plants. The fungus was not, however, pathogenic on *P. quinquefolius*. As both *H. canadensis* and *C. racemosa* are unaffected by two widespread root diseases of ginseng, further studies on their potential cultivation with ginseng is merited.

Use of a commercial paint shaker to extract fungal DNA from soil. R. D. REELEDER (1), B. Capell (1), L. Tomlinson (1), J. Miller (1), and W. Hickey (2). (1) Agriculture and Agri-Food Canada, London, ON N5V 4T3; (2) Dept. Soil Science, University of Wisconsin, Madison, WI 53706. *Phytopathology* 92:S68. Publication no. P-2002-0495-AMA.

A bead homogenization procedure was used in conjunction with a commercial paint shaker to extract DNA from multiple large soil samples. Platforms designed to accommodate eight 50-ml Oakridge tubes were constructed and fitted to the shaker clamps. Soil samples (5-10 g) infested with chlamydozoospores of the fungus *Cylindrocarpon destructans* were used to evaluate efficiency in fracture of spores and release of DNA. Glass and zirconium oxide beads of various diameters were compared for DNA yield and reduction of colony-forming-units (CFU) on agar media after shaking treatments. Shaking for 20 min with 1-mm diam zirconium oxide beads yielded the largest amount of DNA and provided fungal CFU reductions of 98 percent. Effects of various DNA extraction solution components (calcium chloride, proteinase K, sodium dodecyl sulfate) were evaluated for their effects on DNA yield and on amplification with the polymerase chain reaction (PCR). Generally, minimum concentrations of the components maximized yield. Their impacts on PCR amplification were in some cases concentration dependent.

A method for screening prunus cultivars for resistance to gummosis, shot hole and rust. C. C. REILLY and T. G. Beckman. Southeastern Fruit and Tree Nut Research Laboratory, USDA-ARS, Byron, GA. *Phytopathology* 92:S68. Publication no. P-2002-0496-AMA.

A trellis system was established consisting of 4 rows, 46.2 m long, with 6.8 m middles. Support braces 1.9 m tall and 1.2 m wide (6 per row) were used to suspend 3 steel wires and steel fencing (5 cm x 10 cm x 1.2 m) the length of each trellis. A mist system, controlled by an electric timer, was placed on the wire with emitters at 1.7 m intervals. Eight replicates of the 25 most common peach cultivars in Georgia were randomly planted at 0.9 m spacing. Shoots, 1 to 1.5 m in length, pruned from peach trees in January, 2001, were collected and inoculated with *Botryosphaeria dothidea* (5×10^5 conidia/ml) and placed on the trellis wires above the newly planted trees. Misting of the trellis, 5 min per hour, occurred during May, then the pruned shoots were removed. Shot hole *Wilsonomyces carpophilus*, with the inoculum source

being the pruned shoots, was rated in June. Rust *Tranzschelia discolor* inoculum was obtained by removing shoots from infected trees during October, suspending them on the trellis wires, then misting at 5 min per hour for 5 days. Variations in disease susceptibility was detected among the three diseases in the 25 cultivars.

Impact of cell stress on the efficacy of *phlA*-based quantitative competitive PCR in biocontrol *Pseudomonas fluorescens* CHA0. F. Rezzonico (1), Y. MOËNNE-LOCCOZ (2), and G. Défago (1). (1) Phytopathology group, Institute of Plant Sciences, ETH, Zürich, Switzerland; (2) UMR CNRS Ecologie Microbienne, Université Claude Bernard (Lyon 1), 69622 Villeurbanne, France. Phytopathology 92:S69. Publication no. P-2002-0497-AMA.

Monitoring of biocontrol inoculants released into soil is often done by colony counts, but certain bacteria e.g. *Pseudomonas fluorescens* CHA0 can persist in soil as mixed populations of culturable and non-culturable cells. A quantitative competitive (QC) PCR assay was developed based on the gene *phlA*, which is implicated in the synthesis of the biocontrol metabolite 2,4-diacetylphloroglucinol. QC-PCR was tested in vitro. Significant correlations were found between QC-PCR data and CFU when using CHA0 cells in log or stationary phase. However, no correlation was found between QC-PCR data and CFU (or the total number) of CHA0 cells when the latter had been subjected to abiotic stress. The low efficacy of PCR to amplify DNA from stressed cells is a limitation to the use of QC-PCR.

Association of zebra-stem symptoms on processing tomato with the *Pto* gene. M. D. Ricker. Sunseeds Co. Phytopathology 92:S69. Publication no. P-2002-0498-AMA.

A previously unreported disorder of processing tomato has been observed in two breeding programs in the United States since the early 1990s. Symptoms include foliar necrosis, desiccation, and plant death, but the most common and striking symptom is an intricate pattern of stem necrosis. The disorder is consequently called "zebra-stem". Symptoms have appeared in the greenhouse on plug transplants, and on mature plants grown in pots for hybridization. Zebra-stem has been found in the U.S., Canada and Australia. No pathogen or abiotic agent has been associated with this affliction. Incidence of zebra-stem has increased significantly since it was first observed, but so far is limited to experimental genotypes. All of the affected genotypes are thought to contain the *Pto* and *Fen* genes for bacterial speck resistance and fenthion susceptibility, respectively. An additional gene, which is apparently recessive and closely linked to *Pto* and *Fen*, is suggested as the cause of zebra-stem.

Assessment of sugarcane soils suppressive or conducive to ectoparasitic nematode damage. D. Rimé (1,2), S. Nazaret (1), F. Gourbière (1), R. Bally (1), P. Cadet (2), and Y. MOËNNE-LOCCOZ (1). (1) UMR CNRS Ecologie Microbienne, Université Claude Bernard (Lyon 1), 69622 Villeurbanne, France; (2) SASEX and IRD, Mount Edgecombe, 4300 KwaZulu Natal, South Africa. Phytopathology 92:S69. Publication no. P-2002-0499-AMA.

We compared a South-Africa soil suppressive to sugarcane damage caused by ectoparasitic nematodes with a conducive soil located nearby. Suppressiveness was linked to the prevalence of the weak parasite *Helicotylenchus dihystera* amongst ectoparasitic nematodes. The two soils (entisols) were essentially similar, but significant differences were found for organic matter content and pH. Climatic conditions were similar at both sites and differences in *H. dihystera* population were kept when both soils were studied in a same greenhouse. The size of the bacterial community was similar in both soils, but some differences were found by Ribosomal Intergenic Spacer Analysis (RISA) and when counting culturable fluorescent pseudomonads. Thus, apparently-small differences in soil composition seem to lead to significant differences in ectonematode community, ectonematode damage and yield.

Host resistance and cultural practices as measures to control faba bean necrotic yellows virus. L. R. Rizkalla. Plant Pathology Research Institute, Giza, Egypt. Phytopathology 92:S69. Publication no. P-2002-0500-AMA.

Faba bean necrotic yellows virus (FBNYV) is a major economic importance on faba bean in Egypt. A number of cultural practices, such as rouging of infected plants, sowing date and used of insecticides were evaluated as control measures of disease. Early sowing date (November 1) plus rouging and spraying of the aphicide primor reduced virus incidence from 47.05% to 1.76%. Screening of 1213 genotypes for FBNYV resistance during 1999 - 2000 growing season and 204 genotypes during 2000-2001 growing season using artificial inoculation of the virus by the aphid vector. Aphis genotypes as relatively highly tolerant. Twelve plants were selected from ten genotypes for re-evaluation for FBNYV resistance during 2001-2002 growing season.

Survey for faba bean and lentil and chickpea viruses in 2000-2001 growing season in Egypt. L. R. Rizkalla. Plant Pathology Research Institute, Giza, Egypt. Phytopathology 92:S69. Publication no. P-2002-0501-AMA.

The faba bean disease epidemic, which occurred during the growing season of 1992 in El-Minia, Beni Sweef and El Fayoum governorates, culled for intensive efforts to study these phenomena and try to identify the virus and develop appropriate control measures. The survey was conducted in this Governorates in faba bean, Lentil and Chick pea, total of 1760 samples of faba bean were collected from 88 fields of faba bean were surveyed from Middle Egypt (Minia and Beni Sweef), Delta Region (Qalubia, Monofia, Sharkia, Gharbia, Dakahlia, Kafer Elshiegh, Beheara and Nobarria) and Fayum Governorate (Fayume destric, Tamia, Ebshowi and Senoris) and 280 Lentil samples were collected from 28 Lentil field from different governorates (Sharkia, Kafer Elshiegh, Beni Sweef, Giza, Nubarria, and Sinai) and collected 90 samples from Chick pea were collected from 6 fields from Giza, Beni Sweef, and Kafer Elshiegh Governorates. The total samples were tested by ELISA in the virology Lab against the different antiserum and found 24.7% from the total samples of Faba Bean diseased by FBNYV and 41% diseased by BYMV and 3.5% diseased by BBWV and in the Lentil crop found 44 samples from the total diseased by BYMV and 46 samples Diseased by FBNYV, 8 samples diseased by BBSV and one sample diseased by PSbMV, in Chick pea found 29 samples from 90 samples were diseased by BYMV, and 13 samples diseased by FBNYV. Generally, the major viruses problem on Faba Bean, Lentil, and Chickpea in Egypt (FBNY and BYM viruses).

Mutation of a *cyaA* homologue in *Enterobacter cloacae* results in reduced colonization of cucumber but does not affect biocontrol of damping-off. D. P. ROBERTS (1), S. M. Lohrke (1), L. McKenna (1), C. J. Baker (2), W. Li (3), P. D. Dery (1), and J. S. Buyer (1). (1) Sustainable Agricultural Systems Laboratory; (2) Molecular Plant Pathology Laboratory, USDA-ARS, Beltsville, MD 20705; (3) Department of Plant Science, Hubei University, Wuhan, People's Republic of China. Phytopathology 92:S69. Publication no. P-2002-0502-AMA.

Identification of genes involved in seed and root colonization by biocontrol bacteria may allow for strategies for improved ecological fitness, survival, and performance by these strains. *Enterobacter cloacae* strain M59, containing a single transposon insertion, was reduced in colonization of cucumber seeds relative to wild-type strain 501R3 but unaffected in control of *Pythium ultimum* damping-off on cucumber. DNA sequence analysis indicated that the transposon was inserted in *cyaA*, which encodes adenylate cyclase. The growth profile of strain M59 was consistent with a *cyaA* mutant. Adenylate cyclase catalyzes the production of cAMP, which in conjunction with CRP, positively regulates several catabolic and other unrelated functions.

Black leaf spot caused in *Spartina alterniflora* (smooth cordgrass) by *Fusarium moniliforme*. C. L. ROBERTSON (1), R. W. Schneider (1), and S. A. Harrison (2). (1) Dept. Plant Pathology & Crop Physiology, Louisiana State University Agric. Center, Baton Rouge, LA 70803; (2) Dept. Agronomy, Louisiana State University Agric. Center, Baton Rouge, LA 70803. Phytopathology 92:S69. Publication no. P-2002-0503-AMA.

Because of the rapid and extensive land loss that is occurring in coastal Louisiana, a comprehensive project was recently initiated to select and improve ecotypes of *Spartina alterniflora* (smooth cordgrass). This is the predominant species in the Louisiana salt marsh responsible for soil stabilization. Smooth cordgrass plots on a barrier island were evaluated for disease. Leaves throughout the plots had dark brown to black lesions surrounded by chlorotic halos. Isolates from these lesions were tested for pathogenicity on greenhouse-grown smooth cordgrass. Plants were inoculated using spore suspensions and agar plugs. One of the test isolates caused symptoms identical to those found on marsh-grown plants. This isolate was tentatively identified as *Fusarium moniliforme*. This is the first report of *F. moniliforme* as a pathogen on *Spartina alterniflora*.

Plant viruses detected in Alaskan *Streptopus amplexifolius*. N. L. Robertson. USDA, ARS. Phytopathology 92:S69. Publication no. P-2002-0504-AMA.

Streptopus amplexifolius is a member of the *Liliaceae* plant family that is indigenous to south central Alaska, growing in woods and meadows. In late June 2001, plants that bordered a trail in Denali State Park had leaves with uncharacteristic yellow-green dashes on their leaves. A month later, similar appearing plants were found about 50 miles away near Skwentna. Leaf samples were collected from several plants from each site and processed for virus isolation. Protein extracts from the preparations of the Denali and

Skwentna plants consisted of unique proteins about 33kDa and 29kDa, respectively. Long flexuous rods were detected from leaf dips with the electron microscope. Leaf sap and purified preparations were tested against universal potyvirus antiserum (Agdia) resulting in strong reactions from the Denali plants and none from the Skwentna plants. A RT-PCR detection assay using universal primers for potyvirus detection gave the predicted product for only the Denali plants. It was concluded that plants from both sites contained a plant virus that was different from each other, and that the Denali virus was a member of the family *Potyviridae*. This is the first report of plant viruses found in *S. amplexifolius*.

Novel nematocidal activities from *Laetiporus sulphureus* and *Ganoderma lucidum*. L. ROBLES, W. Chun (1), and B. Hiromoto (2). (1) Dept. PSES, University of Idaho, Moscow, ID 83844; (2) Owner, ABR, LL. Phytopathology 92:S70. Publication no. P-2002-0505-AMA.

Cultural and chemical controls for nematode induced diseases are limited in availability and efficiency. In this work, we optimized conditions for the production of nematocidal activity by *L. sulphureus* and *G. lucidum*. Fungi were grown in potato dextrose broth (PDB), rich broth plus 0.2% of canola oil (RB), RB plus 1% canola oil (RBCa), RB plus 1% corn oil (RBCo), or RB without oil (RBNo). At 7-day intervals, culture fluids were collected, centrifuged, filter sterilized, diluted with sterile water, and screened with J2 *M. chitwoodi*. After 36 hours, nematodes that did not recover when transferred to fresh water were considered to be dead. Nematode death was observed in 10^0 to 10^{-2} dilutions of culture filtrates but not in non-inoculated media controls. The highest mortality (100%) was observed in the 10^0 dilutions of RB culture filtrates from 14-35 day-old cultures. Dilution end point data suggests that optimal levels of nematocidal activity occurred from 21-28 days. In PDB, nematocidal activity was significantly less than RB. Thus, nutrition may play a role in the production of nematocidal compounds by these fungi.

Characterization of the biofilm phenotype of *Burkholderia* sp., FP62 and its role in biocontrol. M. M. ROCHE, W. F. Mahaffee, C. M. Press, T. M. Sechler, and A. L. Davis. USDA-ARS, HCRL, Corvallis, OR. Phytopathology 92:S70. Publication no. P-2002-0506-AMA.

Biofilm formation of *Burkholderia* sp. (FP62) on plant leaves and its role in the biocontrol of *Botrytis cinerea* (Bc) was examined on geranium. A library of mini-Tn5 lacZ1 transposon mutants was screened for biofilm formation in a polystyrene microtiter plate assay. Mutants deficient in biofilm formation were identified by the absence of a crystal violet stained biofilm ring normally produced by the wild type. Biofilm mutants were identified and tested for biocontrol efficacy against Bc in a geranium plant assay. Among the biofilm deficient mutants, two (55B1, 62E8) were identified that failed to control Bc in a geranium plant assay despite exhibiting antibiosis to Bc in vitro. Southern analysis indicated a single transposon insertion in each mutant. 55B1 and 62E8 had phenotypic changes in colony morphology and colonization pattern on leaf surfaces (determined by SEM) that are distinct from the wild type and from each other. Biocontrol efficacy of 55E1 was restored using polymers associated with biofilms as spray adjuvants in geranium disease assays. These results suggest that biofilm formation plays a role in biocontrol of Bc by FP62.

Genes for pea pathogenicity and a catabolic gene for a pea root exudate are located on the same "conditionally dispensable" chromosome in *Nectria haematococca*. M. C. RODRIGUEZ and H. D. VanEtten. Dept. Plant Pathology, University of Arizona, Tucson, AZ 85721. Phytopathology 92:S70. Publication no. P-2002-0507-AMA.

Habitat diversity in the fungus *N. haematococca* is partially due to unique habitat-defining genes that are present on CD chromosomes. One of these CD chromosomes contains a cluster of genes (*PEP* cluster for pea pathogenicity), which allows isolates to cause a root rot disease on pea plants. In this study we show that the CD chromosome carrying the *PEP* cluster also carries a gene(s) for utilization of homoserine (HS), an amino acid present in pea root exudates. A screen of isolates from a variety of hosts and geographical locations demonstrated that isolates pathogenic on pea grew on HS, while nonpathogenic isolates did not. Isolates that had lost the CD chromosome also lost the ability to grow on HS. Conventional genetic analyses suggested that HS utilization is encoded by more than one gene, but at least one *HUT* gene is on a CD chromosome. We propose that *HUT* genes provide isolates carrying them a competitive advantage in the pea rhizosphere, prior to the establishment of a pathogenic association with the roots.

Baseline information on the current status of IPM adoption among carrot growers in Wisconsin. P. M. ROGERS and W. R. Stevenson. Dept.

of Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S70. Publication no. P-2002-0508-AMA.

A comprehensive survey of nine commercial carrot growers in Wisconsin, established baseline data on current production practices and adoption of integrated pest management (IPM) methods. Growers were surveyed and interviewed regarding current cropping techniques, implementation of IPM strategies, carrot cultivars grown and pesticide usage. Currently all commercial carrots in Wisconsin are raised for processing contracts on sand or organic muck soils, using 3 to 4 year rotations. Growers utilize F₁ hybrid varieties for increased yield, root quality, sugar content and disease tolerance. The cultivars Carson, Gold King, Danvers and Bolero are the most widely planted varieties. To a limited extent, varieties are blocked and sprayed according to their susceptibility to insects and disease. A season average of 24 scouting trips were conducted by farm personnel, with 30-45 minutes invested in each trip. Growers reported an average of 6 fungicide, 6 insecticide and 7 herbicide applications. Growers are implementing many IPM strategies to reduce cost and minimize environmental impacts.

Sources and prevention of *Erwinia* Early Dying of potato in Kern County, California. M. K. ROMBERG (1), R. M. Davis (1), J. J. Nunez (2), and J. J. Farrar (3). (1) Dept. Plant Pathology, University of California, Davis, CA 95616; (2) UC Cooperative Extension, Kern County, Bakersfield, CA 93307; (3) Plant Science Dept., California State University Fresno, Fresno, CA 93720. Phytopathology 92:S70. Publication no. P-2002-0509-AMA.

Erwinia Early Dying, a disease of potato caused by *Erwinia carotovora carotovora* (Ecc) has been observed since 1992 in Kern County, California, and is becoming a widespread and chronic problem. Since 1998 Ecc has been consistently isolated from plants showing wilt and premature death symptoms. These isolates have been characterized using REP- and ERIC-PCR. Using isolates from 2001 the role of soil versus tuber as primary inoculum has been studied in the greenhouse. The ability of Ecc alone to cause vascular discoloration and wilt symptoms has also been investigated. Isolates taken from plants and soil from various fields have been compared to further elucidate the source of primary inoculum. Field trials involving calcium applications to potatoes grown as seed tubers have resulted in increased vigor of progeny plants and lower incidence of rotting mother tubers.

Association of a fungal endophyte with seed tissue and locoweed toxicity. J. Romero (1), R. CREAMER (1), M. H. Ralphs (2), and D. R. Gardner (2). (1) Dept. of Entomology, Plant Pathology, and Weed Science, New Mexico State University, Las Cruces, NM 88003; (2) USDA, ARS, Poisonous Plants Research Lab, Logan, UT. Phytopathology 92:S70. Publication no. P-2002-0510-AMA.

An endophytic *Alternaria* sp. novo isolated from *Astragalus* and *Oxytropis* locoweed species produces the mammalian toxin swainsonine. The toxin is found in high levels in locoweeds that contain the fungus. We sought to determine the association between fungal presence in seed tissues and locoweed toxicity. The fungus was isolated from seed coats of toxic locoweeds, but not from the embryonic tissues of toxic locoweeds or any seed tissues of non-toxic locoweeds. The endophytic *Alternaria* was detected by PCR with *Alternaria*-specific primers. Tissue culture and growth chamber-grown plants produced with the seed coat removed did not contain PCR-detectable *Alternaria* or detectable levels of swainsonine (<0.001%), whereas similar plants produced with seed coats were found to contain swainsonine (0.21%). These results suggest that non-toxic endophyte-free locoweeds were produced.

Analysis of grapevine xylem sap for evidence of host cell wall degrading enzymes associated with colonization by *Xylella fastidiosa*. M. C. ROPER (1), L. Carl Greve (2), J. Labavitch (2), and B. Kirkpatrick (1). (1) Dept. Plant Pathology, University of California, Davis, CA 95616; (2) Dept. Pomology, University of California, Davis, CA 95616. Phytopathology 92:S70. Publication no. P-2002-0511-AMA.

Xylella fastidiosa, the causal agent of Pierce's disease, poses a major threat to the California grape industry. Infected vines often have occluded vessels in the xylem. The exact origin of these occlusions is unknown as is the mechanism by which *X. fastidiosa* is able to breach the pit "membranes" that separate xylem vessels from one another. It is likely that this bacterium utilizes cell wall degrading enzymes to facilitate systemic movement. GC analysis was used to determine the nature of this cell wall breakdown. The data show an increase in pectin breakdown products in xylem sap from infected vines relative to healthy indicating that pectin digestion may be a means by which the bacteria spread. Direct enzyme activity has thus far been

difficult to detect but immunodetection techniques to test for the presence of cellulases and polygalacturonases are under development. These enzymes are apparently encoded by ORF's in the *X. fastidiosa* genome.

Efficacy of strobilurin fungicides to control potato early blight in Wisconsin. N. ROSENZWEIG and W. R. Stevenson. Dept. Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S71. Publication no. P-2002-0512-AMA.

Potato early blight, caused by *Alternaria solani* plays an important role in potato health and can limit yield. Control of this disease depends primarily on multiple fungicide applications. In WI during 2001, approximately 80% of the total potato acreage was treated with strobilurin fungicides alternating with EBDC or chlorothalonil sprays. Resistance to strobilurin fungicides has now been reported in several plant pathogenic fungi. Foliar early blight progression and relative area under the disease progress curves were determined for spray treatments in 2001 alternating strobilurin (2-6 sprays) and chlorothalonil fungicides (3-7 sprays), for a total of 9 sprays. Isolates of *A. solani* collected from experimental and commercial fields in WI before and after strobilurin exposure in 1998 were compared with isolates collected in 2001 field trials for EC₅₀ of azoxystrobin for the formation of mycelial colonies from germinating conidia (*in-vitro*) and protective activity (*in-vivo*). EC₅₀ values for 2001 isolates are slightly higher than the 1998 isolates.

Screening of biorationals for control of *Phytophthora capsici*. E. N. ROSSKOPF (1), J. P. Albano (1), and E. M. Lamb (2). (1) USDA, ARS, USHRL, Fort Pierce, FL 34945; (2) IRREC, University of Florida, Fort Pierce, FL 34945. Phytopathology 92:S71. Publication no. P-2002-0513-AMA.

Phytophthora blight and root and crown rot of peppers, caused by *Phytophthora capsici*, is considered one of the most limiting diseases in the production of bell peppers in the southeast. Available control measures are often inadequate when weather conditions favoring epidemics of the disease are present. The loss of methyl bromide further intensifies the need to find alternative tactics for control. While there are many biologically based products that have some efficacy for controlling soil borne diseases, few are available that include *Phytophthora* spp. as organisms that they control. Greenhouse trials were conducted testing the efficacy of several biorational products in controlling this disease. All products were applied as soil drenches according to label recommendations for other vegetable crops. A range of application rates was used for experimental materials. Results with organism-based products were highly variable. A mixture of dipotassium phosphonate/dipotassium phosphate, currently in commercial development under the name BIOPHOS, provided consistent control of the disease.

Isolation and characterization of a new closterovirus from grapevine. A. ROWHANI (1), Y. P. Zhang (1), D. A. Golino (1), and J. K. Uyemoto (2). (1) Department of Plant Pathology; (2) USDA-ARS, University of California, Davis, CA 95616. Phytopathology 92:S71. Publication no. P-2002-0514-AMA.

Reports received from different vineyards in California indicated that *Vitis vinifera* cv. Redglobe scions grafted on certain rootstocks succumb a year or two later. In an indexing trial, Redglobe inocula graft-inoculated onto test plants of Cabernet Sauvignon on 4 rootstocks (5BB, 5C, 3309C, and 1103P) declined and died. Close examination of the woody cylinder revealed stem lesion confined to the rootstock portion. A dsRNA was purified from Redglobe canes and used for cDNA cloning and sequencing. The sequence data showed that the isolated virus was a closterovirus with 74% homology to the sequence for *Grapevine leafroll associated virus 2* (GLRaV-2). However, repeated testing in the field of Redglobe vines on the leafroll indicator host, Cabernet Franc, did not produce any symptoms typical of leafroll infection. Results suggest that the new closterovirus was unlike the other closteroviruses associated with grapevine leafroll disease. The name *Grapevine rootstock stem lesion associated virus* was proposed for the Redglobe virus.

Molecular characterization of Indian citrus tristeza virus isolates. A. ROY (1), P. Ramachandran (2), and R. H. Brlansky (1). (1) University of Florida, CREC, Lake Alfred, FL 33850; (2) Advanced Center for Plant Virology, IARI, New Delhi-110012, India. Phytopathology 92:S71. Publication no. P-2002-0515-AMA.

Citrus tristeza virus (CTV) is the causal agent of the most economically important viral disease of citrus worldwide and causes substantial crop losses. CTV infected samples were collected from Bangalore, Delhi, Nagpur and Pune in India. The viral coat protein gene (CPG) was amplified by RT-

PCR yielding an amplicon ~680 bp. for all the isolates. The CPG sequence of the Bangalore stem pitting isolate showed about 97% and 92% homology to cross protecting isolates, PB-61 from Australia and M15A from Japan, respectively. Comparison of the CPG sequences and phylogenetic tree analysis of Indian and exotic isolates was done to determine the position of different Indian isolates in relation to others. Bi-directional PCR, single strand conformational polymorphism and restriction fragment length polymorphism were used to differentiate the four Indian isolates. To further understand the population complexity in these isolates, different segments of the viral genome were amplified using primers specific to well characterized CTV isolates, according to Hilf et al., 1999.

Substrate utilization patterns by *Fusarium solani* f. sp. *glycines* and *F. solani* f. sp. *phaseoli*. J. C. RUPE (1), L. A. Mozzoni (2), and E. B. Gbur, Jr. (1). (1) University of Arkansas, Fayetteville, 72701 (2) Rosario National University, Rosario, Argentina. Phytopathology 92:S71. Publication no. P-2002-0516-AMA.

Utilization of 95 substrates was determined with four isolates each of *Fusarium solani* f. sp. *glycines*, and *F. solani* f. sp. *phaseoli* and one isolate each of *F. oxysporum* and *Colletotrichum acutatum* using FF Microplates (Biolog, Inc). The isolates were grown on malt agar at room temperature for 7 to 14 days. A 10⁴ macroconidia/ml suspension was prepared from each isolate, and 100 ul of the suspension was added to each well. Plates were incubated at 25°C and optical densities at 490 nm were determined at 24, 48, and 72 hours after inoculation using a microplate reader. Each isolate was replicated 3 times and the experiment was conducted twice. Initial analysis using K-means cluster analysis separated the *C. acutatum* and *F. oxysporum* isolates from the other isolates and most of the *F. solani* f. sp. *glycines* from the and *F. solani* f. sp. *phaseoli* isolates. Further analysis will determine which substrates are most useful in identifying f. sp.'s of *F. solani*.

Plant-parasitic nematodes associated with peach production in southern Illinois. J. B. RUSSELL, S. A. Walters, and J. P. Bond. Dept. of Plant, Soil, and General Agriculture, Southern Illinois University, Carbondale, IL 62901-4415. Phytopathology 92:S71. Publication no. P-2002-0517-AMA.

Peaches are an important crop in Illinois with an annual farm-gate value over five million dollars. There are many constraints that hinder peach production in Illinois; however, the influence of plant-parasitic nematodes on peach production is not known. A survey of peach orchards was conducted in 2000 and 2001 to determine the plant-parasitic nematodes that are associated with peach trees in southern Illinois. Eight genera of plant-parasitic nematodes, *Helicotylenchus*, *Meloidogyne*, *Mesocriconema*, *Paratylenchus*, *Pratylenchus*, *Trichodorus*, *Tylenchorhynchus*, and *Xiphinema* were identified. Over the two growing seasons, populations of *Mesocriconema* and *Xiphinema* were found to be at the highest levels. Although *Meloidogyne* was found at low populations in a limited number of samples and have potential to cause excessive damage to peach crops, it appears that *Mesocriconema* and *Xiphinema* have the greatest potential to affect production in southern Illinois.

Two new hosts of *Pseudomonas savastanoi* and variability in strains isolated from different hosts. A. T. SAAD and L. Hanna. Faculty of Agricultural and Food Sciences, American University of Beirut, Beirut, Lebanon. Phytopathology 92:S71. Publication no. P-2002-0518-AMA.

Pathovars of *Pseudomonas savastanoi* are phytopathogens reported to cause hyperplasia of affected tissues resulting in clearly visible knots on especially the branches of several hosts including mainly ash, olive, oleander and privet. The purpose of this study was to survey the host range of *P. savastanoi* in Lebanon and determine the relationships of strains isolated from the different hosts. Disease specimens were collected from symptomatic host species from several localities in Lebanon. All bacterial strains were identified by morphological, biochemical, physiological and pathogenicity tests. Results indicated that, in addition to the previously reported hosts, the pathogen was isolated from two new hosts, namely, myrtle (*Myrtus communis*) and buckthorn (*Rhamnus alaternus*). Biochemical, phenotypic and pathogenicity tests confirmed that the strains isolated from the knots on buckthorn and myrtle are pathovars of *P. savastanoi*. Variations were observed in host range specificity, virulence, biochemical characters and IAA synthesis among strains isolated from the different hosts.

Management of early blight of potato in Khartoum State, Sudan. Z. H. Mohamed Saeed. Dept. Crop Protection, Fac. of Agric. Univ. of Khartoum. Phytopathology 92:S71. Publication no. P-2002-0519-AMA.

Potato *Solanum tuberosum* L production in Sudan started in Khartoum State and spread to other parts of the country. Early blight caused by

Alternaria alternata has caused serious damage to potatoes in Sudan. Thus, field tests were carried out at two localities (Shambat & El Saggai, Khartoum State) in 1989-90 to investigate the effect of three planting dates (Nov. 10, Nov. 25, and Dec. 10, 1989); three alpha seeds (once and twice-grown and farmer's seed); two doses of nitrogen (4.34 - 6.51 Kg/fed.); and five applications (7-day intervals) of Antracol 70 (0.5% a.i.). Early planting in November significantly reduced the development of early blight, that resulted in significant increase in tuber yield. Early blight development was least on the once-grown alpha seed and greatest on the farmer's seeds. Tuber yields also varied significantly among the three seed sources used. Early blight development was significantly lower on potatoes growing in the plots fertilized with the higher nitrogen dose, specially in the less fertile soil of Shambat. Fungicide applications significantly decreased early blight severity and improve tuber yield at Shambat and El Saggai. The integration of these IPM components will definitely slow the progress of early blight epidemics.

Identification of critical factors that influence the screening of cotton cultivars for bacterial blight resistance. U. S. SAGARAM (1), T. A. Wheeler (2), G. L. Schuster (1), and J. R. Gannaway (2). (1) West Texas A&M University, Canyon, TX 79016; (2) Texas Agricultural Experimental Station, Lubbock, TX 79401. Phytopathology 92:S72. Publication no. P-2002-0520-AMA.

Disease resistance is the primary method to control bacterial blight of cotton caused by *Xanthomonas campestris p.v. malvacearum*. The objectives of the studies were to determine how disease incidence was affected by: number of bacterial applications, crop stage, plant density and concentration of bacteria. Cotton plots were inoculated with blight pathogen with the goal of developing a better methodology for disease screening in west Texas. The plots were rated for disease 3-4 weeks after inoculation. Disease incidence was similar for plants inoculated 1, 2, and 3 times. Plants sprayed at 4, 6, and 8-leaf stage exhibited significantly higher disease than plants sprayed at the 2-leaf stage. Plant density (10 to 26 plants/m²) did not have an effect on disease incidence. Disease incidence was linearly related to bacterial concentrations (10⁵ to 10⁷ bacteria / ml) in the most susceptible variety.

Some hydrodynamics characteristics of splash droplet formation and dispersal. S. SAINT-JEAN (1,2), J. K. Hacker (3), and L. V. Madden (1). (1) Dept. of Plant Pathology, Ohio State Univ., Wooster, OH 44691; (2) INRA-EGC, 78850, Thiverval-Grignon, France; (3) LPACT, Ohio State Univ., Wooster, OH 44691. Phytopathology 92:S72. Publication no. P-2002-0521-AMA.

Rain-splash is an efficient physical mechanism of particle dispersal at the local (i.e., small) scale in crop canopies. The process takes place part when particles (e.g., spores) are removed by raindrops and redistributed in splash droplets produced at drop impaction. Spore dispersal can occur when large raindrops or drops falling from higher leaves (i.e., drips) strike spores or a spore suspension and produce spore-carrying splash droplets. We describe some hydrodynamics properties regarding splash droplet production and transport from different biological substrates, including wheat leaves, strawberry leaves, and corn residue, that are impacted by water drops with a range of sizes and velocities. Using data obtained from a phase doppler particle analyzer, we relate the splash process to properties of the impacting drops (e.g., kinetic energy at impact), substrate type, and condition of the substrate (e.g., species of wheat, wet or dry, infected or not).

Evaluation of two biologically derived nematicides for control of lesion nematode, *Pratylenchus sp.*, and effects on predatory nematodes in organically managed soils. K. A. SALINAS and J. B. Kotcon. Div. of Plant and Soil Sci., West Virginia Univ. Phytopathology 92:S72. Publication no. P-2002-0522-AMA.

Two biologically derived nematicides were evaluated for control of a mixed population of lesion nematodes, *P. crenatus* and *P. penetrans*, and their effects on predatory nematode populations. Replicate plots of potato, *Solanum tuberosum* var. Kennebec, were treated per label directions with: 8.5L/ha Nemastroy/Valoram (an extract of chili & essential oil of mustard), 1.8L/ha Nemastop (a plant extract product). Each compound was applied as a soil drench with 0.5 cm irrigation and timed in accordance with the label. Neither product caused a significant change in population density of lesion or predatory nematodes, nor did they affect tuber yield as compared to controls. Lesion nematode populations at planting were low, 3/100cc soil, and increased to 17/100cc soil at harvest. A high percent of lesion nematodes (66%) were colonized by *Pasteuria spp.*, a bacterial parasite, suggesting the presence of naturally suppressive soils. The products did not affect nematode populations in this experiment, but should be tested at higher nematode densities.

Nutritional requirements of *Trichoderma stromaticum*, a mycoparasite of *Crinipellis perniciosa*. S. SANOGO (1), P. K. Hebbar (2), and J. H. Bowers (3). (1) Wye Research & Education Center, University of Maryland; (2) M & M Mars, Inc.; (3) USDA, ARS, Alternate Crops & Systems Lab., Beltsville, MD. Phytopathology 92:S72. Publication no. P-2002-0523-AMA.

Nutritional requirements for developing formulations of *Trichoderma stromaticum*, a biocontrol agent for witches' broom disease on cacao, were evaluated using two carbon and three nitrogen sources mixed to provide four C:N ratios of 5:1, 10:1, 15:1, and 20:1 in liquid and solid growth media. Mineral requirements of *T. stromaticum* were studied using media with complete nutrient status, and media deficient in magnesium, phosphorus, potassium, or sulfur. There were interactive effects of carbon source, carbon rate, and nitrogen source on mycelial growth, conidia production, and chlamyospore formation. Conidia were produced on sucrose-amended media, but few were produced on glucose-amended media. Conversely, chlamyospores were abundantly produced on glucose-amended media compared to sucrose-amended media. Few conidia were formed on media lacking phosphorus. Results from these nutritional studies should provide crucial information for efficient procedures for fermentation and formulation of *T. stromaticum*.

Effect of calcium on peanut pod breakdown caused by *Sclerotium rolfsii*. C. SAUDE and H. A. Melouk. Oklahoma State University. Phytopathology 92:S72. Publication no. P-2002-0524-AMA.

Greenhouse experiments were conducted to evaluate the effect of CaCl₂ at rates of 37 and 74 Kg/ha and CaSO₄ at rates of 368 and 552 Kg/ha on peanut pod breakdown in the cultivar Okrun. Pods were produced in 18-cm dia pots in a soil mix (sand: soil: peat; 2:1:1; v/v/v). Finely ground CaSO₄ and CaCl₂ were applied to soil at 75 days after planting (DAP), and the controls received no additional calcium. At 100 and 120 DAP, pods were singly inoculated with 2 sclerotia of *S. rolfsii*, and with the pegs intact were placed in 7-cm long tube-like pouches made from a 2.5-cm dia dialysis tubing (12,000 MCOW) and returned to soil. Pod breakdown was evaluated periodically and at harvest. Pods inoculated at 100 DAP had similar pod breakdown in all treatments. However, pods inoculated at 120 DAP in the CaCl₂ (37 kg/ha) had 11% pod damage compared with 34% in CaCl₂ (74kg/ha) and 19% in no additional calcium. Both rates of CaSO₄ had 6% pod damage. These results suggest a beneficial role of calcium in reducing peanut pod breakdown caused by *S. rolfsii*.

Use of portable teal-time PCR for same-day on-site field diagnosis of bacterial diseases. N. W. SCHAAD, P. Gaus, and E. Postnikova. USDA ARS Foreign Disease-Weed Science Research Unit, Fort Detrick, MD. Phytopathology 92:S72. Publication no. P-2002-0525-AMA.

Diagnosing bacterial diseases can be very time consuming. Traditional isolation and pathogenicity tests are very sensitive but require 10-20 days or longer. Serological techniques can reduce the time, but the detection threshold is only 10⁴ - 10⁵ cfu/ml. Classical PCR is 10 times more sensitive than serology but requires confirmation tests. Real-time PCR does not require confirmation by a Southern blot, but the equipment is very expensive. We have developed real-time PCR assays using the portable Smart Cycler SC System (Cepheid, Sunnyvale, CA) for on-site detection of several bacteria including *Acidovorax avenae* subsp. *citulli* in watermelon, *Pseudomonas phaseolicola* pv. *phaseolicola* in beans, *Ralstonia solanacearum* in potato, and *Xylella fastidiosa* in grape, citrus, and shade trees. All can be detected in under one hour, including sample preparation. If greater sensitivity is more important than time, samples can be enriched for BIO-PCR in liquid or on solid media.

Geographical distribution of *Typhula* snow molds in Wisconsin using species-specific PCR markers and GIS. E. A. SCHEEF, K. Burke-Scoll, G. Gregos, and G. Jung. Dept. of Plant Pathology, University of Wisconsin, Madison. Phytopathology 92:S72. Publication no. P-2002-0526-AMA.

Snow molds, caused by *Typhula* species, are devastating, psychrophilic fungi that damage turfgrasses and winter cereal crops in the Northern Hemisphere. In Wisconsin, snow mold causes extensive damage to golf courses and sod farms. We collected 3,000 samples of snow mold from 100 golf courses across WI, noting the following variables at each collection site: coordinates, temperature zone, duration of snow cover, species of grass, age of fairway, and fungicides applied to the golf course. DNAs were analyzed by PCR primers specific for *Typhula incarnata*, *T. phacorrhiza*, and *T. ishkariensis* var. *ishkariensis*, var. *idahoensis*, and var. *canadensis*. Data was statistically analyzed to investigate correlations between *Typhula* species/variety distribution and variables, and analyze potential interactions among species and varieties. GIS programs were used to analyze the geographical

distribution of species/varieties across WI. Findings will be used to introduce more effective use of fungicides in the control of *Typhula* snow molds and investigate reasons why more outbreaks occur in Northern regions than in Southern.

Gene expression analysis in a wild type and FadA mutant of *Aspergillus flavus*. K. A. SCHEIDEGGER (1), G. R. O'Brian (1), N. P. Keller (2), and G. A. Payne (1). (1) Dept. Plant Pathology, North Carolina State University, Raleigh, NC 27685; (2) Dept. Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S73. Publication no. P-2002-0527-AMA.

Secondary metabolism in some members of the genus *Aspergillus* results in the production of aflatoxin, a carcinogenic polyketide. Microarrays spotted with expressed sequence tags of genes transcriptionally active during toxin production are being used to study the regulatory mechanisms by which developmental and nutritional cues are linked to aflatoxin production. Asexual reproduction and aflatoxin production are blocked by mutations in *fadA*, encoding an alpha subunit of a heterotrimeric G protein. The specific mechanisms of this regulation have not been fully characterized. Transcriptional analysis under conditions conducive for aflatoxin production identified 165 genes whose transcription was greater in the wild type strain than in the *FadA* mutant, including the aflatoxin genes, *nor-1*, *ver-1*, and *omtA*. Of the 165 genes, 89 have no known function. Selected genes of interest, identified by stringent statistical analyses, are being disrupted to characterize their possible roles in aflatoxin production.

Baseline sensitivity distribution of *Sclerotinia homoeocarpa* (dollar spot) to the novel fungicide nicobifen (BAS 510 F). K. A. Scheidegger (1), F. P. WONG (2), J. S. Barnes (3), H. L. Ypema (3), and H. C. Wetzel III (3). (1) Dept. Plant Pathology, North Carolina State Univ., Raleigh, NC 27695; (2) Dept. Plant Pathology, Univ. of California, Riverside, CA 92521; (3) BASF Corp., Res. Triangle Park, NC 27709. Phytopathology 92:S73. Publication no. P-2002-0528-AMA.

Nicobifen (BAS 510 F) is a new broad-spectrum fungicide currently under development and pending registration in the US for use in controlling a wide number of plant diseases, including dollar spot in turfgrass. Eighty-two isolates of *Sclerotinia homoeocarpa* were tested for sensitivity to the fungicide BAS 510 F using an *in vitro* radial expansion test on fungicide amended water agar. The coefficient of variation for the assay was 37% and 53% for determining ED₅₀ and ED₉₅ values, respectively. ED₅₀ values ranged from 0.018 to 0.94 mg/L (mean = 0.092 mg/L) and ED₉₅ values ranged from 0.20 to 10 mg/L (mean = 1.5 mg/L). No cross-sensitivity to propiconazole or thiophanate-methyl was observed *in vitro*. These results provide useful information for future sensitivity monitoring programs and indicate that BAS 510 F can be used effectively in rotation or tank-mixed with propiconazole or thiophanate-methyl for managing fungicide resistance in *S. homoeocarpa* populations.

Biological control of blueberry flower infection by *Monilinia vaccinii-corymbosi*. H. SCHERM, H. K. Ngugi, and A. T. Savelle. University of Georgia, Athens, GA 30602. Phytopathology 92:S73. Publication no. P-2002-0529-AMA.

The fungus *Monilinia vaccinii-corymbosi* infects blueberry flowers via the stigma-style pathway, followed by ovary colonization and subsequent fruit mummification. The potential for use of the bacterial biocontrol agents *Bacillus subtilis* and *Pseudomonas fluorescens* to suppress infection in this pathosystem was investigated. *B. subtilis* showed strong antibiosis against the pathogen *in vitro*, reducing mycelial growth on agar by >50% after 1 week; growth reduction due to *P. fluorescens* was less pronounced and was statistically significant only after 2 weeks of co-culture. When commercial formulations of the biocontrol agents were applied to stigmatic surfaces of detached flowers, populations of *P. fluorescens* increased over the next 3 days, while those of *B. subtilis* decreased gradually over time. In separate experiments, conidia of *M. vaccinii-corymbosi* were applied 24 h before or after application of the biocontrol agents. *B. subtilis* substantially and significantly reduced the number and growth rates of hyphae penetrating into the stylar canal, while fungal suppression following application of *P. fluorescens* was less pronounced.

Oxalic acid production and pH regulation in liquid cultures of four wood decay fungi. J. S. SCHILLING and J. Jellison. University of Maine. Phytopathology 92:S73. Publication no. P-2002-0530-AMA.

Organic acids such as oxalic acid are important components of basic fungal metabolism and may also play a mechanistic role in the fungal degradation

of wood. Wood decay fungi may produce oxalate to regulate micro-environmental pH. Oxalate may also play a role in increasing iron solubility, an important pre-requisite for an iron-driven Fenton reaction postulated to be involved in brown rot decay. This study focused on oxalate production and pH dynamics in two white rot and two brown rot fungi. Culture pH and soluble and total oxalate production were monitored over time. For all four species, culture pH initially decreased and then stabilized. For the white rot fungi *Trametes versicolor* and *Phanerochaete chrysosporium*, oxalate concentrations were generally very low and not well correlated with pH over time. Brown rot fungi *Postia placenta* and *Fomitopsis pinicola* produced significant amounts of oxalate, and oxalate levels in these species were correlated with culture pH. Additional research characterizing the relationship between oxalic acid production and pH, and the role of oxalate metabolism has been initiated.

Discovery and scale-up of freeze-drying protocols for biomass of Fusarium head blight antagonist *Cryptococcus nodaensis* OH 182.9 (NRRL Y-30216). D. A. SCHISLER and J. E. VanCauwenberge. USDA-ARS, NCAUR. Phytopathology 92:S73. Publication no. P-2002-0531-AMA.

Cryptococcus nodaensis OH 182.9 reduces Fusarium head blight (FHB) of wheat in greenhouse and field environments. Protocols were developed for scaling-up biomass production in 100 liter (L) fermentors, processing biomass using continuous flow centrifugation and storing the biomass as a frozen concentrated paste for subsequent field evaluation at 15 sites across the U.S. in 2001. The current studies were initiated to determine the feasibility of freeze-drying biomass to enhance product stability and maintain biocontrol efficacy. Adding turanose (a disaccharide) and melezitose (a trisaccharide) to liquid culture broth enhanced OH 182.9 survival initially and over time compared to six other cryoprotectants. Melezitose increased the survival of dried OH 182.9 by as much as 2 log units (5×10^8 vs 5×10^6 CFU/ml) after 14 days if it was incorporated at 10 to 100mM at the onset or terminus of biomass production in shake flasks. Cryoprotectants alone occasionally increased FHB. The efficacy of biomass of OH 182.9 produced in a 100 L fermentor and freeze-dried in a 24 L capacity drier will be presented.

Occurrences and severity of diseases of eggplant, pepper, and tomato on Guam. R. L. Schlub. College of Agriculture and Life Sciences, University of Guam, Mangilao, GU 96923. Phytopathology 92:S73. Publication no. P-2002-0532-AMA.

Eggplant, pepper, and tomato are among the ten most popular crops grown on Guam. A list of diseases and their severities was compiled from farm surveys, reviews of Guam Experiment Station reports and publications over the past twenty years. The most difficult and perhaps the only disease that may force a grower out of production is bacterial wilt caused by *Pseudomonas solanacearum*. Southern blight caused by *Sclerotium rolfsii* is more common than bacterial wilt but generally is not as severe. Root knot and mosaic diseases are wide spread and associated with reduced fruit quality and early plant decline. Chemical sprays are routinely used as part of the control program for foliar pathogens such as phomopsis blight of eggplant and bacterial spot of pepper and tomato. Non-infectious diseases include blossom drop, sunscald, off flavor fruits, blossom end-rot, growth cracks, and various nutrient deficiencies. Diseases caused by soilborne and foliar pathogens and non-infections agents increase in severity during the wet season from July-November; tomatoes and bell peppers are not currently recommended for year round production.

Phenotypic and genotypic variation among single-spore isolates from a single basidiome of *Armillaria tabescens*. G. SCHNABEL (1), M. R. Paradkar (1), and G. I. McDonald (2). (1) Dept. Plant Pathology & Physiology, Clemson University, Clemson, SC 29634; (2) USDA Forest Service, RMR5, Forestry Sciences Lab, 1221 South Main, Moscow, ID 83843. Phytopathology 92:S73. Publication no. P-2002-0533-AMA.

Thirty-six single spores were isolated from a basidiome of *A. tabescens* and clamp formation in individual colonies verified haploidy in 32 of 36 isolates. Fluffiness of mycelium, rhizomorph formation, and the ability to stain the growth medium brown varied among isolates when grown on malt extract agar. Sequence analysis of the ribosomal internal transcribed spacer (ITS) and intergenic spacer (IGS) regions revealed two corresponding ITS and IGS alleles in 31 haploid isolates. *AluI* restriction analysis of ITS and IGS regions revealed additional IGS *AluI* restriction patterns in five isolates. These results indicate that sexual recombination in the basidiome was not uniform and produced single spores of different genotypes. Although there was no apparent link between rDNA type and phenotypic characteristics, the genotypic variation may partially explain the phenotypic variability observed *in vitro*.

A rapid ELISA method for detecting *Ralstonia solanacearum*. B. SCHOEDEL and C. Sutula. Agdia, Inc., 30380 County Road 6, Elkhart, IN 46514. Phytopathology 92:S74. Publication no. P-2002-0534-AMA.

Ralstonia solanacearum (Rs) infects a variety of plants worldwide and is a major threat to several crops. To facilitate diagnosis, a rapid ELISA has been developed. This test can detect all races of Rs from infected plant tissues and pure cultures. Samples were prepared by boiling the cells or tissue for 10 minutes in screw cap tubes. After a 10 minute adsorption of the sample to an uncoated ELISA well, the well was washed, blocked, and reacted with peroxidase labeled Rs specific Mab for 10 minutes. The results were determined 5 minutes after substrate addition. The test can detect about 3.5×10^2 CFU/ml in infected geranium tissue under optimum conditions. Specificity was determined by testing with 33 plant pathogen and 10 non-plant pathogen bacterial cultures. This test makes Rs diagnosis possible within 30 minutes.

Development of Rhizoctonia root rot of barley in soils from conventional and no-till fields. K. L. SCHROEDER (1) and T. C. Paulitz (2). (1) Dept. Plant Pathology, Washington State University, Pullman, WA 99164; (2) USDA-ARS, Pullman, WA 99164. Phytopathology 92:S74. Publication no. P-2002-0535-AMA.

Rhizoctonia root rot is a chronic disease of wheat and barley in the Pacific Northwest. *Rhizoctonia solani* AG-8 (Rs) was thought to be the primary causal agent, but recently, *R. oryzae* (Ro) was found to be more predominant in eastern Washington. Ro and Rs at five inoculum densities from 0.8 to 80 propagules/g (ppg) were tested for virulence on barley in soils from two conventional and two no-till fields. Barley plants grown in no-till soils inoculated with Rs or Ro at low inoculum levels had greater plant heights than in conventionally tilled soils. At higher inoculum densities, this trend was reversed for Ro, but there were no differences among soils inoculated with Rs. The number of seminal roots increased with increasing inoculum levels of Rs, with maximum number of roots at 25 ppg. However, with Ro, maximum root numbers were at 2.5 ppg, with reduced numbers at higher inoculum levels. At inoculum levels between 0.8 to 25 ppg, Ro caused greater disease severity than Rs. *R. oryzae* appears to have greater inoculum efficiency than *R. solani*, and both pathogens are influenced by tillage practices.

Effects of seeding rate, row pattern, and fungicide treatment on incidence of peanut stem rot. L. E. SCONYERS (1), T. B. Brennenman (1), and K. L. Stevenson (2). (1) Dept. Plant Pathology, University of Georgia, Tifton, GA 31793; (2) Athens, GA 30602. Phytopathology 92:S74. Publication no. P-2002-0536-AMA.

The effects of seeding rate, row pattern (twin or single), and fungicide (azoxystrobin 0.33 kg/ha, twice) on the development of peanut stem rot, caused by *Sclerotium rolfsii*, were evaluated in 2000 and 2001. In 2000, disease incidence at harvest was significantly higher in single rows (10%) than in twin rows (6%), and azoxystrobin reduced stem rot incidence by 50%. In 2001, incidence of stem rot at harvest was lower in twin rows than in single rows, except at the lowest seeding rate (12.5 seed/m). Higher disease incidence was observed in single rows planted at 22.6 seed/m (11%) than in single rows planted at 17.4 seed/m (6%) or 12.5 seed/m (4%). Disease incidence was higher in non-treated single rows (10%) than in fungicide-treated single rows (4%) or in non-treated twin rows (2%). Disease incidence in twin rows did not exceed 2% and was not affected by seeding rate or fungicide treatment. Our results indicate that growers can reduce stem rot incidence by planting twin rows at lower seeding rates and applying fungicide.

Control of leaf and nut scab in pecans with trifloxystrobin/propiconazole, a new combination fungicide from Bayer Corporation. W. D. SCOTT (1), T. D. Hunt (2), and R. Rudolph (3). (1) Bayer Corporation, Kansas City, MO 64120; (2) Bayer Corporation, Opelika, AL 36801; (3) Bayer Corporation, Peachtree City, GA 30269. Phytopathology 92:S74. Publication no. P-2002-0537-AMA.

Stratego® is a 250 g/L EC formulation containing 125 g/l of trifloxystrobin, a new strobilurin fungicide from Bayer and 125 g/L of propiconazole. Stratego® is currently registered on wheat and peanuts, with pending registrations for corn, rice and pecans. Stratego® applied at a rate of 146-182 g/h gave excellent control of both leaf and nut scab, *Cladosporium effusum* in pecans. In season long disease control programs Stratego® used alternately with other registered products resulted in generally greater control of scab when compared to a similar program omitting Stratego®. Applications of Stratego® have shown no adverse crop effects applied alone or in combination with any tested tank mix partners. Official registration for Stratego® use on pecans is expected by May 2002.

Microscopic evaluation of phyllosphere colonization by a biofilm-forming biocontrol agent. T. M. SECHLER, W. F. Mahaffee, C. M. Press, M. M. Roche, J. A. DiLeone, and A. H. Soeldner. USDA-ARS, Horticulture Crops Research Laboratory, Corvallis, OR 97330. Phytopathology 92:S74. Publication no. P-2002-0538-AMA.

Biofilms of the biocontrol agent, *Burkholderia* sp. strain FP62, in the phyllosphere of geranium leaves were viewed using both fluorescence compound light microscopy (FCLM) and conventional scanning electron microscopy (SEM). Initial work was done by staining with acridine orange and viewed using FCLM. Average number of biofilms was 6, 8.5, and 7 biofilms per cm² leaf area, 6, 12, and 22 days post-inoculation (respectively) on inoculated geranium leaves. Biofilms were strongly associated with trichomes and stomates. They were also observed to be formed on the stalks and bulbs of glandular trichomes. Similar colonization patterns of FP62 were observed on inoculated geranium leaves using SEM techniques. Biofilms 7-day post-inoculation were many cell layers thick and embedded in a smooth polymer-like matrix with bacterial cell-associated fibril strands. On average FP62 biofilms spanned the area of 20 epidermal leaf cells, where some spanned as many as 200 cells. Biofilm formation by FP62 appears to be associated with biological control of *Botrytis cinerea* and FP62 survival on the leaf surface.

Detection of Tomato yellow leaf curl virus and Tomato yellow leaf curl Sardinia virus with a general probe and species specific probes in tomato samples from Agadir, Morocco. M. SEDEGUI (1), M. K. Nakhla (2), T. A. Evans (3), and D. P. Maxwell (2). (1) Ministry of Agriculture, BP 1308, Rabat, Morocco; (2) University of Wisconsin-Madison 53706; (3) University of Delaware, Newark, DE 19717. Phytopathology 92:S74. Publication no. P-2002-0539-AMA.

Tomato yellow leaf curl (TYLC) disease is widespread in the Mediterranean and occurs in Morocco (Plant Dis. 84:490). Tomato leaves with typical symptoms of TYLC disease were collected near Agadir, Morocco in 2001. DNA was extracted from 24 samples and dot blot hybridization was performed at low stringency with a general probe produced by PCR amplification of the most conserved region of the CP gene of TYLCV-[EG1] with primer pair TYCPv369/TYCPc1023. Positive signals were obtained for 23 samples. Specific probes were prepared with primer pair PTYIRv21/PTYIRc287 for TYLCV (Phytopath. Medit. 32:163-173) and MA-14 and MA-15 for TYLCSV (Plant Dis. 84:490) and dot blot hybridization was done at high stringency. All probes were directly labeled with alkaline phosphatase. TYLCV and TYLCSV singly were detected in nine and 11 samples, respectively, and both viruses in four samples.

Management of Phytophthora crown rot of yellow squash with foliar applied fungicides. K. W. SEEBOLD, A. S. Csinos, and T. B. Horten. Dept. of Plant Pathology, University of Georgia, Tifton, GA 31793. Phytopathology 92:S74. Publication no. P-2002-0540-AMA.

Field trials were conducted in Tifton, GA in the spring and fall of 2001 to evaluate the efficacy of several fungicides for the control of crown rot of yellow squash, caused by *Phytophthora capsici*. Materials were applied at 40 gallons per acre beginning at 3 weeks after planting and continuing on a 7-day schedule. Materials tested included mefenoxam, cyazofamid, cyazofamid + pyraclostrobin, cyazofamid + dimethomorph, pyraclostrobin, and azoxystrobin. All treatments reduced disease incidence at season's end (FD) and season-long incidence, measured as area under the disease progress curve (AUDPC). Pyraclostrobin was superior to azoxystrobin with regard to FD and AUDPC. Cyazofamid + pyraclostrobin reduced FD by 88% and AUDPC by 92% as compared to the untreated check. Tank mixes of cyazofamid and pyraclostrobin can be used effectively to manage *Phytophthora* crown rot when applied as high volume foliar sprays.

Partial characterization of a virus serologically related to Johnsongrass mosaic virus. D. L. SEIFERS (1), S. Haber (2), W. Ens (3), Y.-M. She (3,4), K. Standing (3), and R. Salomon (5). (1) Kansas State Univ., ARCH, Hays, KS; (2) Cereal Research Centre, Agriculture & Agri-Food Canada, Winnipeg, Canada; (3) Dept. of Physics, Univ. of Manitoba, Winnipeg, Canada; (4) Hospital for Sick Children, Toronto, Canada; (5) Volcani Center, Bet-D. Phytopathology 92:S74. Publication no. P-2002-0541-AMA.

Comparison of Johnsongrass mosaic virus (JGMV) isolates showed that an isolate from sorghum in Nigeria (Ni-virus) caused different phenotype responses in sorghum. The Ni-virus was positive in enzyme-linked immunosorbent assay to only JGMV antiserum. The capsid was analyzed for relative molecular weight (RMW) by sodium-dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE), and the amino acid (aa) sequence was determined by time-of-flight mass spectrometry (TFMS). In SDS-PAGE, the

capsid had a RMW of 29,428 Da compared to 32,863 Da for JGMV-KS1. The aa sequence had a 10 aa deletion and the remaining sequence differed by 15% from JGMV-KS1. Taken together these data indicate that the Ni-virus is a distinct potyvirus. We propose the name 'dawa mosaic virus'; dawa means sorghum in Hausa, the main language of northern Nigeria.

Study of genetics of resistance and identification of genes involved in resistance in *Phaseolus vulgaris* cv. Othello to Bean dwarf mosaic virus. Y. SEO (1), W. J. Lucas (2) and R. L. Gilbertson (1). (1) Dept. Plant Pathology; (2) Section of Plant Biology, University of California, Davis, CA 95616. Phytopathology 92:S75. Publication no. P-2002-0542-AMA.

Bean dwarf mosaic virus (BDMV) is one of several whitefly-transmitted geminiviruses (Genus *Begomovirus*, Family *Geminiviridae*) that infect common bean. Inoculation experiments revealed that most beans of the Mesoamerican gene pool are resistant to BDMV infection, whereas those of the Andean gene pool are susceptible. The genetics of BDMV resistance in cv. Othello (Mesoamerican) was examined by making crosses with the BDMV-susceptible cv. Topcrop (Andean). F1 progeny were nearly uniformly resistant, whereas in the F2, a 3:1 ratio of resistant: susceptible plants was obtained. This suggests that a single dominant gene be involved in this resistance. By using cDNA subtraction and RT-PCR with degenerate primers, a number of upregulated genes have been cloned from BDMV-infected cv. Othello hypocotyl tissues. These include pathogenesis-related genes and resistance gene analogs (RGAs). Full-length clones of two RGAs (RT4-4 and F110) were obtained by RACE-PCR. The role of these genes in BDMV resistance is being examined.

Interaction of lance nematode populations and cultural practices on creeping bentgrass quality. D. SETTLE (1), J. Fry (2), N. Tisserat (1), and T. Todd (1). (1) Dept. Plant Pathology and (2) Division of Horticulture, Kansas State University, Manhattan, KS 66506. Phytopathology 92:S75. Publication no. P-2002-0543-AMA.

Irrigation and clipping height treatments were applied to four bentgrass cultivars on an experimental putting green naturally infested with lance nematodes (*Hoplotaimus galeatus*). Treatments were arranged in a strip-strip plot design. Population densities ranged from 0 to >1500 lance nematodes 100 g⁻¹ soil in samples collected monthly from each sub-subplot. Nematode populations were negatively correlated with visual quality ($r = -0.20$, $P = 0.06$), reflectance at 813 nm ($r = -0.17$, $P = 0.09$), and color at 1/661 nm ($r = -0.22$, $P = 0.03$) in Jul, and with photosynthesis ($r = -0.28$, $P = 0.005$) in Aug. Nematode densities in Aug averaged 45-50% higher in plots seeded to the cultivar Penncross compared to Crenshaw, L-93, or Providence. Reducing clipping height from 0.40 to 0.32 cm reduced nematode populations and turf quality, but did not significantly alter their relationship. Plots that received supplemental, daily irrigation in mid-afternoon averaged 240% more juveniles ($P = 0.10$) compared to plots that were irrigated every other day.

Molecular methods for detection of *Banana bunchy top virus* from banana tissues and viruliferous banana aphids. A. A. SHALABY (1), A. A. Rezk (1), M. K. Nakhla (2), S. El-Deeb (3), F. Abo El-Abbas (3), M. El-Hammady (3), H. M. Mazyad (1), and D. P. Maxwell (2). (1) Agricultural Research Center, Giza, EG; (2) Univ. of Wisconsin-Madison 53706; (3) Ain Shams University, Cairo, EG. Phytopathology 92:S75. Publication no. P-2002-0544-AMA.

The six DNA components of BBTV from Egypt were amplified with PCR primer pairs specific for each component, and the fragments cloned and sequenced (DNA 1, 2, 3, 4, 5, and 6; AF102780, AF102781, AF102782, AF102783, AF102784, AF148139, respectively). Squash and dot blot hybridization methods were developed for detection of BBTV. The probe (480-bp PCR fragment from DNA 1) was labeled with 32P for radioactive detection, with biotin for a chemiluminescent technique, or with digoxigenin for a colorimetric method. The radioactive method was slightly more sensitive than the chemiluminescent method, whereas the digoxigenin technique was the least sensitive. An immuno-capture PCR (IC-PCR) assay was developed, and it was more sensitive than the hybridization methods. All methods could detect BBTV in extracts of infected banana leaves and in viruliferous banana aphid (*Pentalonia nigronervosa*).

Detection of three potato viruses by multiplex reverse transcription-polymerase chain reaction in a single reaction. A. A. SHALABY (1), M. K. Nakhla (2), A. M. Soliman (1), D. P. Maxwell (2), A. Hadidi (3), and H. M. Mazyad (1). (1) Agricultural Research Center, Giza, Egypt; (2) University of Wisconsin-Madison 53706; (3) NIH, Bethesda, MD 20892. Phytopathology 92:S75. Publication no. P-2002-0545-AMA.

Potato virus Y (PVY), *Potato virus X* (PVX) and *Potato leaf roll virus* (PLRV) are the economically important viruses in commercial potato production in Egypt. M-RT-PCR assays were used for the detection of these three viruses using three specific primer pairs designed to amplify the full-length of the coat protein gene of each virus (810 bp for PVY, 700 bp for PVX, 548 bp for PLRV). Specificity of the three primer pairs was confirmed by sequencing the amplified RT-PCR products. This M-RT-PCR method could detect all three viruses in a single reaction mixture. In naturally infected, field grown potatoes, M-RT-PCR detected the infection of two or three viruses in leaf tissues and tubers. No single virus infections were detected. The M-RT-PCR assay developed is a reliable, rapid and sensitive method for the detection of these three viruses in one plant sample.

Field assessment of partial resistance to *Puccinia triticina* in recombinant inbred lines from wheat cross CI 13227 × Suwon 92. G. SHANER and G. Buechley. Purdue University. Phytopathology 92:S75. Publication no. P-2002-0546-AMA.

Partial resistance in wheat to *Puccinia triticina* may provide more durable protection than monogenically controlled hypersensitive resistance. Long latent period is a major component of CI 13227's resistance, and is conditioned by the action of 4 genes of unequal effect, based on analysis of 98 recombinant inbred lines (RILs) derived from CI 13227 crossed with a susceptible cultivar, Suwon 92. We compared rust severity on RILs in the field to latent period, as measured in the greenhouse. We grew RILs in replicate, single-row, 1-m plots during 3 years and assessed rust severity several times during the growing season. Area under the disease progress curve, standardized to adjust for length of the epidemic each year, was the most informative statistic to compare degrees of partial resistance among RILs. There were large differences in rust intensity among RILs. Lines were reasonably consistent ($r > 0.69$ for comparisons between each pair of years) in expression of partial resistance, and rust intensity in the field was negatively correlated with latent period.

Specific detection and quantification of plum pox potyvirus by real-time fluorescent RT-PCR. D. J. Sherman, W. L. SCHNEIDER, A. L. Stone, V. D. Damsteegt, and R. D. Frederick. USDA ARS Foreign Disease-Weed Science Research Unit, Fort Detrick, MD 21702. Phytopathology 92:S75. Publication no. P-2002-0547-AMA.

Plum pox potyvirus (PPV), economically the most important stone fruit virus in the world, was identified in Adams County, PA, in 1999. Surveys continue in the surrounding areas for detection and eradication of trees infected with PPV. The survey uses a PPV monoclonal antibody ELISA-DASI assay, which is time consuming and somewhat unreliable in detecting low titer viral infections. A real-time fluorescent RT-PCR assay was developed to detect and quantify PPV in tissue samples using multiple platforms (i.e. Smart Cycler, TaqMan, etc.). The target sequence was selected in a conserved region of the coat protein. Detection limits (750 fg of viral RNA) and standard curves were determined using *in vitro* transcripts that included the target sequence. The assay successfully identified European and Pennsylvania strains of PPV in leaf tissue samples from plum, peach, pear, *N. clevelandii* and *N. benthamiana*. Ongoing work includes adapting the assay to index different *Prunus* tissues (i.e. roots, stems, bark, buds and flowers).

Investigation of outbreaks of bacterial canker of tomato in California. Q. J. SHI, R. M. Davis, and R. L. Gilbertson. Dept. Plant Pathology, University of California, Davis, CA 95616. Phytopathology 92:S75. Publication no. P-2002-0548-AMA.

Bacterial canker of tomato caused by *Clavibacter michiganense* subsp. *michiganense* (*Cmm*) reemerged as a serious yield-reducing disease in California in 1998. Isolates from infected tomato plants, crop debris and seed lots were characterized by colony morphology on 523 media, pathogenicity tests, detection with *Cmm*-specific PCR primers and Rep-PCR fingerprinting. All four known rep-PCR types (A, B, C, and D) of *Cmm* were detected. Various seed treatments were evaluated; optimal treatments to eliminate *Cmm* from seed were 1.24% HCl for 30 min and 5% Physan 20 for 5 min. Long-term viability of *Cmm* in association with tomato debris was determined over two seasons. Dried tomato stems infested with *Cmm* in nylon mesh bags were placed on the soil surface or buried (30 cm deep) in soil. Samples were retrieved monthly and assayed for *Cmm*. *Cmm* survived for at least 24 months in infested debris on the soil surface, but for only 6 months in buried debris.

Cercosporin biosynthesis by *Cercospora zea-maydis*: Expression of genes involved in lipid metabolism and secondary metabolism. W. B. SHIM and L. D. Dunkle. USDA-ARS, Dept. Botany and Plant Pathology, Purdue University, W. Lafayette, IN 47907. Phytopathology 92:S76. Publication no. P-2002-0549-AMA.

Cercospora zea-maydis, the fungal pathogen causing gray leaf spot of maize, produces cercosporin, a perylenequinone phytotoxin that is a pathogenicity factor in other *Cercospora* species. Pathway intermediates and genes directly involved in cercosporin biosynthesis have not been determined. We constructed a *C. zea-maydis* cDNA subtraction library and identified a number of genes that are up-regulated during nutritionally controlled synthesis of cercosporin. For further characterization, we selected seven genes suggested to be involved in lipid metabolism and secondary metabolism based on nucleotide sequence analysis (translated BLAST). We analyzed the expression of those genes and cercosporin production in liquid media over a 7-day incubation period. Also, from a *C. zea-maydis* genomic library, we isolated cosmid clones that contain the respective genes. Gene cloning, sequence analysis and targeted gene disruption experiments are in progress to assess the role of cercosporin in pathogenicity of *C. zea-maydis*.

Evaluation of captan/sulfur tank-mixes for scab control on peaches. E. J. SIKORA (1), J. A. Pitts (2), R. Boozer (2), and E. M. Bauske (3). (1) Dept. of Ent. & Plant Path., Auburn Univ., AL 36849; (2) Chilton Area Hort. Sta., Clanton, AL; (3) AWIS Weather Services, Auburn, AL. Phytopathology 92:S76. Publication no. P-2002-0550-AMA.

Peach growers in Alabama often use less costly sulfur instead of captan to control scab (*Cladosporium carpophilum*). Recently, growers have begun tank-mixing lower rates of these products to either improve scab control or reduce production costs. Our objectives were to determine the effectiveness of these tank-mixes and the ratio of sulfur to captan needed to control scab. The five cover spray treatments included: 1) unsprayed control, 2) Captan 50 WP 5 lb/A, 3) Sulfur 80% 9 lb/A, 4) Captan 50 WP 3 lb/A + Sulfur 80% 5.5 lb/A, and 5) Captan 50 WP 2 lb/A + sulfur 80% 3.5 lb/A. All treatments, except the control, received the same bloom and preharvest fungicide applications. At harvest, 40 fruit/plot were rated for scab incidence and fruit marketability. In two of the three years, the Captan 50 WP 5 lb/A and Captan 3 lb/A + sulfur 80% 5.5 lb/A treatments had fewer fruit with scab and higher levels of marketable fruit than the sulfur 80% at 9 lb/A treatment. Scab incidence and fruit marketability among the treatments that used Captan were similar.

Effect of fungicides on rooting of three ornamental species. L. J. SIMMONS and K. L. Bowen. Dept. Entomology and Plant Pathology, Auburn University, AL 36849-5409. Phytopathology 92:S76. Publication no. P-2002-0551-AMA.

Many ornamental species are asexually propagated under conditions of high humidity and temperature. Because these conditions are highly conducive to disease, fungicide applications are needed. However, some fungicides are known to inhibit rooting of particular species. We evaluated nine fungicides for their effects on rooting of three ornamental species (*Spiraea cantoniensis*, *Rhododendron indica* "Pride of Mobile", and *Viburnum prunifolium*). The fungicides, PCNB, etridiazole, myclobutanil, azoxystrobin, captan, benomyl, chlorothalonil, iprodione, and thiophanate-methyl, and a water control were sprayed weekly on cuttings that were under mist irrigation and in soilless media. PCNB and etridiazole were found to inhibit rooting of spiraea and only PCNB inhibited azalea rooting. No fungicide detrimentally affected the viburnum species. These data indicate a need for concern about pesticide use in propagation or a need for additional information concerning disease control in propagation.

Pineapple genetically modified for resistance to plant-parasitic nematodes. B. SIPES (1), C. Nagai (2), M. McPherson (3), H. Atkinson (3), D. Christopher (1), J. Hu (1), R. Paull (1), K. Rohrbach (1), P. Moore (4), C. Oda (5), P. Wood (6), and M. Conway (7). (1) University of Hawaii; (2) Hawaii Agriculture Research Center; (3) University of Leeds; (4) USDA-ARS; (5) Del Monte Fresh Produce, Hawaii; (6) Maui Land and Pineapple Company, Ltd.; (7) Dole Foods Hawaii. Phytopathology 92:S76. Publication no. P-2002-0552-AMA.

Pineapple plants transformed to express a cystatin protease inhibitor were compared to wild type pineapple plants for growth and reproduction of reniform nematode. Twenty transgenic and 20 wild type plants were removed from tissue culture, rinsed free of media, weighed, and transplanted into sterile soil. Fifteen plants of each type were inoculated with 500 eggs of reniform nematode, while the remaining five plants were not. Six months

later, the plants were harvested, root and shoot weight recorded, and nematode eggs extracted from the roots. Uninoculated wild type plants grew an average of 1152% over the 6 months. Uninoculated transformed plants grew 990% which was not different from the wild type plants ($P > 0.1$). Nematode infection reduced plant growth in both the wild type and transformed pineapple plants, 1012% and 809%, respectively. The transformed pineapple had less nematode reproduction than occurred on the wild type plants ($P < 0.05$). An average of 11,593 eggs/plant were recovered from the transformed plants and 15,221 eggs/plant from the wild type pineapple. The range of nematode reproduction per plant was greater on the transformed plants than on the wild type plants suggesting chimerism within the transformed pineapple plants. The transformation did not produce visible off phenotypes but seems to have adversely affect plant growth rate.

Chemical and genetic analyses of pigment biosynthesis in the chestnut blight fungus. T. M. SIRVENT (1), H. McLane (1), C. Tsang (1), S. Krasnoff (2), D. M. Gibson (2), and A. C. L. Churchill (1). (1) Boyce Thompson Institute; (2) USDA/ARS Plant, Soil, and Nutrition Laboratory; Ithaca, NY. Phytopathology 92:S76. Publication no. P-2002-0553-AMA.

Cryphonectria parasitica synthesizes orange and yellow pigments, which are aromatic polyketides that exhibit numerous biological activities *in vitro*. The roles of the pigments in the biology of the fungus are unknown. We isolated 15 mitotically stable mutants that are altered in pigment production by insertional mutagenesis. We developed a HPLC method for the characterization of the pigments from crude extracts. Chemical analyses of pigment mutants showed significant alterations in amounts and ratios of known and unknown pigments. Aloe-emodin was detected for the first time in *C. parasitica* in several of the pigment mutants, and several unknown compounds are being characterized by LC-MS. Using PCR amplification with degenerate primers designed to conserved domains of polyketide synthase (PKS) genes, we cloned 10 unique fragments with similarity to known PKSs. Complementation analyses of pigment mutants with PKS-containing cosmids are in progress to identify a cosmid that restores polyketide pigment production.

Sclerenchyma cell deterioration of CTV-infected Mexican lime. M. SKARIA and H. Miao. Texas A&M University-Kingsville Citrus Center. Phytopathology 92:S76. Publication no. P-2002-0554-AMA.

Mexican lime leaf veins infected with a severe *Citrus tristeza virus* (CTV) strain (CTV-3) and a mild strain (T-TX 8) were compared. It is a universal indicator plant for virtually all strains of CTV. Translucent areas in the vein are prominent in the leaves infected with a severe CTV and less prominent with mild strains. Cross sections of veins from a prominently translucent, slightly translucent, non-translucent, and 'healthy' areas were compared for sclerenchyma cell degradation. These cells form a sheath around the leaf vascular bundles. They are thick-walled and lignified cells that provide mechanical support. Specimens for microscopy were prepared with free hand sections and stained with Fungi-Fluor™ and examined under a Nikon Eclipse TE 300 fluorescence microscope. With severe CTV, the sclerenchyma cell degradation was 63% of the total in sections from the prominent vein clearing area and 34-48% in less prominent areas. With mild CTV, the highest cell degradation was only 40%. Non-translucent and 'healthy' sections had all cells intact. This appears to be the first quantitative report of sclerenchyma cell degradation associated with CTV.

Fluorescence microscopy of leaves from five citrus cultivars affected by greasy spot disease. M. SKARIA and H. Miao. Texas A&M University-Kingsville Citrus Center. Phytopathology 92:S76. Publication no. P-2002-0555-AMA.

Cross sections of grapefruit and sweet orange leaves with greasy spot symptoms (caused by *Mycosphaerella citri*) were compared. The samples were comparable in leaf maturity and infection stages. The analyses were conducted using Fungi-Fluor™ for fluorescence and the specimens were examined under a Nikon Eclipse TE 300 fluorescence microscope. Infections of Amber sweet orange leaves extended deeper into the parenchyma cells. Both Amber sweet and Rio Red grapefruit had numerous infections on both upper and lower epidermis. Rio Red grapefruit showed very pronounced hypertrophy of parenchyma below the palisade tissue. Orlando tangelo, Marrs orange, and Star Ruby grapefruit had more infection sites on the lower epidermis than the upper epidermis. In a related study with 80 grapefruit trees, tape-lift samples were collected from the lower epidermis of young leaves and the number of conidia and ascospores of *M. citri* were assessed. Apart from the ascospores, conidia of *M. citri* were prevalent. New information from fluorescence microscopy will help disease management strategies.

Identification of quantitative resistance in *Lycopersicon pennellii* to *Phytophthora infestans*. C. D. Smart and W. E. Fry. Dept. of Plant Pathology, Cornell University, Ithaca, NY 14853. Phytopathology 92:S77. Publication no. P-2002-0556-AMA.

Resistance in tomato to *Phytophthora infestans*, was investigated in two mapping populations derived from a cross between *Lycopersicon esculentum* × *L. pennellii*. The first was an F2 population that is ideal to work with because a high-density molecular marker map is available. Using this population, a quantitative trait locus for resistance was identified on chromosome 6, which accounted for 25% of the phenotypic variance with an LOD score of 3. This QTL is different from either of the two previously identified resistance genes from *L. pimpinellifolium*, Ph1 and Ph2. The second population was a series of near-isogenic introgression lines (ILs), each containing a segment of an *L. pennellii* chromosome introgressed into an *L. esculentum* background. IL6-2 had the highest level of resistance and contains the region of chromosome 6 that was associated with the QTL in the F2 population. Because IL6-2 is near-isogenic with *L. esculentum* cv. M82, the only difference being the region of chromosome 6 which contains the QTL, these two lines will be invaluable for the comparison of the host response to a tomato-specialized isolate of *P. infestans*.

Genomics of compatibility between potato and *Phytophthora infestans*. C. D. SMART (1), S. Restrepo (1), A. Hart (2), R. Buell (2), and W. E. Fry (1). (1) Dept. Plant Pathology, Cornell University, Ithaca, NY; (2) The Institute for Genomic Research, Rockville, MD. Phytopathology 92:S77. Publication no. P-2002-0557-AMA.

The various interactions between the late blight pathogen *P. infestans* and the potato or tomato host have been studied from a variety of perspectives for many years. It is now possible to study these interactions using microarray technology to understand global gene expression. The recent availability of 60,000 ESTs from various potato cDNA libraries and the generation of microarrays containing 5,000 of these cDNA clones have enabled the study of global gene expression during compatible interactions. We are using microarrays in time course experiments to follow gene expression over the course of a compatible interaction. Initial analyses have identified genes (such as pathogenesis-related genes) that, as expected, are induced throughout the course of the compatible interaction. We have also identified a unique group of proteinase inhibitors that appear to be induced early in the infection process, but are repressed later in the infection process. These data will enable us to identify and study genes that may be involved in the compatible interaction.

Comparison of elicitor genes in closely related *Phytophthora* species. C. D. SMART, K. T. Hodge, C. H. Haney, and W. E. Fry. Dept. Plant Pathology, Cornell University, Ithaca, NY 14853. Phytopathology 92:S77. Publication no. P-2002-0558-AMA.

We have recently identified several clonal lineages of *Phytophthora infestans* that are pathogenic on tomato, petunia and *Nicotiana benthamiana*, as well as potato. Elicitins (10 kd proteins produced by *Phytophthora spp*) have been implicated in pathogenicity, and the presence of the elicitor, INF1, has been described as an avirulence factor to *N. benthamiana* in at least one strain of *P. infestans*. We wanted to determine if the pathogenicity of several lineages to *N. benthamiana* was due to modification of the elicitor gene *Inf1*. Sequence analysis of *Inf1* genes from *P. infestans* isolates representing multiple lineages confirmed that in several lineages, the *Inf1* genes were identical to the first reported sequence, thus a modification of the INF1 elicitor was not responsible for pathogenicity to *N. benthamiana*. Additionally, the sequence of the major elicitor gene from the closely related species, *P. mirabilis*, revealed that the deduced amino acid sequences were identical, while the untranslated regions were variable. Interestingly, the major elicitor from *P. phaseoli*, which is also closely related to *P. infestans*, displayed several amino acid differences within the protein sequence.

Root-lesion nematode populations from dryland field crops in a semiarid environment. R. W. SMILEY (1), K. Merrifield (2), and R. E. Ingham (2). Oregon State University, Dept. of Botany and Plant Pathology, (1) Columbia Basin Agric. Res. Center, Pendleton; (2) Corvallis. Phytopathology 92:S77. Publication no. P-2002-0559-AMA.

Annual no-till spring crops are replacing winter wheat-summer fallow rotation in eastern Oregon and Washington. Wheat sampled in annual crop systems during 1999 had *Pratylenchus neglectus* and/or *P. thornei* (root-lesion nematode, RLN) densities up to 3,970 per kg soil and 4,369 per g fresh-weight root. RLN was surveyed in 109 fields in 10 OR and WA counties during 2000. RLN were detected in 94% of cereal (wheat, barley), brassica (canola, mustard) and legume (lentil, chickpea, lupin) crops

sampled. Densities ranged from 0 to 2,449 per g root and 0 to 35,960 per kg soil. *P. neglectus* was more prevalent than *P. thornei*. Densities were influenced by crop rotation but not by tillage. Wheat rotated with fallow always had fewer than 100 per g root. High densities (above 300 per g root for dry environments) occurred in 44% of fields cropped more than 2 of 4 years. Densities were slightly lower from cereals following cereals than following legumes or brassicas. Few RLN occurred in flax or safflower. Overwintering grass and volunteer cereal seedlings were highly colonized.

Crop damage assessment for *Fusarium* crown rot of winter wheat in the inland Pacific Northwest. R. W. Smiley. Oregon State University. Phytopathology 92:S77. Publication no. P-2002-0560-AMA.

Fusarium pseudograminearum and *F. culmorum* damage winter wheat (*Fusarium* crown rot, dryland foot rot, *Fusarium* root rot) in semiarid eastern Oregon and Washington. All plants in 5-foot row sections were collected from 4 areas in 13 randomly selected winter wheat fields in 4 OR and WA counties. Tillers were separated into subsamples for disease severity classes 0 (none) to 4 (browning to fourth node). Crown rot increased grain protein and reduced kernels per head, kernel weight and test weight. Potential yield was calculated by adding numbers of headed tillers in each subsample and multiplying the sum by mean grain weight for healthy tillers. Crop damage was calculated as the difference between measured and potential yield. Crown rot reduced yield by 0 to 35%, with the average (9%) equating to \$17.11 per acre. Annual economic damage in the eastern OR and WA wheat belt was estimated at 3.6 million bushels, or \$14 million, using average county yields, acreage and market prices. Estimates did not include reduced market grade from lower test weight, or grain protein percent too high for soft-white wheat produced for low-protein markets.

Limited fungicide applications affect berry rot severity and resveratrol content of muscadine grapes (*Vitis rotundifolia* Michx.). B. J. SMITH and J. B. Magee. USDA-ARS, Small Fruit Research Station, Poplarville, MS 39470. Phytopathology 92:S77. Publication no. P-2002-0561-AMA.

Berry rot diseases reduce yield and quality of muscadine grapes, but those losses may be minimized by fungicide applications. The fungicides, myclobutanil, azoxystrobin, and tebuconazole, were applied sequentially to two muscadine cultivars every ten days beginning at early bloom and stopping at pre-harvest intervals (PHIs) of 64, 42, 28, 14, 7, 4, 2, 1, and 0 days. Objectives were to determine spray schedule effects on foliage and berry diseases and the relationship between disease incidence and berry resveratrol content. Resveratrol, a phytoalexin, has shown potential for prevention and treatment of cardiovascular disease and certain types of cancer. Resveratrol content in berry skins was determined by HPLC. Foliar and berry diseases were rated visually at harvest. All fungal diseases were lower on 'Doreen' than on 'Summit' and were reduced by fungicide treatments. There were no differences in the number of asymptomatic berries among the 9 PHIs. Resveratrol content of berry skins from fungicide treated vines was significantly lower than those from untreated vines.

Intercropping with tomato resistant variety 'Juliet' reduces early blight on susceptible variety 'Brandywine'. L. J. SMITH and J. Kotcon. Div. of Plant and Soil Science, West Virginia Univ. Phytopathology 92:S77. Publication no. P-2002-0562-AMA.

Disease development was compared in plots where cv. 'Brandywine' was intercropped with resistant cv. 'Juliet' versus 'Brandywine' monoculture. A spore suspension of *Alternaria solani* was sprayed on corner plants in replicate plots in early July. Disease progress was monitored weekly on 'Brandywine' throughout the season. Early blight defoliation over the season was 23.2% greater on 'Brandywine' in monoculture than when intercropped. Rate of disease increase was linear and 'r' values were greater in monoculture (1.05) than in intercropped (0.91) plots (P = 0.0006). Average lesion expansion on 'Brandywine' in monoculture was 0.326 cm/week compared to 0.289 cm/week when intercropped with 'Juliet' (P = 0.09). Yield from 'Brandywine' plants was 27.3% greater when intercropped with 'Juliet' than when grown in monoculture. The presence of resistant foliage in the plot reduced spread of the pathogen to susceptible foliage. Reduction in lesion expansion on 'Brandywine' when intercropped with the resistant variety 'Juliet' suggests an interaction initiating a defense response in 'Brandywine'.

Control of oomycete diseases of vegetable crops with Gavel™ or Zoxium™ fungicides. R. L. SMITH (1), A. E. Duttie (1), B. D. Olson (1), A. G. McFadden (1), and G. M. Kemmitt (1). (1) Dow AgroSciences, 9330 Zionsville Rd., Indianapolis, IN 46268. Phytopathology 92:S77. Publication no. P-2002-0563-AMA.

Oomycete pathogens pose a serious problem for vegetable growers throughout all the major production areas of the United States. Infections of highly virulent strains of diseases such as tomato late blight (*Phytophthora infestans*) are able to quickly cause significant economic crop damage. Gavel (zoxamide plus mancozeb) and Zoxium (zoxamide) fungicides were found to be highly effective at controlling several important oomycete diseases of vegetable crops in studies established in major growing regions. Several years of replicated experimental field trials have characterized the efficacy of these two formulations on key vegetable pathogens such as tomato late blight (*P. infestans*), white rust of spinach (*Albugo occidentalis*), downy mildews of cucurbit (*Pseudoperonospora cubensis*), lettuce (*Bremia lactucae*), broccoli (*Peronospora parasitica*), and onion (*P. destructor*). Results indicated that Gavel at 1.5 lbai/A and/or Zoxium at 0.25 lbai/A applied on a 7 to 10 day schedule provided control of the fore mentioned pathogens that was equivalent or superior to other commercial standard fungicides.

Changes in symptom severity of soybean associated with single and double infections of Alfalfa mosaic virus and Tobacco streak virus. B. J. SORENSEN, C. R. Grau, and N. C. Kurtzweil. Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S78. Publication no. P-2002-0564-AMA.

Alfalfa mosaic virus (AMV) and Tobacco streak virus (TSV) are among a group of viruses found to frequently co-infect soybean (*Glycine max*). In a plot near Janesville, WI, AMV and TSV were found alone in 40% and 37% (respectively) of samples testing positive for virus, compared to a 26% incidence of co-infection with both viruses. Strains of TSV and AMV were isolated from locations in Walworth Co., WI. Plants inoculated with AMV had an average foliar symptom rating of 1.0 (on a five point scale, 0 being asymptomatic and 5 being dead), TSV inoculated plants averaged 3.2, and plants co-infected with both viruses averaged 1.3 symptom severity. Co-infection with AMV and TSV resulted in lower symptom severity than TSV alone. TSV and AMV were purified, and a calibration curve based on mean absorbance values from different concentrations of pure virus was established using ELISA. Virus titer will be compared in plants infected singly or doubly with TSV and AMV. Local lesion counts on Berna Bean inoculated singly or double will be performed for comparison.

Development and application of a PCR detection method to study the distribution and rate of movement of Beet curly top virus in the beet leafhopper (*Circulifer tenellus*). M. J. SOTO and R. L. Gilbertson. Department of Plant Pathology, University of California, Davis, CA 95616. Phytopathology 92:S78. Publication no. P-2002-0565-AMA.

Beet curly top virus (BCTV; Genus *Curtovirus*, Family *Geminiviridae*) is transmitted by the beet leafhopper to dicotyledonous plants in a circulative manner. To investigate the BCTV-vector interaction, a PCR-based method for the detection of BCTV in the insect was developed. Total DNA was extracted from individual insects and used in the PCR with a BCTV-specific primer pair to direct the amplification of an 1132-bp fragment. Using this method, BCTV DNA was detected in the different compartments involved in circulative transmission: digestive tract, hemolymph and salivary glands. To study the temporal distribution of BCTV in its vector, groups of leafhoppers were given acquisition access periods (AAP) ranging from 1 to 48 hours on BCTV-infected plants. Regardless of AAP, BCTV was detected in the insect gut. However, virus was detected in the hemolymph and salivary glands after AAPs of 4 hours or greater, with the amount of virus detected in the hemolymph and salivary glands positively correlated with AAP. The virus persisted for as long as 30 days in the insect.

Population biology of *Aureobasidium pullulans* on apple leaves. R. N. SPEAR (1) E. V. Nordheim (2), and J. H. Andrews (1). (1) Dept. Plant Pathology; (2) Depts. Forest Ecology and Management, and Statistics; University of Wisconsin, Madison, WI 53706. Phytopathology 92:S78. Publication no. P-2002-0566-AMA.

Spatial colonization of the fungus *Aureobasidium pullulans* (Ap) on apple leaves from field trees was studied over two years. Microscopic counts of Ap on leaf transects by fluorescence in situ hybridization (FISH) yielded population estimates \log_{10} 0.5-2.6 units higher than by leaf washing and plating methods. Transect positional data were imported into ArcView™ GIS software for display and subsequent statistical analysis of both occupancy and density. Microscope fields of midveins, smaller veins and interveinal areas were occupied by Ap, on average, of 70, 57 and 15%, respectively. The density of Ap as cells per unit area was significantly greater on veinal vs interveinal areas. Fungal morphotypes observed were primarily chlamydospores, swollen cells and blastospores; hyphae and pseudohyphae were rarely seen. Microcolonies (>10 Ap cells) increased in both number and size during the year and were greater over the veinal areas

and wounds. Thus, Ap colonizes the leaf in a heterogeneous fashion at sites conducive to growth.

Implications of current orchard renovation practices and market forces on the management of fire blight: The need for a new antibiotic for disease control and resistance management. R. A. Spitko. New England Fruit Consultants. Phytopathology 92:S78. Publication no. P-2002-0567-AMA.

Erwinia amylovora is the most serious bacterial pathogen of pome fruits and is a limiting factor in their production. Despite more than 200 years of research, this disease causes major losses annually and continues to spread world-wide. The risk of major fire blight epiphytotics is increasing due to current orchard renovation practices. Market forces are imposing a shift to the production of highly fire blight-susceptible cultivars and to horticultural practices that favor disease development. Lack of control products is compounding the seriousness of the situation. Antibiotics are the only materials which are consistently effective against the most destructive phases of the disease but their usefulness is declining due to widespread bacterial resistance. The integration of gentamicin, a new antibiotic, into future control programs will be discussed.

Effect of powdery mildew infection of 'Bing' sweet cherries on surface pitting. R. A. SPOTTS and L. A. Cervantes. Oregon State University Mid-Columbia Agricultural Research and Extension Center. Phytopathology 92:S78. Publication no. P-2002-0568-AMA.

Surface pitting of sweet cherry fruits is a disorder related to bruising. Both pitting and powdery mildew infection of sweet cherry fruits reduce quality and cause commercial losses. This research was done to study the effect of powdery mildew infection on pitting. Sweet cherry fruits cv. Bing were harvested on 6 July, 2001 from two orchards. Fruits were rated for powdery mildew severity as none, light, or severe. Fruits were cooled to 4 C, then bruised by dropping a 10.0 g weight with a 2.42 mm diameter head a distance of 6.0 cm onto individual fruit. Fruits were stored at 1°C for 14 days, and pitting rated as none, slight, moderate, or severe. Fruits not infected with powdery mildew had significantly more slight pitting than either light or severely infected fruits. Fruits with severe powdery mildew infection had significantly more moderate and severe pitting than healthy fruits. The severity of pitting of mildew-infected fruits may be related to bruising of softer tissues affecting a large area of the fruit. Conversely, bruising of healthy fruits may only affect a small number of cells at the impact point, resulting in a smaller pit.

Proposed chromosome location of Fusarium head blight resistance genes in additional sets of durum disomic substitution lines. R. W. Stack (1), J. D. MILLER (2), and L. R. Joppa (2). (1) Plant Pathology Dept., North Dakota State Univ., Fargo, ND 58105; (2) USDA-ARS, Northern Crop Sci. Lab., Fargo, ND 58105. Phytopathology 92:S78. Publication no. P-2002-0569-AMA.

We recently reported that substitution lines prepared by replacing individual chromosome pairs of durum wheat with those from *Triticum dicoccoides* (TDIC) showed highly significant differences to Fusarium head blight (FHB). From over 400 TDIC accessions, we identified several with useful levels of FHB resistance. Two of these TDIC lines were used to produce new sets of chromosome substitution lines in durum. The purpose of this study was to determine which chromosomes held the resistance loci in these FHB resistant TDIC accessions. Each substitution line was grown in replicated trials in the greenhouse and inoculated at anthesis with *Fusarium graminearum* by the single spikelet method. FHB response was determined visually 3.5 weeks after inoculation. Five lines in the new substitution sets had significantly less disease than the durum parent check. The three chromosomes (1A, 5B, 7A) substituted in those lines are proposed as sites of FHB resistance genes in the two TDIC accessions.

Effect of calcium, nitrogen, and potassium fertilization on white mold of stored carrots. R. W. STACK (1), L. J. Cihacek (2), C. W. Lee (3), and J. M. Hansen (1). (1) Dept. of Plant Pathology, (2) Dept. of Soil Science, (3) Dept. of Plant Sciences, No. Dak. St. Univ., Fargo, ND 58105. Phytopathology 92:S78. Publication no. P-2002-0570-AMA.

A major consideration for carrot production in northern regions is the need to store the crop over the winter. In 1995, 1996, and 1997, growers in North Dakota and Minnesota suffered severe losses of stored carrots due primarily to white mold (WM) caused by *Sclerotinia sclerotiorum*. The disease was not evident at harvest but appeared after several months in storage. Raising the calcium content of carrot tissue has been suggested as a way to reduce

susceptibility to WM. A Carrot field trial using several rates of nitrogen and potassium fertility and a mid-season side dressing of calcium nitrate was established in 1998. Carrots were harvested from each plot, washed, graded and placed in cold storage. After four months, stored carrots were visually scored for presence of WM. Incidence of WM in the checks was 18 to 37%. Neither nitrogen nor potassium fertility showed a consistent response for WM. There was no effect of calcium side dressing on WM in the stored carrots.

Spectral properties of sugar beets with rhizomania. K. STEDDOM, G. Heidel, D. Jones, and C. M. Rush. Texas Agricultural Experiment Station, Bushland TX. *Phytopathology* 92:S79. Publication no. P-2002-0571-AMA.

We have investigated the feasibility of remote sensing of rhizomania of sugar beets caused by beet necrotic yellow vein virus in the presence of nitrogen depletion. Samples were collected from healthy and diseased areas from 7 fields in 2000 and 6 fields in 2001 from mid season through to harvest. Vegetation coverage was calculated from radiometer readings taken in the field at the time samples were collected. Infected areas had significantly lower amounts of vegetation than healthy areas at mid season, but not at the end of the season. Vegetation indices were calculated from reflectance spectra of individual leaves to estimate relative chlorophyll and anthocyanin contents. Chlorophyll concentrations remained constant from mid season through harvest, and were significantly lower in infected beets than in healthy beets. Anthocyanins increased over time in both healthy and infected beets. Individual leaf spectra showed the greatest differentiation between healthy and infected beets in the green region (550 nm) and in the red edge region (700 nm) and the least in the near infra-red (750 – 1000 nm). Rhizomania can be effectively detected with remote sensing instruments.

The use of soil solarization on managing mosaic viral and phyllosticta foliage diseases of snapbeans. C. STEVENS (1), V. A. Khan (1), R. Rodriguez-Kabana (2), J. E. Brown (3), M. A. Wilson (4), D. J. Collins (5), and A. E. Fyffe (1). (1) Dept. of Agri. Science, Tuskegee University; (2) Dept. of Entomology Plant Pathology, Auburn University; (3) Dept. of Hort., Auburn University; (4) Dept. of Agri. Southeast Missouri State University; (5) Urban Forestry Program, Southern University. *Phytopathology* 92:S79. Publication no. P-2002-0572-AMA.

A combination of solarized soil and a mixture of 13% allyl isothiocyanate in 270L/ha of furfural was evaluated for the control of mosaic viral disease a complex of cucumber mosaic virus (CMV) and southern bean mosaic virus (SBMV) and Phyllosticta leaf spot *Phyllosticta phaseolina* of snapbean on Norfolk sandy loam soil at Tuskegee, AL. Two years following solarization, plots arranged in a split plot on a randomized complete block design were planted with 'Kentucky Wonder' pole beans plants in 2000. Solarized soils amended without or with furfural mixture reduced mosaic symptoms and Phyllosticta leaf spot on snapbeans compared to non-solarized bare soil. There were no significant differences of these foliage diseases among solarized treatments. A uniform pattern of viral mosaic disease observed in field plots indicated a seedborne viral disease. Two seed lots of about 100 bean seeds each were used for the identification of mosaic viruses as determined serologically by the enzyme linked immunosorbent assay (ELISA) test, by the Agdia testing Services (Agdia Incorporated, Elkhart, IN). The results of the ELISA serological test conducted in snapbean seeds tested positive for CMV and SBMV. The furfural mixture amended in non-solarized bare soil showed significant reduction of both diseases compared to the non-amended bare soil treatments.

Resistance to azoxystrobin in the gummy stem blight pathogen in Georgia. K. L. STEVENSON (1), D. B. Langston, Jr. (2), and K. W. Seebold. Dept. of Plant Pathology, University of Georgia, (1) Athens, GA 30602; (2) Tifton, GA 31793. *Phytopathology* 92:S79. Publication no. P-2002-0573-AMA.

Azoxystrobin has been used in Georgia since 1998 for control of gummy stem blight, caused by *Didymella bryoniae*. Unsatisfactory disease control was first observed in Georgia as early as 1999 in fields treated with azoxystrobin. In 2001, isolates of the fungus were obtained from 25 commercial watermelon fields and research sites in Georgia. Sensitivity of each isolate to azoxystrobin was determined using a spore germination assay. Conidia of each isolate were transferred to water agar amended with azoxystrobin and salicylhydroxamic acid (SHAM) or SHAM only. Germination was assessed after 48 h of incubation at 23-25°C and the ED₅₀ value was determined for each isolate. An isolate was considered resistant if the ED₅₀ value was greater than 10 micrograms/ml. Of the 65 isolates tested to date, 54 (83%) were found to be resistant to azoxystrobin. The mean ED₅₀ value of the sensitive isolates was 0.702 micrograms/ml and only three isolates had an ED₅₀ value less than 0.20 micrograms/ml. These preliminary

results provide evidence of widespread resistance to azoxystrobin in *D. bryoniae* populations in Georgia.

Nicobifen (BAS 510 F) - Biological characteristics and redistribution properties. R. STIERL, W. Hanke, G. Lorenz, H. Koehle, and E. Ammermann. BASF AG, Agricultural Center, 67 114 Limburgerhof, Germany. *Phytopathology* 92:S79. Publication no. P-2002-0574-AMA.

Nicobifen, developed under code name BAS 510 F, is the new active ingredient of a series of fungicides from BASF. It shows excellent preventative properties against a broad range of pathogens. Microscopic studies including CLSM and SEM indicate that this is due to its strong inhibition of spore germination, as exemplified by the activity against *Botrytis cinerea* and *Monilinia laxa*. In some fungi, nicobifen also demonstrates an effect against mycelial growth and spore development. Laboratory studies with radio-labeled nicobifen demonstrate translaminal movement and movement with the water stream in an acropetal direction to the leaf tip and margins. The vapor pressure and the resulting vapor phase activity are low. These redistribution properties and the high intrinsic activity characterize nicobifen as a fungicide with classical systemic properties.

Managing the race structure of the tobacco black shank pathogen with variety rotation. M. J. SULLIVAN, T. A. Melton, and H. D. Shew. Dept. Plant Pathology, NC State University, Raleigh, NC 27695. *Phytopathology* 92:S79. Publication no. P-2002-0575-AMA.

Predominance of race 1 of *Phytophthora parasitica* var. *nicotianae* has increased in NC due to the deployment of tobacco varieties that contain single-gene or complete resistance to race 0. Variety rotation studies were conducted using three varieties in three fields to assess how different levels and types of resistance affect the race structure when deployed in fields containing single or mixed races. In a mixed race field, the high level of partial resistance in K 346 was most effective in reducing disease incidence. The proportion of race 1 in the pathogen population also decreased with this variety over the growing season. The use of complete resistance in NC 71 resulted in intermediate levels of disease and race 1 predominated. K 326, with a low level of partial resistance, had the highest levels of disease and race 0 remained dominant. In a field where no race 1 was detected initially, disease incidence was high with the use of either partially resistant variety. Complete resistance was very effective, but race 1 was recovered after only one year. A high level of partial resistance was most effective in a third field where race 1 was predominant.

Nucleotide sequence analysis of pPSR1, a *Pseudomonas syringae* pPT23A-family plasmid. G. W. SUNDIN (1), C. T. Mayfield (1), and M. Ullrich (2). (1) Dept. Plant Pathology and Microbiol., Texas A&M University, College Station, TX 77843; (2) Max Planck Inst. Terr. Microbiol., Marburg, Germany. *Phytopathology* 92:S79. Publication no. P-2002-0576-AMA.

We report a comprehensive nucleotide sequence analysis of pPSR1, a pPT23A-family plasmid (PFP) from *Pseudomonas syringae* pv. *syringae*. PFPs are believed to have arisen from a common ancestor because they share a gene (*repA*) that encodes a protein essential for replication. PFPs contain genes that encode effector proteins, toxin biosynthesis, and other virulence factors that contribute significantly to bacterial-plant interactions in diverse *P. syringae* pathosystems. We study the evolution of PFPs and are determining the contribution of these plasmids to virulence and ecology. The conjugative plasmid pPSR1 contains *repA*, a novel *avr* effector gene, an operon (*dapCE*) presumably involved in epiphytic fitness, a putative bacteriocin-production locus, the *ruIAB* UV-radiation tolerance determinant, the streptomycin-resistance transposon Tn5393, and several *tra* genes and loci involved in plasmid maintenance. We will also discuss results of functional and comparative analyses of pPSR1 with PFPs from distinct *P. syringae* pathovars.

Phylogenetic analysis of the *Puccinia andropogonis* complex. L. J. SZABO and A. P. Roelfs. USDA-ARS, Cereal Disease Laboratory, Saint Paul, MN, 55108. *Phytopathology* 92:S79. Publication no. P-2002-0577-AMA.

The rust *Puccinia andropogonis* is a pathogen of North American grasses including *Andropogon gerardii* (big bluestem) and *Schizachyrium scoparium* (little bluestem). *P. andropogonis* is a macrocyclic heteroecious rust with uredinia and telia on grasses, and pycnia and aecia on wide range of dicots in six families. Given the diversity of the aecial hosts, *P. andropogonis* is likely a species complex. DNA sequence analysis of the nuclear ITS rDNA from aecial and telial collections separated *P. andropogonis* into four groups that corresponded to aecial hosts: 1.) *Penstemon gracilis*, *P. grandiflorus*

(Scrophulariaceae); 2.) *Comandra umbellata* (Santalaceae); 3.) *Ampicarpa comosa*, *Lupinus perennis* (Fabaceae); 4.) *Xanthoxylum americanum* (Rutaceae). Samples from the grass host *A. gerardii* fell into groups 2 and 4, while samples from *S. scoparis* fell into group 1. This data supports the division of *P. andropogonis* into groups based on aecial hosts as describe by Arthur (1962). However, the genetic distances between the groups (39-83 bp changes) suggest that these represent separate species.

Resistance to *Phialophora gregata* is expressed in the stem of resistant soybeans. G. M. TABOR, G. L. Tylka, and C. R. Bronson. Department of Plant Pathology, Iowa State University, Ames, IA. Phytopathology 92:S80. Publication no. P-2002-0578-AMA.

Growth chamber experiments (at 22°C) were conducted to test if resistance to *Phialophora gregata*, causal agent of brown stem rot of soybeans, is expressed in the stems of soybeans. *P. gregata* was introduced into the base of the stems of two-week-old plants and advance of the fungus in the stems was assessed. Fungus was isolated from all heights (base to apex) of the stems of 7 susceptible and 11 resistant cultivars one week after inoculation. However, in succeeding weeks, the pathogen advanced with the growing tip more readily in susceptible than in resistant cultivars. The mean percent stem length colonized five weeks after inoculation for susceptible cultivars (87 ± 7%) was significantly greater than the mean for resistant cultivars (57 ± 6%). Mean percentage of symptomatic trifoliate leaflets was 57 ± 17% and 10 ± 2% for susceptible and resistant cultivars, respectively. These results indicate that resistance can be expressed in the stems of resistant soybeans and thus suggest that stem inoculation methods may be useful in resistance screens.

Suppression of Rhizoctonia disease of potato by biological control and a ryegrass rotation. M. M. TALBOT and R. P. Larkin. USDA-ARS New England Plant, Soil, and Water Laboratory, University of Maine, Orono, ME 04469. Phytopathology 92:S80. Publication no. P-2002-0579-AMA.

Rhizoctonia solani is an important fungal pathogen of potato capable of reducing tuber yield and quality. Integrated, sustainable control options, including effective crop rotations and biocontrol, are needed to reduce pathogen losses. The use of ryegrass as a rotation crop was compared with barley, clover, and no rotation in greenhouse tests for effects on inoculum levels and disease suppression. Early results indicate that ryegrass treatments are more effective than clover at reducing disease. Biocontrol organisms, including *Trichoderma* spp., *Paenibacillus polymyxa*, *Penicillium* spp., *Waitea* spp., *Pseudomonas fluorescens*, *Burkholderia cepacia*, *Bacillus subtilis*, *Verticillium biguttatum*, *Laetisaria arvalis* and *Cladophium foecundissimum*, were also screened for disease reduction in greenhouse experiments. Efficacy of the rotation crops alone and in combination with selected biocontrol organisms is being evaluated for reduction of *Rhizoctonia* inoculum and disease of potato in ongoing field trials at two locations in Maine. Efficacy and interactions among treatments will be discussed.

Detoxification of the toxin cercosporin by the bacterium *Xanthomonas campestris*. T. V. TAYLOR, T. K. Mitchell, and M. E. Daub. Dept. Plant Pathology, North Carolina State University, Raleigh, NC 27695. Phytopathology 92:S80. Publication no. P-2002-0580-AMA.

The nonspecific toxin cercosporin is hypothesized to play a role in the pathogenesis of *Cercospora* spp. *Xanthomonas campestris* pv. *zinniae* (XCZ) rapidly degrades this toxin. Growth of XCZ strains in cercosporin-containing medium leads to the breakdown of cercosporin and to the formation of a non-toxic breakdown product called xanosporolactone. Xanosporolactone is hypothesized to be formed via a cytochrome P450-mediated oxygen insertion into one of the quinone rings of cercosporin. Three non-degrading mutants of one XCZ strain (XCZ-3) were created through EMS mutagenesis. By means of a helper cell line containing the plasmid pRK2013, the XCZ-3 mutants were conjugated with an ordered wild type library created in the vector pLAFR6. A complementing clone was found and used to complement the three mutants of XCZ-3. An 8 kb fragment of the subclone that complements the mutants is being sequenced. Isolation of the gene(s) encoding the breakdown of cercosporin may allow for engineering of *Cercospora*-resistant plants.

Field efficacy of root-knot nematode resistance in ‘Charleston Belle’ and ‘Carolina Wonder’ bell peppers. J. A. THIES (1), R. L. Fery (1), J. D. Mueller (2), G. Miller (2), and J. Varn (2). (1) USDA, ARS, Charleston, SC 29414; (2) Clemson University, EREC, Blackville, SC 29817. Phytopathology 92:S80. Publication no. P-2002-0581-AMA.

Reactions of two pairs of near-isogenic bell pepper (*Capsicum annuum* L.) cultivars that differ for resistance to root-knot nematodes (RKN) conferred by the *N* gene were evaluated in field tests at Blackville, S.C. (black plastic-

mulched beds), and Charleston, S.C. (bare-soil beds). The isogenic pepper sets were Charleston Belle (CB - NN) and Keystone Resistant Giant (KRG - nn), and Carolina Wonder (CW - NN) and Yolo Wonder (YW - nn). CB and CW were highly resistant; root gall indices (GI) = 1.1 for both cultivars. KRG and YW were highly susceptible; GI = 4.7 and 4.6, respectively. CB had 96.3% fewer eggs per g fresh root than KRG and CW had 96.9% fewer than YW. CB had greater marketable fruit yield than the other cultivars at Blackville, but there were no differences at Charleston. RKN-resistance conferred by the *N* gene is effective in bell pepper grown on black plastic mulch and on bare-soil beds. RKN-resistant bell peppers should provide economical and environmentally compatible alternatives to methyl bromide and other nematicides for managing RKN.

Development of a rapid take-all assay for bentgrass. S. L. THOMAS, M. J. Boehm, J. W. Rimelspach, and L. H. Rhodes. The Ohio State University, Columbus, OH 43210. Phytopathology 92:S80. Publication no. P-2002-0582-AMA.

Take-all of bentgrass (causal agent is *Gaeumannomyces graminis* var. *avenae*; Gga) can be a serious disease on new golf course putting greens and fairways. Gga is believed to be the most active during cool, moist conditions of spring and fall while symptoms appear during periods of heat and water stress. Our objective was to develop a growth chamber assay to assess the effects of microbial inoculants, organic amendments, and chemical controls on take-all incidence and severity. Conetainers were set up using a sand root-zone mix amended with various amounts of millet seed colonized by Gga as inoculum. The cones were seeded with creeping bentgrass (*Agrostis palustris*, cv. Penneagle) and placed in a growth chamber on a 12 hr day with 18°C day and 12°C night. Symptoms (chlorotic leaf blades and tip dieback) were observed at 16-20 days after set-up. The mean percent infected roots were positively correlated to the percent symptomatic turf observed. The effect of microbial inoculants on disease incidence and severity will be assessed.

IR-4 fungicide registration update. D. C. THOMPSON, V. R. Starner, J. S. Corley, M. Arsenovic, and H. Chen. IR-4 Project, Rutgers University, North Brunswick, NJ 08902. Phytopathology 92:S80. Publication no. P-2002-0583-AMA.

The IR-4 Project had a record year in 2001 with 512 new food-use clearances, 289 new ornamental-use labels and 20 new biopesticide clearances. IR-4 data supported 62 Section 18 uses. More than 70% of the food-use clearances are chemicals and uses identified as “reduced risk” by EPA. IR-4 is committed to working with chemistries needed for each pest; however, we strongly encourage evaluation of safer chemistries such as acibenzolar, azoxystrobin, BAS 510, dimethomorph, fenamidone, fenhexamid, pyraclorobin, quinoxifen, trifloxystrobin, Switch (fludioxonil + cyprodinil), and zoxamide. Triazole fungicides are under review due to an additional metabolite of concern. Greenhouse needs were evaluated and projects initiated for *Botrytis* and *Pythium* control. IR-4’s methyl-bromide alternatives program has identified promising individual products and combinations of products for use in strawberries and tomatoes. IR-4 continues to expand the efficacy program to support adding uses to labels where residue data exists, but the registrant requires additional performance data before labeling the new use.

Mn oxidation in plant pathogenic fungi. I. A. THOMPSON (1), L. Li (1), D. M. Huber (1), and D. G. Schulze (2). (1) Depts. Botany and Plant Pathology; (2) Agronomy, Purdue University, W. Lafayette, IN. Phytopathology 92:S80. Publication no. P-2002-0584-AMA.

Manganese oxidation by the soil-borne fungus *Gaeumannomyces graminis* var. *tritici* (Ggt) has been implicated in pathogen virulence in lab and field experiments. The manganese oxidation factor (MOF) produced by Ggt is an extracellular protein with an estimated size of 50 kD. Spectrophotometric analysis of agar extracts containing the MOF have shown that Mn oxidation activity is absent in Ggt isolates that do not visibly oxidize Mn in culture. Manganese oxidation is inhibited by phenanthroline and other metal sequestering chemicals. This parallels research in Mn oxidizing bacteria where all MOFs to date have been shown to have copper binding motifs. *Pyricularia grisea*, the rice blast pathogen, also oxidizes Mn in culture. A screen of 1,000 REMI generated *P. grisea* mutants has identified several Mn oxidation minus isolates (*mn-*). Pathogenicity tests on barley, rice and wheat roots are being conducted. The isolation of genes associated with Mn oxidation from *mn-* REMI mutants using plasmid rescue is in progress.

Sequencing status of the *Magnaporthe grisea* chromosome 7. M. R. THON, T. Mitchell, D. Brown, and R. A. Dean. Dept. Plant Pathology, North Carolina State University, Raleigh, NC 27695-7251. Phytopathology 92:S80. Publication no. P-2002-0585-AMA.

Magnaporthe grisea, a filamentous fungus and the causal agent of rice blast disease, is a serious pathological threat to food supplies worldwide. This fungus has been the focus of intense studies in recent history that have increased our understanding the molecular determinants of pathogenesis and biology for this and related fungi. These activities provide a starting point and the necessary tools to more thoroughly elucidate the mechanisms involved in host pathogen interactions. Previously, a physical map of the *M. grisea* genome was constructed using a 25X BAC library and a minimum tiling path of 42 BAC clones spanning 95% of chromosome 7 was determined. Using this BAC library, we have initiated large-scale sequencing of chromosome 7 following a BAC-by-BAC approach. Sequencing is being performed by first sequencing 10 seed BACs that are spaced along the chromosome and then selecting contiguous, overlapping BACs to join the seed BACs. Shotgun libraries are prepared for each BAC clone and sequenced to a 5X depth of coverage. We will present our current results from BAC sequencing including analyses of repetitive elements, gene content and order, EST location, and chromosome-wide synteny with related fungi.

Phylogenetic analyses of *Magnaporthe grisea* based on internal transcribed spacer and translation elongation factor sequences. Y. TIAN (1), S. B. Goodwin (2), and M. Levy (3). (1) Botany and Plant Pathology Department; (2) USDA-ARS; (3) Biological Science Department, Purdue University, W. Lafayette, IN 47907. Phytopathology 92:S81. Publication no. P-2002-0586-AMA.

Rice blast is caused by *Magnaporthe grisea* and it is the most important disease of rice in the world. Due to its economic significance, *M. grisea* has become the model pathogen for molecular plant pathology research. Its pathogenicity has been studied extensively. However, the phylogenetic relationships of *M. grisea* are less studied. Previous studies on *M. grisea* populations have focused on its host diversity, DNA fingerprint and RFLP variation. We performed phylogenetic analyses based on sequences of its ribosomal DNA region (ITS1, ITS2 and 5.8S gene) and intron sequences of TEF alpha. Twenty-four isolates of *M. grisea* from different geographical places and hosts were analyzed for ITS and TEF sequence. Similar sequences were downloaded from GenBank and incorporated into the phylogenetic tree. We have found several distinct subgroups among these 24 isolates. We also suggest that *Magnaporthe poae* may be more related to *Gaeumannomyces* than to *Magnaporthe* based on its ITS sequence downloaded from GenBank.

Possible repeat induced point mutation (RIP) in coding and flanking regions of a transposable element from the wheat pathogen *Mycosphaerella graminicola*. Y. TIAN (1) and S. B. Goodwin (2). (1) Purdue University; (2) USDA-ARS, W. Lafayette, IN 47907. Phytopathology 92:S81. Publication no. P-2002-0587-AMA.

We previously identified a putative transposable element in a DNA fingerprint probe from the wheat leaf blotch pathogen *Mycosphaerella graminicola*. This transposable element is very stable. However, it moved occasionally in one isolate during asexual reproduction. The active copy and four inactive copies of the reverse transcriptase gene were cloned and sequenced. A high frequency of RIP mutations (G:C to A:T transitions) were found by DNA sequence comparison of these five clones. These mutations occurred not only in the reverse transcriptase gene coding region but also in the flanking region. All copies except the active one contain one or more stop codons within the coding region. Furthermore, most of the stop codon mutations were G:C to A:T, most likely due to RIP. By comparison, a single-copy gene from *M. graminicola* showed no evidence of RIP. This may provide the first evidence for RIP in a Loculoascomycete and indicates that RIP may be important for inactivating transposable elements for fungi in the genus *Mycosphaerella*.

Characterization of *Phytophthora capsici* in south Florida. A. M. TOHAMY, R. R. Urs, and P. D. Roberts. University of Florida, Dept. Plant Pathology Southwest Florida Research and Education Center, Immokalee, FL 34142. Phytopathology 92:S81. Publication no. P-2002-0588-AMA.

Thirty-three isolates, (8 A1 and 25 A2 mating types), of *Phytophthora capsici* obtained from infected pepper, squash, tomato and watermelon in South Florida in 1997, 98, and 2001 were evaluated for their ability to cause disease on each of their respective and the other three hosts. Some isolates were highly virulent on their primary hosts and less virulent on the other ones. Other isolates were proved to be highly cross-pathogenic. Age-related resistance to *P. capsici* was demonstrated in pepper plants. Plants inoculated at 12-week old showed less necrotic dark brown lesions length on stem when compared to plants inoculated at 6, 8, and 10 weeks old. *P. capsici* isolates were recovered from water detention pond using lemon leaves bait technique. The recovered *P. capsici* isolates were identified according to

their microscopic morphological features, mating with authentic A1 and A2 mating types on V-8 agar and their pathogenic potential on pepper seedlings.

Infectivity of *Phytophthora ramorum* on selected Ericaceae host species. P. W. TOOLEY (1) and L. Englander (2). (1) USDA ARS FDWSRU, Fort Detrick, MD 21702; (2) Dept. of Plant Sciences, University of Rhode Island, Kingston, RI 02881. Phytopathology 92:S81. Publication no. P-2002-0589-AMA.

Phytophthora ramorum, suspected causal agent of sudden oak death in California, was evaluated for its ability to infect ornamental plant species in the family Ericaceae. *P. ramorum* was reported by European workers to attack plants in the genera *Rhododendron* and *Viburnum*, and has been isolated from rhododendrons and other varied plant species in California. Leaves on whole plants were inoculated by cutting off the tips (2-3 mm) and dipping them in *P. ramorum* mycelial suspension, followed by incubation for 7 days in a dew chamber at 20°C in darkness. Mean lesion areas, as percentage of leaf areas were 10.2% for *Arctostaphylos uva-ursi*, 5.9% for *Rhododendron maximum*, 15.9% for Girard's Rose azalea, 4.4% for florist azalea Inga, 31.8% for *Kalmia latifolia* Madeline, 27.8% for *Pieris floribunda*, 32.6% for *Zenobia pulverulenta*, and 17.4% for Cunningham's White rhododendron. Isolate 0-217 from rhododendron in California appeared less virulent than *P. ramorum* type culture 9/95. Additional research is needed to determine the role these potential hosts may play in the epidemiology of sudden oak death.

REMI mutagenesis in the wheat scab fungus *Fusarium graminearum*. M. TRACY (1), Z. Hou (1), H. C. Kistler (2), and J. R. Xu (1). (1) Dept. of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907; (2) USDA-ARS, Dept. of Plant Pathology, University of Minnesota, St. Paul, MN 55108. Phytopathology 92:S81. Publication no. P-2002-0590-AMA.

Fusarium graminearum is an important pathogen of small grains and maize in many areas of the world. Infected grains are often contaminated with mycotoxins harmful to humans and animals. In the past decade, wheat scab (head blight), primarily caused by *F. graminearum* in North America, has emerged as a major threat in wheat production. To better understand the molecular mechanism of plant infection and virulence of *F. graminearum*, we used the REMI (restriction-enzyme mediated integration) approach to generate random targeted mutants. Over 3500 hygromycin-resistant transformants have been generated by transforming pCB1003 into *F. graminearum* PH-1. Nine of 1,500 transformants in a corn-silk infection assay had reduced virulence. Preliminary data indicated that some of them were dramatically reduced in their ability to infect and colonize flowering wheat heads. Genetic analysis and plasmid rescue are under way to identify and characterize genes disrupted in these mutants.

Characterization of oxygenases involved in the *Aspergillus* seed interaction. D. I. TSITSIGIANNIS, T. M. Kowieski, and N. P. Keller. Dept. Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S81. Publication no. P-2002-0591-AMA.

Sexual, asexual reproduction and aflatoxin (AF)/sterigmatocystin (ST) biosynthesis in *Aspergillus* species is affected by both seed and fungal derivatives of linoleic acid (LA). Seed defense enzymes, lipoxygenases (*lox*) convert LA to 13S-hydroperoxylinoleic acid, 13S-HPODE (inhibits AF/ST production) and/or 9S-HPODE (promotes AF/ST gene transcription). Both compounds stimulate *Aspergillus* asexual development, possibly by mimicking effects of psi factors (precocious sexual inducers); endogenous *Aspergillus* sporogenic factors derived from LA. Two putative novel peanut seed *lox* genes have been subcloned from *Aspergillus* infected and non-infected peanut seeds. Characterization of these peanut *lox* genes will reveal the LA derived products (9S-HPODE, 13S-HPODE or both) and could represent the first molecular markers with potential to identify genotypes that enhance or suppress AF contamination. In *A. nidulans* three putative *ppo* (psi producing oxygenases) genes have been cloned. Deletion of *ppoA* increases the sexual to asexual spore ratio in *A. nidulans* and has no prominent effects on ST production.

Spatial patterns of grapevines with Pierce's disease in the lower San Joaquin valley. K. M. TUBAJIKA (1), E. L. Civerolo (1), D. Bartels (2), and J. M. Hashim (3). (1) USDA ARS, Davis, CA; (2) USDA APHIS, Mission, TX; (3) UCCE, Bakersfield, CA. Phytopathology 92:S81. Publication no. P-2002-0592-AMA.

Incidence of Pierce's disease (PD, caused by *Xylella fastidiosa*), continues to increase in many grape varieties in California due to the establishment and spread of the vector, the glassy-winged sharpshooter. Eleven vineyards were

surveyed during 2001 season and each vine was assessed visually for PD and Geo-referenced using GPS/GIS technology. The spatial patterns of infected vines were analyzed by ordinary runs and two-dimensional distance class analyses, and illustrated using multispectral images. Disease incidence ranged from < 1% to 80%. No disease gradient or edge effect was detected in any vineyard. The spatial disease gradient analyses consistently described the non-randomness of the patterns of diseased vines, and an increase in the degree of clustering of diseased vines as disease incidence increased. Based on these results, effective PD management is likely to be based on practices that reduce initial inoculum and use of resistant varieties.

Endophytic activity of *Aspergillus niger* in onion. J. W. TUFFLEY and J. W. Lorbeer. Dept. of Plant Pathology, Cornell University. Phytopathology 92:S82. Publication no. P-2002-0593-AMA.

Endophytic activity of *A. niger* was investigated in onion seedlings, mature plants, and bulbs. Onions utilized in the experiment were propagated in one of two ways: untreated seed sown in soil artificially infested with *A. niger* (T-1), and untreated seed sown in sterilized soil (T-2). At different life stages, these onions were subjected to one of two assay techniques and evaluated for the presence of *A. niger*: seedlings and mature plants were dissected, plated on APDA, and incubated; plants were harvested, dried, topped, and the bulbs then incubated in humidity chambers at either 30°C or 33.3°C. High levels of *A. niger* were detected in T-1 seedling and plant tissues. Upon gaining ingress into the seedlings, *A. niger* appeared to move systemically through the plants. The fungus was consistently recovered from root, basal plate, lower leaf, and leaf tip sections of T-1 plants. *A. niger* was not recovered from T-2 seedlings. After the second assay, T-1 bulbs exhibited the greatest degree of infection by *A. niger*. The combination of high humidity and temperature appeared to trigger the transformation from latent to active black mold infection in the bulbs.

Phytoplasma typing based on PCR of 16S rDNA and genetic sequences. E. TUMBAN, J. Rascoe, and M. Shaw. Department of Life Sciences, New Mexico Highlands University, Las Vegas, NM. Phytopathology 92:S82. Publication no. P-2002-0594-AMA.

Phytoplasmas have been difficult to classify and characterize because they cannot be grown in axenic culture. Formerly, classification was based entirely on biological characteristics, while current classifications are based almost entirely on 16S ribosomal DNA sequences, exemplified by RFLP analysis or sequencing. A few other genes from specific phytoplasmas have been identified and sequenced. To investigate whether similarities in other genes will confirm phylogenetic relationships based on 16S rDNA sequence information, we have used PCR, with either published primers or primers constructed from sequences available in GenBank, to attempt to amplify products from periwinkle plants infected with one of several aster yellows isolates, beet leafhopper virescence agent isolates, or Spiroplasma citri isolates. Primer pairs R16F/ R16R (specific for phytoplasmas) and RPF1/ RPR1 (specific for aster yellows group phytoplasmas) have been used to classify phytoplasmas and were used as positive controls and to confirm group placement. Amplification with the tested primer pairs was generally consistent with group placements made from 16S data.

Effect of desert corn flea beetle on sudangrass bacterial leaf blotch severity. T. A. TURINI (1), H. R. Azad (2), D. A. Cooksey (2), M. D. Rethwisch (3), and M. R. Ambrozio (1). (1) University of California Coop. Ext., Holtville, CA 92250; (2) Dept. Plant Pathology, UC, Riverside, CA 92521; (3) UC Coop. Ext., Moreno Valley, CA 92557. Phytopathology 92:S82. Publication no. P-2002-0595-AMA.

Pantoea ananas (Serrano) Mergaert and *P. stewartii* (Smith) Dye causes leaf blotch symptoms on sudangrass *Sorghum sudanense* (Piper) Stapf. This disease discolors hay and greatly reduces marketability. The effect of reducing desert corn flea beetle *Chaetocnema ectypa* Horn populations on disease severity was evaluated in a replicated field experiment in Imperial County California in 2001. Lambda-cyhalothrin (0.28 L/ha), a pyrethroid insecticide, was applied to sudangrass at weekly intervals from 25 July to 18 August. The insecticide treatment had a seasonal average of 77% lower *C. ectypa* population densities than the untreated control. Disease severity in the insecticide treated sudangrass was 65% lower than the untreated control on 24 August. In addition, *P. ananas* and *P. stewartii* were isolated from *C. ectypa* that were feeding on infected sudangrass. These results suggest that *C. ectypa* may have a role in spread and development of bacterial leaf blotch of sudangrass.

A *Crinivirus* is associated with the Strawberry Pallidosis disease. I. E. TZANETAKIS (1,2), A. B. Halgren (2), K. E. Keller (3), and R. R. Martin

(1,2,3). (1) Molecular and Cellular Biology Program, Oregon State University, Corvallis, OR 97331; (2) Dept. Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331; (3) USDA-ARS, Corvallis, OR 97330. Phytopathology 92:S82. Publication no. P-2002-0596-AMA.

Pallidosis is a disease of strawberry first identified in 1957. A survey conducted in Maryland in 2000 revealed that 71% of field plants tested (grafted on indicator plants) were positive for pallidosis, suggesting its importance to the strawberry industry. Graft transmissibility and existence of dsRNA in infected plants indicate the viral nature of the disease. DsRNA was extracted from 22 infected plants and cloned. After sequencing ~100 clones we have identified 8 genes of a virus with homology to Lettuce infectious yellows virus, the type member of the *Crinivirus* group. The major coat protein is most closely related to Cucurbit yellows stunt disorder virus, another member of the group. Using the sequence information we developed a sensitive RT-PCR test that detected 37 of the 38 isolates of the virus available to us. We cloned and expressed the major coat protein of the virus and are now obtaining antibodies to the virus for developing a reliable ELISA test.

Interactions between *Pyricularia grisea* and *Rhizoctonia solani* in gray leaf spot pathosystem in perennial ryegrass turf. W. UDDIN, J. Shelton, and G. Viji. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802. Phytopathology 92:S82. Publication no. P-2002-0597-AMA.

Three avirulent isolates of *Rhizoctonia solani* from tall fescue, wheat, and poinsettia, were evaluated for suppression of gray leaf spot of perennial ryegrass (*Lolium perenne* L.) turf caused by *Pyricularia grisea*. Eight-week-old Legacy II perennial ryegrass plants were inoculated with *P. grisea* two days prior to, concurrently, or two days after spraying the plants with mycelia of the isolates of *R. solani*. Gray leaf spot developed in perennial ryegrass plants four days after inoculation with *P. grisea*. There were significant ($P = 0.05$) effects of timing of inoculation with *P. grisea* on gray leaf spot incidence (percent leaf blades symptomatic), and severity (index 0-10; 0=plants asymptomatic, 10 ≥ 90% leaf area necrotic). Disease incidence and severity in plants inoculated with *P. grisea* two days after spraying the mycelial suspension of *R. solani* were significantly lower than those of the plants inoculated concurrently, or two days prior to spraying *R. solani*. This study shows that *R. solani* interacts with *P. grisea* in the gray leaf spot pathosystem in perennial ryegrass turf.

Analysis of soil fungal community composition using an array-based oligonucleotide fingerprinting approach. L. VALINSKY (1), G. Della Vedova (2), M. Chrobak (1), T. Jiang (1), and J. Borneman (1). (1) University of California, Riverside; (2) Università degli Studi di Milano-Bicocca. Phytopathology 92:S82. Publication no. P-2002-0598-AMA.

Technological limitations currently hinder comprehensive descriptions of fungal community composition. In this report, we describe a new approach for analyzing fungal diversity and community composition termed oligonucleotide fingerprinting of ribosomal RNA genes (OFRG). OFRG is an array-based approach, which has been previously used for analysis of bacterial taxa. OFRG sorts arrayed ribosomal RNA gene (rDNA) clones into taxonomic clusters through a series of hybridization experiments, each using a single oligonucleotide probe. For this analysis, a simulated annealing algorithm was used to design a probe set consisting of 26 oligonucleotides. To demonstrate this approach, we analyzed 1510 fungal rDNA clones derived from soil. A large fraction of the clones were affiliated with the genera *Raciborskiomyces* (165 clones) and *Fusarium* (422 clones). Smaller assemblages of clones were affiliated with the *Alternaria*, *Ascobolus*, *Cephalophora*, *Chaetomium*, *Cryptococcus*, and *Rhizoctonia* clades.

Evolution of the resistance of *Botrytis cinerea* isolates to benzimidazole and dicarboximide in southwestern Spain. I. VALLEJO, M. Carbú, F. J. Fernández-Acero, L. Rebordinos, and J. M. Cantoral. Laboratory of Microbiology and Genetics, University of Cádiz, Spain. Phytopathology 92:S82. Publication no. P-2002-0599-AMA.

Botrytis cinerea Pers. is a phytopathogenic fungus which causes grey mould on over 230 hosts. The widespread use of fungicides, such as dicarboximides and benzimidazoles, in the prevention or elimination of fungal attack has resulted in the appearance of resistant strains. The goal of our study was to assess the sensitivity response to benzimidazoles and dicarboximides of 36 strains of *B. cinerea* isolated from strawberries cultured in Huelva (South Spain), in order to assess the level of resistance of isolates. Three phenotypes exhibiting resistance to the benzimidazole benomyl and carbendazim were detected in *B. cinerea* populations. No dicarboximide-resistant strains were found, which has been related to the heterokaryotic nature of the isolates and

the low fitness of the homokarotic resistant strains. All strains showed low or intermediate virulence on plant material and no relationships were established between sensitivity to fungicides and virulence of the strains.

Transformation of *Xylella fastidiosa* using replicative shuttle vector pUFR047. A. VANAMALA, R. Harakava, and D. W. Gabriel. Plant Molecular and Cell Biology Program, University of Florida, Gainesville, FL 32611. Phytopathology 92:S83. Publication no. P-2002-0600-AMA.

Xylella fastidiosa (Xf) is a xylem-inhabiting bacterium that causes serious diseases in a wide range of plant species. Two of the most serious of these are Pierce's Disease (PD) of grape and Citrus Variegated Chlorosis (CVC). Both PD and CVC genomes have been completely sequenced. Functional genomic analyses of Xf have been severely limited by lack of a stable replicative shuttle vector. pUFR047 is a small, stable, wide host range, conjugative, replicative, repW shuttle vector with a long history of use in *Xanthomonas*. pUFR047 was transferred into a rifampicin-resistant Florida PD strain, PD1A, by both conjugation and electroporation. pUFR047 was also electroporated into the sequenced California PD strain Temecula. Transfer by conjugation was inefficient due to background growth of *E. coli*. Transfer by electroporation occurred with or without DNA inserts into both PD strains at a frequency of ca. 50 transformants/ microgram DNA. The vector was replicative in both Xf strains and was reisolated after ten generations of growth and used to transform *E. coli*. Stability tests are underway.

Characterization of *Pantoea agglomerans* isolated from cranberry stem galls. A. VASANTHAKUMAR, V. M. Best, and P. S. McManus. Department of Plant Pathology, University of Wisconsin, Madison, WI 53706. Phytopathology 92:S83. Publication no. P-2002-0601-AMA.

Bacteria isolated from cranberry stem galls were identified as *Pantoea agglomerans* (formerly *Erwinia herbicola*) by 16S rRNA gene sequencing and physiological tests. A dose of 10^7 cfu or greater of *P. agglomerans* from cranberry (*Pac*) was required for symptom formation on micropropagated plants. *Pac* did not cause galls on Gypsophila or beet while both *E. herbicola* pv. *gypsophila* (*Ehg*) and *E. herbicola* pv. *betae* (*Ehb*) caused galls on micropropagated cranberry. Pulsed field gel electrophoresis of *Pac* DNA revealed a large plasmid (ca.195 kb). At high stringency, there was no hybridization between *Pac* DNA and probes based on hormone synthesis genes from known gall-forming bacteria. However, *Pac* produced similar amounts of IAA to *Ehg* and *Ehb* (ca. 2 micrograms per ml). There was no hybridization between *Pac* DNA and probes based on *hrp* genes and host specificity genes from *Ehg*. Our data indicate that the 195 kb plasmid in *Pac* is not closely related to the pathogenicity plasmids in *Ehg* or *Ehb*.

Segregation and characterization of two components within CTV isolate SY568 by aphid transmission. J. J. VELAZQUEZ-MONREAL, D. M. Mathews, and J. A. Dodds. Dept. of Plant Pathology, University of California, Riverside, CA 92521. Phytopathology 92:S83. Publication no. P-2002-0602-AMA.

Four subcultures of citrus tristeza virus (CTV) isolate SY568 obtained by grafting were used as inoculum sources for transmission to sweet orange by *Aphis gossypii*. Plants were tested using polyclonal (PAb) and monoclonal (MCA13) antibodies, growth, symptoms and aphid transmission rates were recorded, and RNase Protection Assays (RPA) were done using three different CTV probes: coat protein (CP), p65 HSP, and the 5' NTR. Aphid transmission ranged from 0-50% and symptoms ranged from asymptomatic to severe stunting and vein corking. Using the p65 RPA probe, 3 possible patterns were found indicating 2 independently segregating populations, components A, B, or A+B. A single plant with component A did not react with MCA13 and was asymptomatic. The A component has never been separated from B using graft transmission. All plants with the B or A+B components reacted with MCA13 and had severe symptoms. The A component accumulated to lower levels than B, but was found to attenuate the symptoms of B in some mixed inoculation treatments.

Fungal mannitol metabolism in plant-pathogen interactions. H. VÉLEZ, M. Ehrenshaft, J. D. Williamson, D. M. Pharr, and M. E. Daub. Depts. of Plant Pathology and Horticultural Science, NC State University, Raleigh, NC 27695. Phytopathology 92:S83. Publication no. P-2002-0603-AMA.

Many plants, fungi, and bacteria produce mannitol, a six-carbon sugar alcohol and a quencher of reactive oxygen species. Research in our labs suggests that mannitol is involved in host-pathogen interactions, as transgenic tobacco plants expressing a celery mannitol dehydrogenase and inoculated with *Alternaria alternata*, showed enhanced resistance to the fungus. We are investigating the role of mannitol in protection of A.

alternata against active oxygen-mediated plant defense responses by creating a fungal strain unable to synthesize mannitol. Fungal mannitol metabolism has been reported to occur via a cycle in which the enzymes mannitol dehydrogenase (MtDH) and mannitol-1-phosphate dehydrogenase (M1PDH) are involved in synthesis and degradation. Enzyme analysis of extracts of A. *alternata* confirmed that it contains an NADP-dependent MtDH, but not M1PDH. PCR with degenerate primers yielded a 252bp fragment with strong homology to published fungal MtDHs. Once the gene encoding MtDH is isolated, it will be disrupted, and the mutant strain tested for pathogenicity on tobacco.

Supervised control of *Cercospora beticola* in sugar beet using action thresholds and weather in the Netherlands. J. VEREIJSEN and J. H. M. Schneider. Institute of Sugar Beet Research (IRS), P.O. Box 32, 4600 AA, Bergen op Zoom, The Netherlands. Phytopathology 92:S83. Publication no. P-2002-0604-AMA.

Cercospora leaf spot (CLS), incited by the fungus *Cercospora beticola* Sacc. causes sugar yield losses up to 40%. In 1998, the IRS started to advise on the management of CLS using action thresholds. Defined stages of disease incidence or severity indicated a threshold for fungicide application. Action thresholds in the first year consisted of fungicide applications early (incidence) and late (severity) in the epidemic. In 2000, one action threshold was selected which, for two years, gave no statistical significant difference in sugar yield compared to fixed schedule spraying. The selected action threshold is defined as two early fungicide applications. In 2001, fungicide applications solely based on weather conditions were introduced. These objects were also sprayed twice, resulting in a higher relative sugar yield (122%), but not statistically significant different from the selected action threshold (116%). Both were significantly different from the unsprayed treatment (100%). These results indicate that both methods may be used to manage CLS in the Netherlands.

The effect of a North Carolina isolate of tomato spotted wilt virus on symptom development and gene expression in resistant and non-resistant cultivars of pepper. M. E. VIGIL (1), B. Bailey (2), and M. A. Smith (1). (1) Dept. Biology, North Carolina A & T State University, Greensboro, NC 27411; (2) Biocontrol of Plant Diseases Laboratory, ARS-USDA, Beltsville, MD 20705. Phytopathology 92:S83. Publication no. P-2002-0605-AMA.

Tomato Spotted Wilt Virus (TSWV) and related viruses infect over 900 species of plants including 99 species in 20 genera of the family Solanaceae, which includes the crop plants tomatoes, potatoes, tobacco and peppers. Among the many economically important pepper varieties and cultivars, very few are resistant to TSWV. However this resistance is not ensured because TSWV is an RNA virus, which can mutate easily and produce new strains. The development of more resistant cultivars is needed to control the spread of the virus and protect commercially important crops. Development of resistant plants by genetic engineers is possible and may be enhanced by increased understanding of strain diversity, virulence and the genes responsible for a host response. The purpose of this study was to determine the effect of an unknown NC isolate of TSWV on symptom development and gene expression in resistant and non-resistant cultivars of pepper. Two cultivars of resistant and non-resistant peppers were mechanically inoculated with the isolate of TSWV. Double antibody sandwich enzyme-linked immunosorbent assay (DAS ELISA) was used to test for viral infection. RNA was isolated from susceptible and control plants and used for fluorescence differential display. Non resistant pepper cultivars displayed typical symptoms of a systemic infection while the resistant cultivars exhibited a hypersensitive response. Preliminary results from differential display showed 40 differences in gene expression between the control and susceptible peppers. After repeating differential display, genes will be selected from susceptible and control plants and used as probes to test for the presence of those genes expressed in resistant cultivars. Support from U.S. Department of Education Graduate Assistance in Areas of National Need (GAANN) Fellowship Program grant # P200A980117 and Partnership for Underrepresented Scientist United for Education (PURSUE) grant # RFA GM-99-011 at UNC-Chapel Hill.

Evaluation of *Sclerotinia homoeocarpa* isolates from creeping bentgrass from various geographic regions in the United States. G. VIJI (1), W. Uddin (1), M. P. S. Camara (2), and N. R. O'Neill (2). (1) Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802; (2) Molecular Plant Pathology Laboratory, USDA-ARS, Beltsville, MD 20705. Phytopathology 92:S83. Publication no. P-2002-0606-AMA.

Sixty-seven isolates of *Sclerotinia homoeocarpa*, causing dollar spot in creeping bentgrass greens (*Agrostis palustris* Huds.), collected from various geographical regions in the U.S., were characterized by vegetative

compatibility, pathogenicity, and genetic similarity tests. Eleven vegetative compatibility groups were identified among the isolates, five of which were new. Results from the pathogenicity study indicated that there were significant differences ($P = 0.05$) in virulence among the isolates of *S. homoeocarpa* on Pennfine creeping bentgrass. Genetic analyses by Amplified Fragment Length Polymorphism (AFLP) revealed polymorphic DNA bands among the isolates, suggesting that the population is genetically diverse. Isolates collected from various golf courses exhibited unique AFLP patterns that clustered into several AFLP groups. The results of this study suggest that the causal agent of dollar spot consists of multiple genotypes.

Vertical redistribution of motile *Phytophthora parasitica* zoospores in water and implications for pathogen management in recycling irrigation systems. S. L. von Broembsen. Dept. Entomology & Plant Pathology, Oklahoma State University, Stillwater, OK 74078. Phytopathology 92:S84. Publication no. P-2002-0607-AMA.

Motile zoospores of *Phytophthora* spp. are known to accumulate at water surfaces because they are negatively geotropic. Vertical redistribution of motile zoospores of *Phytophthora parasitica* isolate GLN 9-3 was measured in 64 × 4 cm columns constructed from polyvinyl chloride pipe with rubber septa at 0, 30 and 60 cm heights to allow sampling using syringes. Columns filled with uniform suspensions of motile zoospores in water were sampled from 0 to 24 h. Samples were assayed by plating and counting germlings after 24 h. The number of viable propagules was proportionally greatest at the top (60 cm) of the water column after 0.5 to 1 h, became equally distributed by 6 h, and then greatest at the bottom by 24 h. Total viable propagules decreased to less than 10% by 24 h. Although water should not be drawn for reuse from the upper 60 cm of stored water, loss of viability may have more important implications for irrigation water management in recycling systems than vertical distribution.

A nitric oxide synthase in *Streptomyces turgidiscabies* affects thaxtomin biosynthesis. M. WACH (1), D. Gibson (2), and R. Loria (1). (1) Department of Plant Pathology, Cornell University, Ithaca, NY 14853; (2) USDA Agricultural Research Service, US Plant, Soil, and Nutrition Laboratory, Ithaca, NY 14853. Phytopathology 92:S84. Publication no. P-2002-0608-AMA.

We have identified a gene, *strNOS*, in the plant pathogen *Streptomyces turgidiscabies*, homologous to nitric oxide synthases (NOSs) from *Bacillus halodurans* and iNOS_{OX}, an inducible murine NOS. *strNOS* is conserved among plant pathogenic streptomycetes and is on a mobilizable pathogenicity island, upstream of a peptide synthetase which produces thaxtomin A and related congeners (cyclo-(L-4-nitrotryptophyl-L-phenylalanyl)). Eukaryotic NOSs produce active nitrogen species involved in cell signaling and host defense. *NOS* genes have been identified in Gram-positive bacteria, but the function of these genes is unknown. To determine if *strNOS* is involved in thaxtomin biosynthesis, *ie.*, through the nitration of thaxtomin precursors, we evaluated the effects of several characterized NOS inhibitors on thaxtomin A production by *S. turgidiscabies* and other pathogenic species. NOS inhibitors had no effect on bacterial growth but reduced thaxtomin production, indicating a role for *strNOS* in thaxtomin biosynthesis.

Role of watermelon blossoms in seed infection by *Acidovorax avenae* subsp. *citrulli*. R. R. WALCOTT (1), A. C. Castro (1), and R. D. Gitaitis (2). (1) Dept. Plant Pathology, University of Georgia, Athens, GA 30602; (2) Tifton, GA 31793. Phytopathology 92:S84. Publication no. P-2002-0609-AMA.

Bacterial fruit blotch (BFB), caused by *Acidovorax avenae* subsp. *citrulli*, is a major threat to watermelon production. The bacterium is seedborne but little is known about the mechanisms of seed infestation. To investigate the role of blossoms in seed infestation, watermelon plants were grown under greenhouse conditions. At anthesis, female blossoms were pollinated and inoculated with 0, 10⁷ or 10⁹ CFU of a green fluorescent protein mutant of *A. avenae* subsp. *citrulli* (AAC8-1ST) suspended in 0.1M phosphate buffered saline solution. At harvest maturity seeds were extracted from the resulting asymptomatic fruits and tested for the bacterium. While all seedlots from the control blossoms were negative for *A. avenae* subsp. *citrulli*, 31.8 and 40.3% of the seedlots from blossoms treated with 10⁷ and 10⁹ CFU AAC8-1ST, respectively, were infested. The average BFB transmission rates for seedlots from blossoms inoculated with 0, 10⁷ and 10⁹ AAC8-1ST CFU were 0, 7 and 8.7%, respectively. The data suggest that *A. avenae* subsp. *citrulli* can penetrate blossoms and infest seeds within symptomless fruits.

Jackbean accessions, as soil amendments, vary in suppressing root-knot nematode. J. T. WALKER (1) and J. B. Morris (2). (1) Dept. Plant Pathology, University of Georgia, Griffin, GA 30223; (2) USDA/ARS, Griffin, GA 30223. Phytopathology 92:S84. Publication no. P-2002-0610-AMA.

Jackbean, *Canavalia ensiformis*, is a valued cover crop and source of nutraceuticals, pharmaceuticals, and industrial products. Dried above-ground tissue of this legume as a soil amendment reduced *Meloidogyne incognita* infection of tomato in greenhouse studies. The Plant Genetic Resources Conservation Unit (USDA/ARS) has a collection of jackbean from diverse regions. We evaluated tissue from 16 accessions to determine if they varied in nematode suppressiveness. Ground tissue was mixed with infested (6000 eggs/kg) soil at 0, 1, and 2 percent, stored for one week, and then planted with Rutgers tomato. After eight weeks, tomato roots were examined and galls counted. No interaction occurred between tissue type and amendment rates. Five of 16 accessions reduced gall numbers. Rates affected gall numbers and plant heights, but not plant dry weights. Genetic variation in root-knot nematode suppressiveness occurs among jackbean accessions.

Effects of sand particle size on populations of the ring nematode, *Criconebella ornata*. N. R. WALKER (1) and D. L. Martin (2). (1) Dept. Entomology and Plant Pathology; (2) Dept. Horticulture and Landscape Architecture, Oklahoma State University, Stillwater, OK 74078. Phytopathology 92:S84. Publication no. P-2002-0611-AMA.

Sand used for golf course putting green construction can vary in particle size distribution. The effects of sand particle size on populations of ring nematode, *Criconebella ornata*, was evaluated in the greenhouse. Pots (10.5 cm × 10.5 cm × 9.5 cm h) containing autoclaved sand were planted with SR1020 creeping bentgrass. Course sand (particles between 1 - 2 mm), fine sand (less than 1 mm), and sand obtained from a commercial source were examined. Pots were inoculated with 75 nematodes per pot 30 days after planting. Turfgrass was maintained at 1 cm height and the study was terminated 40 days after inoculation. Turfgrass quality, rated on a scale of 1 to 5 where 1 = thin turf and 5 = thick, healthy turf, was greatest for pots containing fine sand (4.0) and lowest for course sand (3.1). Nematode populations were highest for fine sand (112/pot) and lowest for course sand (9/pot). These results indicate that sand particle size may influence nematode populations.

Genes for seedling and adult-plant resistance to leaf rust in soft red winter wheats. Y. A. WAMISHE, E. A. Milus, and K. C. Thomson. University of Arkansas, Fayetteville. Phytopathology 92:S84. Publication no. P-2002-0612-AMA.

The objective of this research was to determine the genetic basis for resistance to leaf rust among 116 contemporary genotypes of soft red winter wheat. Infection types produced on seedlings inoculated with 22 races of the leaf rust pathogen and incubated under controlled conditions were analyzed using a computer program that was developed to assist the identification of resistance genes. Fourteen genotypes had no seedling *Lr* genes. Seedling genes *Lr1*, *2a*, *2c*, *3*, *3ka*, *9*, *10*, *11*, *14a*, *18*, *23*, *24*, *26*, and *30* were identified, and 20 genotypes had unknown genes. Only the combination of *Lr9*, *24*, and *26* in five genotypes provided protection against all races. Genotypes also were evaluated for adult-plant resistance, and genes *Lr12*, *13*, and *34* were identified either singly or combined in about 50% of the genotypes. *Lr34* provided intermediate resistance by itself and higher resistance in combination with *Lr12* and *13*. These genes appear to provide resistance in the field, and some of the unknown genes detected in the seedling stage are likely *Lr13* or *34* that are also expressed at the seedling stage and interact with certain seedling genes.

Occurrence of the *ros* gene in marine organisms. S.-Y. Wang, N. Bouhouche and C. I. KADO. University of California, Davis. Phytopathology 92:S84. Publication no. P-2002-0613-AMA.

The chromosomal *ros* gene of *Agrobacterium tumefaciens* encodes a 15.5 kDa protein called Ros. Ros is a regulatory protein containing a novel C2H2 zinc finger that was thought to only occur in eukaryotic cells. Ros down regulates the *virC* and *virD* operons and the cytokinin synthesis gene, *ipt* on the Ti plasmid. Ros upregulates succinoglycan synthesis. Molecular evolutionary tree analysis revealed no candidate *ros* genes in plant genomes, but distant homologues were found in a marine fish. We analyzed a number of marine microorganisms and lower eukaryotes for the presence of *ros*. Southern hybridizations revealed novel *ros* counterparts in certain marine species. This discovery supports the data obtained from the e-tree analysis, and suggests a marine origin for *ros*.

Pathogenic variation of *Phoma* spp. from alfalfa and chemical control of *Phoma* pathogens in Alberta. H. WANG (1), S. F. Hwang (1), K. F. Chang (2), G. D. Turnbull (1), and R. J. Howard (2). (1) Crop and Plant Management, Alberta Research Council, Vegreville, Alberta T9C 1T4; (2) Plant Pathology, Crop Diversification Centre South, Brooks, Alberta T1R 1E6. *Phytopathology* 92:S85. Publication no. P-2002-0614-AMA.

Spring black stem and leaf spot of alfalfa, caused by *Phoma medicaginis* var. *medicaginis*, is one of the most common foliar diseases observed in Alberta. To determine the potential virulence of *Phoma* isolates from commercial alfalfa fields, inoculation experiments were conducted on selected alfalfa cultivars. In addition, fungicides were evaluated for efficacy against *Phoma* in laboratory trials. Three isolates of *Phoma* spp. were highly infective to most cultivars in an excised leaf study, although the cultivars Pickseed 3006, Algonquin and Anik exhibited consistently low disease levels. Cultivar 630 was the most susceptible amongst 15 cultivars inoculated with all isolates. Benomyl and propiconazole were highly effective at inhibiting *Phoma* growth, with EC₅₀ values ranging from 1.0 to 14.4 ng/mL. The inhibition effect declined slightly as the incubation time increased. Further results from greenhouse experiments will be presented.

Effect of *Crotalaria juncea* amendment on *Meloidogyne incognita*. K.-H. WANG and R. McSorley. Dept. Entomology and Nematology, University of Florida, Gainesville, FL 32611. *Phytopathology* 92:S85. Publication no. P-2002-0615-AMA.

Sunn hemp (*Crotalaria juncea*) (Cj), a tropical green manure crop, is recently recognized for its plant-parasitic nematode suppressive properties. Effects of Cj amendment on *Meloidogyne incognita* (Mi) infecting squash (*Cucurbita pepo*) were examined in 2 greenhouse tests. Objectives of these experiments are to determine if Cj amendment 1) enhances beneficial organisms, and 2) suppresses Mi in soils from various farming systems. The Cj-amended soil reduced root galls and enhanced ($P < 0.05$) plant weight of squash 8 weeks after nematode inoculation in both tests. Effects on Mi juveniles and free-living nematodes in soil varied between the 2 tests. Amendment of Cj enhanced nematode-trapping fungal (NTF) population densities in test 1 and a nematode-endoparasitic fungus, *Harposporium anguillulae*, in test 2. Soil collected from farming systems plots with yard waste compost had higher ($P < 0.05$) NTF population levels than soil from plots without yard waste compost. Performance of Cj amendment on Mi suppression is influenced by the availability of nematode-antagonistic organisms, which is often higher in soil with higher previous organic inputs.

Characterization of left end of the pathogenicity island of *Erwinia amylovora*. L. WANG (1), J. F. Kim (2), and S. V. Beer (1). (1) Department of Plant Pathology, Cornell University, Ithaca, NY 14853, USA; (2) Microbial Genomics Laboratory, Korea Research Institute of Bioscience and Biotechnology (KRIBB), P.O. BOX 115, Yusong, Taejeon 305-600, Republic of Korea. *Phytopathology* 92:S85. Publication no. P-2002-0616-AMA.

The left terminus of pathogenicity island (PAI) of *E. amylovora*, is clearly bounded by a tRNA gene. Eighteen open reading frames (ORFs) were identified between the *dspEF* operon and the left terminus of the PAI. The functions of the eighteen ORFs, except for *rlsA* a regulator for levan production, are not known. BLAST searches with this sequence (ca. 20 kb) showed that one of the ORFs is similar to plant chitinases and several ORFs showed homology to putative proteins from *Salmonella typhi* and/or *Yersinia pestis*. To determine if the left end of the PAI is involved in the virulence of *E. amylovora*, it was deleted by marker exchange. The deletion mutant was confirmed by PCR and Southern hybridization. The mutant was tested for pathogenicity on apple trees and for the ability to elicit the hypersensitive response on tobacco plants. It did not cause any symptoms on apple trees, but it elicited a hypersensitive response. Complementation tests with the deleted 20 kb fragment and portions of it are underway to identify ORFs critical to pathogenesis and virulence.

Construction of a secretory gland cell-specific cDNA library of *Heterodera glycines* through an amplified RNA approach. X. WANG (1), T. Maier (2), R. Hussey (3), T. Baum (2), and E. Davis (1). (1) Dept. Plant Pathology, North Carolina State University, Raleigh, NC 27695; (2) Dept. Plant Pathology, Iowa State University, Ames, IA 50011; (3) Dept. Plant Pathology, University of Georgia, Athens, GA 30602. *Phytopathology* 92:S85. Publication no. P-2002-0617-AMA.

Secretions from the esophageal glands of the soybean cyst nematode, *Heterodera glycines* play important roles in nematode parasitism of plants. mRNA from the microaspirated esophageal gland cell contents of the parasitic stages of SCN was isolated by biotinylated oligo-dT containing a T7 promoter sequence that was attached to streptavidin-coated beads and

used for initial cDNA synthesis. The synthesized cDNA products attached to the beads were used as templates to direct several rounds of in-vitro transcription using T7 RNA polymerase. The amplified RNA pool was utilized to construct a cDNA library without using PCR to obtain a more representative library. Sixty-seven unique sequences were obtained through sequencing of ninety randomly selected clones. Expressed sequence tag analysis of the mRNA gland cell-specific cDNA library and identification of potential gland secretion genes are being conducted

Apparent lack of strong selection pressure on non-functional *avrBs2* for change to a functional *avrBs2*. T. WANGSOMBOONDEE, P. B. Lindgren, and D. F. Ritchie. Dept. of Plant Pathology, NC State Univ., Raleigh, NC 27695. *Phytopathology* 92:S85. Publication no. P-2002-0618-AMA.

Mutations in the 5-bp repeat region of the avirulence gene *avrBs2* were detected in field strains of *Xanthomonas axonopodis* pv. *vesicatoria* (Xav) that defeat the *Bs2* resistance gene. We hypothesized that if the *avrBs2* gene is required for full fitness of Xav, in the absence of selection pressure by the *Bs2* resistance gene, functional *avrBs2* should be favored. Seven strains representing races 4, 5, and 6 resistant to rifampicin were tested for the potential to change from non-functional to functional *avrBs2*. This hypothesis was tested in the laboratory using broth cultures and potted pepper plants and in a field experiment. No strains with functional *avrBs2* were detected in the laboratory experiments. Although 3 (0.16%) of 1900 colonies from the field experiment had functional *avrBs2*, these strains did not result in a detectable population shift. This suggests that the selection pressure for functional *avrBs2* in the absence of resistance gene *Bs2* is weak or absent.

Pathogenicity region of *Streptomyces*, cause of potato common scab. L. A. Wanner. USDA-ARS Vegetable Lab, Beltsville, MD 20705. *Phytopathology* 92:S85. Publication no. P-2002-0619-AMA.

Common scab is the fourth most important potato disease. Symptoms reportedly vary on the same cultivar grown in different locations. Though no cultivar is immune, there are differences in resistance among cultivars. Streptomycetes are a diverse group of soil-inhabiting gram positive bacteria. Most are not plant pathogens. Those that cause scab are phylogenetically diverse, but nearly all produce the toxin thaxtomin. Pathogenicity seems to result from a pathogenicity island (PAI) containing a gene for a pathogenicity factor (*necl1*) within a >26 kb conserved region that may be horizontally transferred into distantly related streptomycetes to produce new plant pathogenic strains (1, 2). We isolated streptomycetes from scabby potatoes from several regions of the US and assessed their pathogenicity in radish and potato. Many of our isolates do not produce the pigment melanin. We are analyzing variability in the PAI using PCR with the *necl1* gene as anchor point. We plan to identify additional cultural or environmental factors contributing to variability in scab morphology and susceptibility in different cultivars. (1) Bukhalid et al. 1998 *MPMI* 11:960-7; (2) Bukhalid et al. 2002 *AEM* 68: 738-44.

***Phytophthora cinnamomi* on loblolly decline sites in Alabama.** A. Weber (1), J. JONES (1), N. Hess (2), E. Carter (3), and J. Stienman (4). (1) Dept. Plant Path. and Crop Phys., La. Ag. Expt. Stn, LSU Ag. Ctr, Baton Rouge, LA; (2) US Forest Service, FHP, Pineville, LA; (3) US Forest Service, SRS, Auburn University, AL; (4) US Forest Service, FHM, Asheville, NC. *Phytopathology* 92:S85. Publication no. P-2002-0620-AMA.

Loblolly decline resembles littleleaf disease of shortleaf pine and has been considered the same disease, implying a role for *Phytophthora cinnamomi* (Pc) in the etiology of loblolly decline. Thirty-nine plots in Alabama were selected that showed a range of symptoms. Pc was isolated from the rhizosphere of ~50% of these plots, but not from roots. This is consistent with the literature of littleleaf in which Pc is rarely found in roots. Results of a pathogenicity study were varied, but indicated that Pc isn't pathogenic to loblolly seedlings. Pc was recovered from roots and soil of inoculated seedlings, and did reduce height and crown diameter. Pc isolations did correlate with certain hardwoods, so we did pathogenicity tests with loblolly and shortleaf pine as well as oak, dogwood, and sweetgum, the hardwoods most numerous on loblolly decline sites where Pc was isolated from the soil. We have also returned to the sites to isolate from hardwood roots and rhizosphere to determine presence of Pc.

Antifungal activity of naturally occurring quinones. D. E. WEDGE (1), F. E. Dayan (1), and G. Meazza (2). (1) USDA-ARS, Natural Products Utilization Research Unit, The National Center for Natural Products Research, University, MS 38677; (2) Isagro Ricerca Srl, via G. Fauser, 4, 28100 Novara, Italy. *Phytopathology* 92:S85. Publication no. P-2002-0621-AMA.

Increasing incidence of chemical resistance in fungal plant pathogens and loss of available fungicides for disease control are two factors that drive the need to search for new plant protectants. As part of a program to discover natural product-based fungicides with low environmental and mammalian toxicity, we evaluated the antifungal activity of several classes of naturally occurring phenolics, especially quinones, and other related compounds. Bioautography provides a rapid and simple technique to visually screen large numbers of compounds or extracts for antifungal activity against *Colletotrichum* spp. Quinones are common in nature, occurring as benzo-, naphtho- and anthraquinones in higher plants. Naphthoquinones are often responsible for the pigmentation of colored heartwood and bark. The compounds tested belong to the class of 1,4-naphthoquinones, 1,2-naphthoquinones, 1,4-benzoquinones, anthraquinones, and other miscellaneous compounds. About half of the 46 quinones tested demonstrated moderate to good antifungal activity against the *Colletotrichum* test fungi. *C. fragariae* appeared to be the most sensitive species to quinone-based chemistry, and *C. acutatum* was the most resistant to these compounds. *C. gloeosporioides* often demonstrated intermediate sensitivity.

Small RNA production and transgene methylation in gene silenced transgenic plants expressing sequence of red clover necrotic mosaic virus. Z. WENG and Z. Xiong. Dept. of Plant Pathology, Univ. of Arizona, Tucson, AZ 85721. Phytopathology 92:S86. Publication no. P-2002-0622-AMA.

RCNMV is a small virus with a bipartite RNA genome. Transgenic lines of *Nicotiana benthamiana* expressing the 5'1.2 kb of RNA-1 exhibited either self-activated (in pre-silenced lines) or inducible (in inducible lines) gene silencing against RCNMV infection. To further understand the mechanisms of gene silencing, production of small RNA homologous to the transgene and transgene methylation were analyzed. While the transgene-specific small RNA was detected in the pre-silenced lines, no such small RNA was found in the inducible lines prior to RCNMV infection. The transgene was methylated in the pre-silenced lines but not in the inducible lines. However, the transgene became methylated in the inducible lines after recovery from RCNMV infection. These data indicated the positive correlations between gene silencing and production of the transgene-specific small RNA and between gene silencing and transgene methylation. The effect of plant developmental stage on the activation of gene silencing is also analyzed.

Timing of spore release by *Colletotrichum acutatum* in Michigan blueberry fields. P. S. WHARTON, J. S. Dickman, and A. M. C. Schilder. Dept. Plant Pathology, Michigan State University, East Lansing, MI 48824. Phytopathology 92:S86. Publication no. P-2002-0623-AMA.

To determine the timing of spore dispersal by *Colletotrichum acutatum*, causal agent of blueberry anthracnose, spore traps were placed in one blueberry field in SW Michigan in 2000 and two fields in 2001. Traps consisting of 15 cm-diameter funnels attached to 2-L plastic bottles were placed inside the canopy of four bushes per field. Rain water in the traps was collected every week from late April/early May until early/mid August. Samples were concentrated by centrifuging and numbers of spores were counted using a hemacytometer. Two main peaks of inoculum production occurred during the growing season: one early on (before and during bloom) and a smaller peak later in the season when ripe fruit was present. The later peak presumably is the result of secondary sporulation on berries infected earlier in the season. These results correlated well with observations from a field trial in which two early sprays of captan + benomyl (at green tip and bloom) significantly reduced (by 62%) the incidence of anthracnose fruit rot. They also emphasize the importance of protecting fruit with fungicides when inoculum levels may be high.

ropA, a negative regulator of phenazine biosynthesis in *Pseudomonas aureofaciens* 30-84. C. A. WHISTLER (1) and L. S. Pierson III (2). (1) University of Hawaii, Honolulu, HI 96813; (2) Dept. Plant Pathology, University of Arizona, Tucson, AZ 85721. Phytopathology 92:S86. Publication no. P-2002-0624-AMA.

Pseudomonas aureofaciens 30-84 suppresses take-all disease of wheat through the production of phenazine antibiotics. Although much is known about regulatory systems that stimulate phenazine gene expression, such as PhzR/PhzI and GacS/GacA, little is known regarding negative regulation. Tn5 mutagenesis of a 30-84 derivative containing a *phzB::lacZ* fusion identified a mutant with increased *phzB* expression. Insertion of the mutated region into the wild type 30-84 genome also resulted in increased phenazine production. Sequence analysis of the mutated region identified a gene named *ropA* (repressor of phenazine). The predicted sequence of RopA shared similarity with *E. coli* RstB, a putative two component regulator of unknown function. The *ropA* phenotype includes phenazine gene expression at lower

cell densities without an increase in *N*-acyl homoserine lactone production. The mutation does not restore phenazine production to *gacS/gacA* mutants and its effect is greatest in minimal medium.

Baculovirus expression of soluble TSWV glycoproteins. A. E. WHITFIELD (1), D. E. Ullman (2), and T. L. German (1). (1) Dept. of Entomology, University of Wisconsin, Madison, WI 53706; (2) Dept. of Entomology, University of California, Davis 95616. Phytopathology 92:S86. Publication no. P-2002-0625-AMA.

Tomato spotted wilt virus (TSWV) is an enveloped plant virus with a worldwide distribution and large plant host range. Two transmembrane glycoproteins (G1 and G2) decorate the surface of the virion. We developed baculoviruses to produce large amounts of the glycoproteins for subsequent experiments designed to determine the roles of G1 and G2 in TSWV acquisition by its thrips vector. Soluble forms of both glycoproteins (G1 and G2) were expressed individually from recombinant baculoviruses in SF21 cells. The proteins were expressed with a hexahistidine tag at the C-terminus for affinity purification. When analyzed by Western blot, the soluble proteins were recognized by monoclonal antibodies raised against the glycoproteins. These proteins will be valuable tools to characterize the events that are involved in the acquisition of TSWV by its primary insect vector (*Frankliniella occidentalis*).

Pathogenic and genetic relationships among strains of *Xylella fastidiosa* isolated from various hosts. R. L. Wichman, C. M. Thompson, and D. L. HOPKINS. Mid-Florida REC, University of Florida, Apopka, 32703. Phytopathology 92:S86. Publication no. P-2002-0626-AMA.

Diseases are produced in a wide range of hosts by strains of *Xylella fastidiosa*. However, some strains are host specific, indicating that different pathotypes or subspecies may exist. In this study, strains isolated from grapevine, elderberry, oak, and oleander were characterized by host range, by SDS-PAGE of proteins, and by pulsed field electrophoresis of DNA digested with rare-cutting restriction endonucleases. The strains of *X. fastidiosa* from grapevine, oak, and oleander were most virulent on their host of origin and also were separated into three similar, but distinct clusters based on cluster analysis of DNA fingerprints. Strains from elderberry were not distinct from the grapevine strains. One strain originally isolated from elderberry was more virulent on grapevine than elderberry. DNA fingerprinting of the *X. fastidiosa* strains separated them into groups that corresponded with host range results. This supports the separation of these strains into three or four pathovars of *X. fastidiosa*.

Multi-locus sequencing typing and single nucleotide polymorphism (SNP) genotyping of *Xylella fastidiosa* from citrus and coffee in Brazil. E. WICKERT (1), P. C. Ceresini (2), M. A. Machado (3), and E. G. M. Lemos (1). (1) Dept. Tecnologia, UNESP - Universidade Estadual Paulista, Jaboticabal, SP 14884-900; (2) Dept. Fitossanidade, UNESP, Ilha Solteira, SP 15385-000; (3) IAC - Instituto Agronomico de Campinas, Cordeiropolis, SP 13490-970, Brazil. Phytopathology 92:S86. Publication no. P-2002-0627-AMA.

Multi-locus sequencing typing and single nucleotide polymorphisms (SNPs) were used to evaluate the molecular evolution of genes and the genetic diversity of 40 Brazilian isolates of *Xylella fastidiosa* (*Xf*) from citrus and coffee. Other five isolates from grapes, *Morus rubra*, or plums were added to the sample. The hypothesis that populations of *Xf* from citrus and coffee are not genetically subdivided was tested. SNPs were detected in 17 out of 20 randomly chosen loci of *Xf*. None of the molecular haplotypes of *Xf* was shared by different host populations. This indicates complete absence of gene flow between citrus and coffee populations of *Xf*. However, the reconstruction of haplotypes network by statistical parsimony supported the hypothesis of contemporaneous evolution of *Xf* genes from citrus and coffee.

Effect of pre-treatment with benzoic acid on disease development on stems of *Banksia attenuata* inoculated with *Phytophthora cinnamomi*. M. G. WILLIAMS (1), T. Senaratna (1,2) and K. Sivasithamparam (1). (1) Dept. Soil Science and Plant Nutrition, University of Western Australia, Nedlands, WA 6907, Australia; (2) Research Laboratory, Kings Park and Botanic Garden, West Perth, WA 6005, Australia. Phytopathology 92:S86. Publication no. P-2002-0628-AMA.

The use of a host's own resistance shows great potential for the control of the plant pathogen *Phytophthora cinnamomi*. *Banksia attenuata* plants were pre-treated with foliar sprays or soil drenches of benzoic acid (BZA) to determine if the compound could confer resistance in the host. Plants were stem inoculated with *P. cinnamomi* 1 week after pre-treatment. An estimate of resistance was determined by measuring *P. cinnamomi* lesions on stems.

All soil drench and foliar spray pre-treatments with 0.10, 0.25 or 0.50mM BZA caused a reduction in lesion sizes compared to control plants. Soil drenching with 0.50mM BZA was the most effective treatment. Biochemical assays of leaf tissue from 0.50mM BZA treated plants revealed enhanced activity (45% more) of the defence enzyme peroxidase and an overall increase (100%) in total protein content. Thus, BZA affects pathogen development, presumably by stimulating defence mechanisms in the host.

Sensitivity of *Ceratocystis fimbriata* f. sp. *platani* and *Botryosphaeria rhodina* to triazole fungicides. A. D. WILSON, T. D. Leininger, and C. S. Oberle. USDA Forest Service, Southern Hardwoods Laboratory, P.O. Box 227, Stoneville, MS 38776. Phytopathology 92:S87. Publication no. P-2002-0629-AMA.

Canker stain, caused by *Ceratocystis fimbriata* f. sp. *platani*, and sycamore anthracnose, caused by *Botryosphaeria rhodina*, are among a complex of dieback and decline diseases that affect sycamore pulpwood production in southern fiber-farm plantations. Five triazole fungicides, including difenoconazole, myclobutanil, propiconazole, terbuconazole, and triadimefon, were tested in vitro for their effectiveness in inhibiting growth of these fungal pathogens. Minimum effective concentrations (MECs) required to totally inhibit growth were higher (>500 ppb) than levels needed to inhibit growth of *Ceratocystis fagacearum*. *Ceratocystis fimbriata* strains were more sensitive than *Botryosphaeria rhodina* strains. This information will be used to test dosages of these fungicides applied in automated fertigation (combined fertilization/irrigation) systems. The use of permanently-installed, metered fertigation systems are particularly well suited for application of protective systemic fungicides during times when fertilizers are not being applied in irrigation water.

Host specific differences in preharvest grain infection by toxigenic fungi in dryland pearl millet and corn. J. P. WILSON (1), W. W. Hanna (1), D. M. Wilson (2), and A. E. Coy (3). (1) USDA-ARS Crop Genetics & Breeding Res. Unit; (2) Dept. Plant Pathology; (3) Dept. Crop & Soil Sciences, Univ. Georgia, Coastal Plain Expt. Stn., Tifton, GA 31793. Phytopathology 92:S87. Publication no. P-2002-0630-AMA.

Pearl millet is a promising alternative feed grain for the Southern Coastal Plain. Preharvest grain infection by potentially toxigenic fungi in pearl millet and corn were compared in 2000 and 2001. Hybrids were grown in dryland conditions at several planting dates to allow variation in flowering time. Grain was harvested and fungi were isolated in the laboratory. Fungal isolation differed by host species in both years. Across years, mean isolation frequencies of *Aspergillus flavus*, *Fusarium verticillioides*, *F. semitectum*, and *F. chlamyosporum* from corn were 4.3, 62.3, 0.1, and 0.0%, respectively; those from pearl millet were 0.1, 0.3, 55.9, and 23.8%. Isolations of *F. verticillioides* and *F. semitectum* differed by planting date. In 2000, aflatoxins in corn and pearl millet averaged 135.6 and 0.1 ppb, and fumonisins averaged 6.1 and 0.0 ppm, respectively. Differences in preharvest mycotoxins are likely due to host specific differences in preharvest fungal infection of the grain.

Granular application of *Aspergillus flavus* as an inoculation technique of corn in the field. G. L. WINDHAM (1), W. P. Williams (1), P. M. Buckley (1), and H. K. Abbas (2). (1) USDA, ARS, Mississippi State, MS; (2) USDA, ARS, Stoneville, MS. Phytopathology 92:S87. Publication no. P-2002-0631-AMA.

Field studies were conducted to compare conventional *Aspergillus flavus* inoculation techniques with granular application techniques. Conventional inoculation techniques included injecting spores under husks using the side-needle technique or spraying spores on ears weekly for 5 weeks with a Solo backpack sprayer. The granular inoculations consisted of broadcasting *A. flavus* infected wheat in field plots. Granular applications were made 2 weeks prior to mid silk, at mid silk, or 2 weeks after mid silk. Ears inoculated with the side-needle technique had significantly higher levels of aflatoxin compared to ears inoculated using all other techniques. Aflatoxin levels were similar in plots sprayed with *A. flavus* and in plots treated with infected wheat 2 weeks prior to mid silk. In a late planted study, aflatoxin levels were highest in plots inoculated using the side-needle technique and infected wheat at 2 weeks prior to mid silk. Corn from plots inoculated with wheat at mid silk had higher levels of aflatoxin compared to corn from plots sprayed with *A. flavus*. Application of *A. flavus* infected wheat prior to mid silk may be a useful tool in evaluating corn for aflatoxin resistance.

A qualitative risk assessment regarding use of gentamicin to control fire blight disease and human health implications. V. W. Winkler and G. L. MELCHIOR. Gowan Company. Phytopathology 92:S87. Publication no. P-2002-0632-AMA.

It has been suggested that use of gentamicin to control fire blight disease on apples will select for bacteria that are resistant to gentamicin. There has been further speculation that gentamicin-resistant genes resulting from the use of gentamicin as a plant protection agent could be transferred to bacteria that infect humans, which would decrease the effectiveness of gentamicin as a human antibiotic. In response to these concerns, we present a detailed analysis of the risks associated with 1) occupational exposure to gentamicin, 2) dietary exposure to gentamicin, and 3) potential dietary exposure to gentamicin-resistant bacteria. We will also address concerns expressed by the EPA and an interagency panel. In the final analysis these risks are negligible so that the proposed use meets the reasonable certainty of no harm for FFDA, and exceeds the bar of risk versus benefit requirements of FIFRA.

Survival and dispersal of *Colletotrichum acutatum* on strawberry leaves under field conditions. K. A. WISE, L. F. S. Leandro, S. N. Wegulo, and M. L. Gleason. Dept. Plant Pathology, Iowa State University, Ames, IA 50011. Phytopathology 92:S87. Publication no. P-2002-0633-AMA.

Survival and dispersal of *Colletotrichum acutatum* on strawberry (cv. Tristar) were observed in an Iowa field for 7 wks. Strawberry plants in a 9-m² area in the center of a plot were spray-inoculated before bloom. Inoculated and non-inoculated leaves from the inoculated area and leaves from adjacent sampling areas were sampled weekly. Inoculated leaves were observed microscopically, and leaflets from all areas were observed for presence of acervuli after freezing and incubation. Fruit developing after inoculation were assessed for symptoms. Conidia populations on leaves changed significantly ($P < 0.05$) over time. The number of appressoria peaked 3 wks after inoculation. Acervuli were found on inoculated and non-inoculated leaves in the inoculated area for 7 wks, and on leaves from non-inoculated areas 3 wks after inoculation. Anthracnose fruit rot was present in the inoculated area and two adjacent sampling areas 3-4 wks after inoculation. Results indicated that *C. acutatum* can survive on symptomless leaves under field conditions and that asymptomatic leaves may harbor inoculum for fruit rot epidemics.

Effects of two soil-borne viruses of sugarbeet and their fungal vector, *Polymyxa betae*, on virus accumulation and plant growth in sugarbeet. G. C. Wisler (1), R. T. Lewellen (2), J. L. Sears (2), J. Wasson (2), H.-Y. Liu (2), and W. M. WINTERMANTEL (2). (1) Department of Plant Pathology, University of Florida, Gainesville, FL 32611; (2) USDA-ARS, Salinas, CA 93905. Phytopathology 92:S87. Publication no. P-2002-0634-AMA.

Soils naturally infested with cultures of avirulent *Polymyxa betae* and *P. betae* infected with the two sugar beet benyviruses BNYVV and BSBMV, alone and in combination, were compared to non-infested soil with regard to their effects on virus content, fresh plant weight, and seedling emergence. Both a variety with resistance to BNYVV (Rz), the cause of Rhizomania, and a Rhizomania-susceptible variety (rz) were examined. These studies clearly demonstrated that the Rz resistance gene does not confer resistance to BSBMV. Additionally, *P. betae* alone had a significant negative effect on growth of sugarbeet. BSBMV titers were significantly higher in single infections than in mixed infections with BNYVV in both resistant and susceptible varieties. In contrast, BNYVV titers were very high in single and in mixed infections in the Rhizomania-susceptible variety, but low in the resistant variety. It appears that even at low levels, BNYVV either out competes or suppresses BSBMV.

Nitric oxide synthase in higher plants: A novel protein different from NOS in animals. M.-Y. WONG, C.-C. Chou, and J.-S. Huang. Department of Plant Pathology, North Carolina State University, Raleigh, NC 27695. Phytopathology 92:S87. Publication no. P-2002-0635-AMA.

Nitric oxide (NO) is a signaling molecule involved in intercellular communication and immune responses in mammalian cells. It is produced during the conversion of L-arginine to L-citrulline catalyzed by nitric oxide synthase (NOS). The presence of NO and its involvement in disease resistance in higher plants have been demonstrated. Hitherto NOS has not been isolated from higher plants. Our attempts to isolate a protein from pea with NOS-like activity indicate that the protein is novel and significantly different from animal NOS. Immunoblot revealed no specific band for NOS when probed with rabbit anti-brain NOS, anti-endothelial NOS and anti-macrophage NOS antibodies but several positive bands that were not animal NOS. The pea protein does not bind to 2',5'-ADP Sepharose 4B affinity gel, in contrast to animal NOS, implying the absence of NADPH domain. Also, in contrast to animal NOS, unidentified small molecular cofactor(s) from the crude extract is(are) required to restore partially or fully the pea NOS activity after partial purification. Other characteristics of the protein are also discussed.

Evaluating the efficacy of *Arthrobotrys oligospora* to reduce plant parasitic nematodes in putting greens. J. E. WOODWARD (1), N. R. Walker (1), J. W. Dillwith (1), D. L. Martin (2), and H. Zhang (3). (1) Dept. Entomology & Plant Pathology; (2) Dept. Horticulture & Landscape Design; (3) Dept. Plant & Soil Sciences, Oklahoma State University, Stillwater, OK 74078. Phytopathology 92:S88. Publication no. P-2002-0636-AMA.

Plant parasitic nematodes are common in golf course putting greens, but few options exist to manage nematodes in established greens. The objective of this study was to determine the efficacy of the nematophagous fungus, *Arthrobotrys oligospora*, to reduce nematode populations in simulated putting green soils in the greenhouse. Pots (10.5 cm × 10.5 cm × 9.5 cm h) containing 1100 g sand were planted with creeping bentgrass (*Agrostis palustris*) cv. SR1020 seed. Fifty days after planting, pots were inoculated with 100 *Criconebella ornata* /pot and 20 *A. oligospora* conidia /g of soil. Pots were arranged in a randomized complete block design with seven replications. The study was terminated 70 days after inoculation and was repeated once. Results and implications of this study for the management of plant parasitic nematodes in golf course putting greens will be presented.

Sclerotial survival of *S. minor* and *S. sclerotiorum* in California. B. M. WU and K. V. Subbarao. Dept. of Plant Pathology, Univ. of Cali. Davis, Salinas, CA 93905. Phytopathology 92:S88. Publication no. P-2002-0637-AMA.

Significant yield losses result from lettuce drop, caused by *Sclerotinia minor* and *S. sclerotiorum* in the Salinas and central valleys of California, respectively. To understand if the predominance of *S. sclerotiorum* in the central valley is due in part to the better sclerotial survival of *S. sclerotiorum* in the summer when soil temperatures reach >30°C and no lettuce is grown there, sclerotial survival of the two species was compared in laboratory and in commercial fields. Results from laboratory experiments demonstrated that >90% sclerotia of both species survived for 3 months in sterilized dry soils at temperatures from 15 to 40°C. When sclerotia were buried at depths of 5 and 15 cm in fields in the central valley, >50% of *S. sclerotiorum* compared with only 20% of *S. minor* remained viable after 24 wk. In the Salinas valley, however, no consistent differences were observed in survival of the two species. The results explain in part why *S. minor* is less prevalent in the central valley of California. Factors contributing to the poor survival of *S. minor* in the central valley and low infection by *S. sclerotiorum* in the Salinas valley are being investigated.

Pathogenicity and infection courts of *Phacidiopycnis piri* in pears. C. L. XIAO and R. J. Boal. Washington State Univ., TFREC, Wenatchee. Phytopathology 92:S88. Publication no. P-2002-0638-AMA.

Phacidiopycnis piri is the causal agent of Phacidiopycnis fruit rot, a newly recognized postharvest disease of pears in U.S. Currently there is little information on *P. piri* as a decay-causing pathogen in stored pears. Thirteen isolates derived from single pycnidiospores and ascospores of the fungus recovered from orchards were tested for their ability to cause fruit rot at 0°C. To determine infection courts of the fungus in d'Anjou pears, decayed fruit were sampled from storage and sorted by symptoms. Isolation was made from decayed fruit to confirm causal agents. In the laboratory, fruit were inoculated shortly after harvest with spore suspensions of the fungus at stem, calyx, and fruit surfaces with or without wounding and then stored at 0°C. All isolates tested were pathogenic to pear fruit at 0°C. *P. piri* caused stem-end rot, calyx-end rot, and wound-associated rot in stored pears. It took 3 to 4 months at 0°C for the fungus to ramify the stem tissues, reach fruit flesh and cause fruit rot. No fruit rot was observed in non-wounded fruit inoculated with spores of the fungus. Laboratory experiments and storage observations indicated that fruit-to-fruit spread by mycelia of the fungus also occurred in storage.

Prevalence and incidence of Phacidiopycnis rot, a newly recognized postharvest disease of pears in U.S. C. L. XIAO (1), R. J. Boal (1), and R. G. Roberts (2). (1) Washington State Univ., TFREC; (2) USDA ARS, Wenatchee. Phytopathology 92:S88. Publication no. P-2002-0639-AMA.

Phacidiopycnis rot, caused by *Phacidiopycnis piri*, is a newly recognized postharvest disease on pears in U.S. During Mar to May 2001 (late storage) and Nov 2001 to Jan 2002 (early storage), decayed d'Anjou pears were randomly sampled from different lots to determine the incidence of the disease. *P. piri* caused three types of symptoms, i.e. stem-end rot, calyx-end rot and wound-associated rot. Symptoms in the early stage were very similar to those caused by *Botrytis cinerea*. During early storage, fruit from 30 of 33 orchards had this disease, which accounted for 20% of decayed fruit sampled, ranging from 2 to 49%. During late storage, fruit from 22 of 26 orchards had this disease. The disease accounted for 42% of decayed fruit

from conventional orchards, ranging from 17 to 71% and 20% of decayed fruit from organic orchards, ranging from 5 to 42%. Most of Phacidiopycnis rot that occurred during early storage originated from wound infections of fruit; while the fruit rot during late storage mostly originated from stem infections. Calyx-end rot caused by *P. piri* accounted for 3 to 33% of the Phacidiopycnis rot in 8 of 22 orchards.

Intraspecific variation of ITS rDNA among geographical isolates of the soybean cyst nematode *Heterodera glycines*. Y. XU (1), S. Bekal (2), A. Colgrove (2), and T. L. Niblack (2). (1) Heilongjiang Institute of Agricultural Modernization, CAS, Division of Crop Sciences, China; (2) Dept. Crop Sciences, University of Illinois, Urbana, IL 61801. Phytopathology 92:S88. Publication no. P-2002-0640-AMA.

The internal transcribed spacers (ITS) of the ribosomal genes are frequently used in nematode diagnostics. We used polymerase chain reaction with ITS universal primers to amplify ITS1 and ITS2 including 5.8 S of eighteen soybean cyst nematode (SCN) isolates, *Heterodera glycines*, originating from Argentina, Canada, China and the United States. A 1Kb product was amplified from all SCN isolates; which was then digested with fourteen different restriction enzymes. Seven restriction enzymes generated cleaved fragments among the SCN isolates but only *Ava*I and *Eco*RI showed polymorphism between the Chinese SCN isolates and all other SCN isolates. Sequence analysis of both ITS regions from one Chinese isolate used in this study showed higher variation in ITS1 than ITS2 when compared with both ITS sequences of the US SCN isolates deposited in GenBank data base.

Virulence of wheat yellow (stripe) rust disease in western and central Asia. A. H. Yahyaoui. International Center for Agricultural Research in the Dry Areas. Phytopathology 92:S88. Publication no. P-2002-0641-AMA.

Surveys of pathogen populations and the genetic characterization of resistance provide valuable information for plant breeders. Yellow rust populations were characterized using a trap nursery including available differential sets, Avocet near-isogenic lines, and lines with known resistance genes. The nursery was planted at 30 sites in twelve countries for three consecutive seasons. Each line's response to yellow rust was evaluated at adult growth stage. The lines 'Morocco' and 'Avocet S' were used as susceptible controls. The remaining genotypes exhibited different degrees of resistance to natural populations of *Puccinia striiformis* f. sp. *tritici*. Avocet near-isogenic lines permitted assessment of the reaction of genotypes to yellow rust compared to other differential lines. Virulence on *Yr10* was noted in Syria. Virulence on *Yr1* and *Yr10* was observed in Tajikistan. Virulence on *Yr9* was found at all the testing sites except those in Turkey and Azerbaijan. *Yr5* remains as an effective resistance gene in this region. This pathotype diversity has important implications for regional plant breeding projects.

Distribution of *Ceratocystis fagacearum* in actively wilting red oaks. M. YAMATO (1), J. Juzwik (2,3), and K. W. Kromroy (3). (1) University of Tokyo, Japan; (2) USDA Forest Service, North Central Research Station, St. Paul; (3) Dept. Plant Pathology, University of Minnesota, St. Paul, MN 55108. Phytopathology 92:S88. Publication no. P-2002-0642-AMA.

Transpirational flow of water in oak wilt-affected trees is disrupted as the pathogen, *Ceratocystis fagacearum* (CF), invades xylem vessels in the outer sapwood and the host responds to invasion, resulting in crown wilt. The distributions of viable CF in xylem tissue of two infected northern red oaks (85 and 60 percent crown wilt) were investigated. Dye was injected intravascularly into root flares to show functioning xylem, and trees were felled 8-15 days after injection. Cross sections (4 cm thick) of the main stem(s) were cut at 1-m intervals from the base to the top of each tree and isolations for CF attempted from fungus-associated stained, green-dyed, and clear tissue of the sapwood. CF was recovered throughout the height (11 and 17-m) of both infected trees. Fungus stained xylem yielded CF more often than dyed or clear xylem. The dye failed to consistently indicate functioning xylem. Isolation frequencies were highest (34 – 88 percent) for the most severely wilted tree compared to the other one (7 – 50 percent).

Genomic diversity of *Erwinia carotovora* subsp. *carotovora*. M.-N. YAP and A. O. Charkowski. Dept. Plant Pathology, Univ. Wisconsin, Madison, WI 53706. Phytopathology 92:S88. Publication no. P-2002-0643-AMA.

Over 40 serotypes of *Erwinia carotovora* subsp. *carotovora* (Ecc) have been described, but only a few of these serotypes are typically isolated from potatoes. Our objective is to develop DNA-based techniques to identify Ecc strain types that are highly virulent and commonly found on potato and to characterize the genes that promote the virulence of these types. We isolated Ecc strains varying in virulence from diseased potatoes in 2001 and used a

variety of DNA-based and biochemical techniques to characterize them. We sequenced a portion of the malate dehydrogenase gene, a housekeeping gene, from 23 strains and found that the sequences ranged from 93% to 100% identical. In comparison, the homologous *mdh* region from diverse *Escherichia coli* types, K12 and O157:H7, are 99% identical. To date, we have identified 7 *I-CeuI* restriction fragment length polymorphism (RFLP) groups in *Ecc*. In contrast, the reported *I-CeuI* RFLP patterns of *Salmonella* spp. from 8 subgenera are nearly identical. Overall, our data suggests that there is considerable genetic variation in *Ecc* strains compared to related human pathogens.

CMVdelta2b can interfere with the accumulation of wild type CMV in zucchini squash plants in a cross-protection like phenomenon that involves RNA silencing. M. N. YASSI and M. J. Roossinck. Plant Biology Division, The Samuel Roberts Noble Foundation, Ardmore, OK 73402. Phytopathology 92:S89. Publication no. P-2002-0644-AMA.

Zucchini squash plants at the cotyledon stage were inoculated with inoculation buffer alone, or with wild type (wt) CMV or CMVdelta2b, a CMV mutant that does not express the 2b protein. Five days after this initial inoculation, symptoms characteristic of wt CMV and CMVdelta2b were observed in the first true leaves of infected plants. When the second true leaf of the plants previously inoculated with CMVdelta2b (non-symptomatic) were further inoculated with wt CMV, CMV symptoms were restricted to the inoculated leaves of these plants and wt CMV was not detected in upper leaves. Symptoms of wt CMV were observed in both inoculated and upper leaves of the plants that were initially inoculated with buffer alone, indicating that they were infected with CMV. Small RNAs (23-25-mer) were detected in both CMV and CMVdelta2b infected plants at 15 days post inoculation indicating that even with wt, CMV undergoes some silencing during infection. It seems likely that the presence of the 2b protein in wt CMV prevents the virus from degrading to non-detectable levels in the upper leaves. These observations also indicated that the CMV 2b protein could not reverse the RNA silencing already initiated in infection with CMVdelta2b even if wt CMV was inoculated to a leaf that did not show the virus symptoms.

Fungi associated with soil suppressiveness against *Heterodera schachtii* detected by oligonucleotide fingerprinting of ribosomal RNA genes. B. YIN, L. Valinsky, J. O. Becker, and J. Borneman. University of California, Riverside. Phytopathology 92:S89. Publication no. P-2002-0645-AMA.

The objective of this work was to identify fungi associated with suppressiveness against the plant-parasitic nematode, *Heterodera schachtii*. The experimental strategy was to identify fungi from *H. schachtii* cysts, which were isolated from soils possessing varying levels of suppressiveness. Since cysts isolated from suppressive soils can transfer this beneficial property to non-suppressive soils, analysis of the microorganisms within or on the cysts should lead to the identification of the target organisms. Five soils possessing varying levels of suppressiveness were inoculated with *H. schachtii* juveniles four weeks after seeding with mustard greens. Cysts were collected eleven weeks after inoculation. The fungi were identified through a ribosomal RNA gene (rDNA) analysis. Cysts obtained from soil mixtures consisting of 10% and 100% suppressive soil were inhabited by fungi with high sequence identities to *Cephalophora muscicola* and *Termitomyces* sp. Fungi from mixtures comprised of 0.1% and 1% suppressive soil had high sequence identity to *Fusarium oxysporum*.

***Rhizopus oryzae* produces macerating enzymes in infected mulberry roots.** S. YOSHIDA (1), S. Tsuyumu (2), T. Tsukiboshi (1), H. Shinohara (1), and S. Tsushima (1). (1) Inventory Center for Natural Resources, National Institute for Agro-Environmental Sciences, Tsukuba 305-8604, Japan; (2) Faculty of Agriculture, Shizuoka University, Shizuoka 422-8529, Japan. Phytopathology 92:S89. Publication no. P-2002-0646-AMA.

Rhizopus oryzae causes rot symptom on the roots of mulberry-grafted saplings. From the observation of the symptom, this fungus was thought to secrete the macerating enzymes in the infected tissues that lead to the collapse of the cortical tissues of stocks. Crude enzyme solutions were prepared from roots of the mulberry infected with the fungus and from the culture filtrate of the fungus grown in mulberry-roots liquid medium. When healthy root segments were used as the substrates to measure the maceration activity in these solutions at 30 degrees for 24 h, both solutions macerated the segments and its pH optimum was approximately 5.0. The cup plate assay revealed that they contained cellulase, polygalacturonase, pectate lyase and pectin lyase, but not protease. These results suggest that the rot symptom caused by *R. oryzae* may be the result of the maceration caused by these enzymes.

Effects of zoxamide on sporangia and zoospores of *Phytophthora capsici*. D. H. YOUNG and U. Vjugina. Dow AgroSciences, 9330 Zionsville Rd., Indianapolis, IN 46268. Phytopathology 92:S89. Publication no. P-2002-0647-AMA.

Zoxamide is highly effective in controlling potato tuber blight caused by *Phytophthora infestans*. To explore the underlying mechanism, we have studied effects of zoxamide on sporangia and zoospores in vitro using *P. capsici* as a model system. No direct effect on motility of swimming zoospores was found, and release of zoospores from healthy sporangia was only partially inhibited at a high zoxamide concentration (10 ppm). A "sandwich" plate technique was developed which enabled the production of sporangia in the presence of zoxamide. Such sporangia, produced with zoxamide at 0.4 - 10 ppm, contained nuclei of abnormal shape, size and distribution, and were severely compromised in their ability to release motile zoospores. Production of sporangia was also reduced at zoxamide concentrations of 2 - 10 ppm. We hypothesize that exposure of *P. infestans* to zoxamide during emergence of sporangia from infected leaves may result in tuber blight control by reducing the production of sporangia and motile zoospores.

BAS 516 F: A new broad-spectrum premix fungicide for use on fruits, vegetables, and treenuts. H. L. YPEMA (1), T. R. Bardinelli (1), J. S. Barnes (1), W. Fletcher (1), and W. Barton (2). (1) BASF Corp. Research Triangle Park, NC 27709; (2) BASF, London, Ontario N6N 1K8, Canada. Phytopathology 92:S89. Publication no. P-2002-0648-AMA.

BAS 516 F is a new broad-spectrum premix fungicide being developed by BASF. It contains the active ingredients pyraclostrobin (F500) and nicobifen (BAS 510 F). This broad-spectrum fungicide effectively controls the majority of foliar pathogens on small berries, bulb vegetables, carrots, grapes, onions, treenuts, stonefruits and strawberries. BAS 516 F has shown excellent activity on berry diseases, such as those caused by *Botrytis* sp. and *Monilinia* spp., anthracnose, and powdery mildew. Carrot pathogens controlled include powdery mildew, *Cercospora* leaf spot and *Alternaria* blight. On grapes, it is effective against powdery mildew, downy mildew, black rot, Phomopsis, and *Botrytis* bunch rot. On stonefruits and treenuts, BAS 516 F was shown to provide excellent control of blossom blight, shothole, scab, anthracnose, and brown rot. BAS 516 F's broad-spectrum efficacy and combination of two modes of action provide for an excellent foundation for disease control programs and management of fungicide resistance.

In vitro evaluation of the efficacy of Serenade® and other new biopesticide candidates in controlling grape canker disease caused by *Eutypa lata*. C. YUAN and S. O'Brien. AgraQuest, Inc., Davis, CA 95616. Phytopathology 92:S89. Publication no. P-2002-0649-AMA.

One of the most common canker diseases of grapevines in California is *Eutypa* caused by the fungal pathogen, *Eutypa lata*. The spread of this disease requires wind driven rain to disperse the ascospores to fresh pruning wounds in the grapevines where infection takes place. Without remedial action after infection, grapevine production declines steadily over a period of years, ultimately leading to its death. The preventive microbial fungicide, Serenade® (*Bacillus subtilis* strain QST713) and several biopesticide candidates developed by AgraQuest Inc., were evaluated for their efficacy against this disease under laboratory conditions. Experiments were conducted with two types of in vitro assays, a standard agar diffusion (SAD) and an assay with grape stems. Benlate was used as the chemical standard and a water treatment served as the positive control. Results of the SAD assay indicated that Serenade whole broth (WB) at 1/4 x, QST 713 technical powder (TP) at 3%, and 1/4 x concentrations of biopesticide candidates AQ 12365, AQ 4686 and AQ 4800 all resulted in significant inhibition of mycelia growth after 2 weeks of incubation. Assay with grape stems in which a piece of autoclaved grape stem was placed over a Bar-Lok® Cable Tie loop with the fungal inoculum placed nearby in a petri dish, indicated that all tested materials showed significant mycelia growth inhibition compared with those treated with water. Serenade WB and QST 713 TP had the same efficacy as Benlate in inhibition to mycelia growth with no mycelia growth on stems after 5 weeks of incubation. In contrast, the water-only treated stems were fully covered with mycelia. Further trials are needed to confirm efficacy under field conditions.

A survey of *Phytophthora* species in Hainan Province of China. H. C. Zeng (1), F. C. Zheng (1), and H. H. HO (2). (1) Plant Protection Research Institute, Chinese Academy of Tropical Sciences, Danzhou City, Hainan 571737, China; (2) Dept. Biology, State University of New York, New Paltz, NY 12561. Phytopathology 92:S89. Publication no. P-2002-0650-AMA.

A comprehensive study was initiated to isolate *Phytophthora* species from diseased plant materials, soil and water samples from Hainan Island of South China. To date, twelve *Phytophthora* species have been recovered: *P. capsici*, *P. cinnamomi*, *P. citrophthora*, *P. colocasiae*, *P. cyperi*, *P. cryptogea*, *P. drechsleri*, *P. heveae*, *P. insolita*, *P. meaditii*, *P. nicotianae* (*parasitica*) and *P. palmivora* with *P. nicotianae* (*parasitica*) as the most dominant plant pathogenic species. Of special interest is the discovery of *P. cyperi* causing leaf spot disease of *Digitaria* sp., a new host, the first record of *P. cryptogea* causing root rot disease of *Gerbera jamesonii* on the island, and the common occurrence of *P. insolita* in forest soil, streams and ponds causing no apparent plant diseases.

Structure-function analysis of the *Arabidopsis* disease resistance gene *RPS4*. X. Zhang and W. GASSMANN. Dept. of Plant Microbiology & Pathology, Univ. of Missouri-Columbia, Columbia, MO 65211. Phytopathology 92:S90. Publication no. P-2002-0651-AMA.

The *Arabidopsis* TIR-NBS-LRR gene *RPS4* confers specific resistance to *Pseudomonas syringae* pv. tomato expressing *avrRps4*. The non-functional *rps4* allele in susceptible ecotype RLD encodes a protein possessing two unique amino acid (aa) changes as compared to other functional alleles. We constructed chimeric *RPS4-Ler* genes with these aa changes. Results with stable transgenic RLD lines suggest that the N195D affects *RPS4* function more strongly than the Y950H, with the double-exchange showing an additive effect. A common feature of TIR-NBS-LRR genes is the presence of alternative transcripts. We removed the 2 introns that are frequently not spliced out and generated stable RLD transformants with these constructs. The truncated *RPS4* proteins are shown to be required for resistance. Further, susceptible transgenic RLD lines containing only full-length *RPS4-Ler* protein are not complemented by a second transgene encoding truncated *RPS4-Ler* protein. We hypothesize that dynamic regulation of the ratio of full-length to truncated *RPS4* proteins during the infection course is critical to this signaling pathway.

Characterization of the spread of two aster yellows phytoplasma strains in lettuce. J. ZHANG (1), S. Hogenhout (2), and S. A. Miller (1). (1) Dept. Plant Pathology; (2) Dept. Entomology, The Ohio State University, OARDC, 1680 Madison Ave. Wooster, OH 44691. Phytopathology 92:S90. Publication no. P-2002-0652-AMA.

To gain a better understanding of aster yellows phytoplasma (AYP) spread in plants after inoculation by leafhoppers, we used a polymerase chain reaction (PCR) assay to map distribution of two strains of the pathogen within *Lactuca sativa* plants. The phytoplasmas were first detected in the midrib of inoculated leaves one week after exposure to inoculative leafhoppers, then in the stem and growing points. AYP was not detected in the margins of inoculated leaves, suggesting that AYP spread in the leaves was unidirectional. Lateral translocation was observed in newly unfolded leaves one week before symptoms appeared. The AYP *severe* strain was detected earlier than the *bolt* strain in the unfolded leaves. AYP *severe* was detected in the mature leaves that had unfolded before exposure of plants to AYP-infected leafhoppers, whereas AYP *bolt* was not detected in those leaves, suggesting that AYP *severe* spreads more easily than AYP *bolt* in lettuce.

Analysis of temporal and spatial dynamics of mycosphaerella blight in field pea. J. X. ZHANG (1), W. G. D. Fernando (1), and A. G. Xue (2). (1) Department of Plant Science, University of Manitoba, Winnipeg, MB, R3T 2N2, Canada; (2) Agriculture and Agri-Food Canada, ECORC, Ottawa, ON K1A 0C6, Canada. Phytopathology 92:S90. Publication no. P-2002-0653-AMA.

The temporal and spatial dynamics of mycosphaerella blight of field peas, caused by *Mycosphaerella pinodes*, was investigated in 2000 and 2001. Inoculum source was in the centre 6 × 6 m area. Disease severity and spore concentration were monitored over time. Disease gradients were estimated based on directional disease severity values starting from the central disease focus. Spore concentration and disease severity decreased with increase in distance from inoculum source. The best model that describes disease progress varied with time, and distance from the inoculum source. At the early stage of disease development, the linear model described the temporal increase of disease better than the monomolecular, exponential, logistic or Gompertz models. However, the exponential model described disease progress best for both years during the late stage. The model best describing disease gradients varied in different directions.

Evaluation of Armicarb-100 for green mold control on Florida oranges. J. X. Zhang. Florida Dept. of Citrus, 700 Expt. Sta. Road, Lake Alfred, FL 33850. Phytopathology 92:S90. Publication no. P-2002-0654-AMA.

Armicarb-100 is a new EPA registered fungicide for the control of diseases on ornamental and other crops. Its active ingredient is potassium bicarbonate. Evaluation of this product for the control of citrus green mold caused by *Penicillium digitatum* was conducted using both inoculated and naturally infected different orange fruits and an aqueous packingline dripping system. In six trials Armicarb-100 significantly reduced green mold incidence, and demonstrated a similar or better control of the disease compared with standard fungicides imazalil and thiabendazole. The application rate at 4% product showed the best control compared with lower or higher concentrations. No fruit damage was observed under the test conditions. *In vitro* Armicarb-100 actively suppressed *P. digitatum* spore germination and its mycelial growth with an ED₅₀ of 0.06% and 0.08%, respectively. Since Armicarb-100 is a bicarbonate based product, its risk for consumers and environment should be minimal since its residues are not of any concern. It could be an alternative fungicide for citrus green mold control, and may be integrated into a postharvest chem-free fruit treatment and marketing system.

Genotypic typing of *Serratia marcescens* strains associated with cucurbit yellow vine disease. Q. ZHANG (1), R. Weyant (2), U. Melcher (1), B. Bruton (3), and J. Fletcher (1). (1) OSU, Stillwater, OK 74075; (2) CDC, Atlanta, GA 30333; (3) USDA-ARS, Lane, OK 74555. Phytopathology 92:S90. Publication no. P-2002-0655-AMA.

The bacterium that causes cucurbit yellow vine disease (CYVD) has been placed in the species *Serratia marcescens* (Sm) based on 16S rDNA and *GroE* sequence analysis. However, phenotypic comparison of the organism with Sm strains isolated from other niches showed disparity. In this study we compared the whole genomes of Sm strains from different niches through DNA-DNA hybridization and rep-PCR. With the former, relative binding ratio greater than 75 percent between the type strain of Sm and CYVD strain C01 confirmed the placement of C01 in species Sm. CYVD associated strains appeared as a closely related group in that they showed relative binding ratios greater than 95 percent with C01. This conclusion was further supported by homogeneous rep-PCR patterns among CYVD isolates, which suggested that CYVD strains might have a single common ancestor. However, since the rep-PCR patterns of CYVD isolates of Sm differ substantially from those of Sm from other niches, the origin of the plant pathogenic CYVD strains remains unclear.

QTL analysis for partial resistance to early blight in an F1 diploid hybrid potato population. R. ZHANG (1), B. J. Christ (1), and K. G. Haynes (2). (1) Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802; (2) USDA/ARS Vegetable Lab, Beltsville, MD 20705. Phytopathology 92:S90. Publication no. P-2002-0656-AMA.

Early blight, caused by *Alternaria solani*, is an important potato disease worldwide. Durable resistance is needed for disease control. A diploid out-crossing F1 population from *Solanum phureja* × *S. stenotomum* was evaluated for early blight resistance over three years using a randomized complete block design. Disease severities were assessed, and area under disease progress curve (AUDPC) values were calculated. The data were analyzed using PROC MIX in SAS®. There were significant differences among the 219 F1 clones for AUDPC. Based on the continuous distribution of AUDPC values, resistance to early blight appears to be inherited quantitatively, with several of the progeny being more resistant to early blight than either parent. Using Amplified Fragment Length Polymorphism (AFLP) markers, a linkage map was constructed for this F1 out-crossing population. So far, it contains 167 markers, covers 1,018 cM. Quantitative trait loci associated with early blight resistance will be mapped on the potato genome.

Mutagenic DNA repair in *Pseudomonas* spp. and its effect on ecological fitness. S. ZHANG and G. W. Sundin. Dept. of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843-2132. Phytopathology 92:S90. Publication no. P-2002-0657-AMA.

Twenty-nine strains of *Pseudomonas* spp. and three strains of *Burkholderia cepacia* were examined for UV radiation (UVR) sensitivity and UVR-induced mutagenic DNA repair (MDR). Strains of *P. cichorii*, *corrugata*, *fluorescens*, and *syringae* were UVR-mutable, and most were of plant origin. The MDR determinant was cloned from the highly UVR-mutable *P. cichorii* 302959, sequenced, and shown to be an allele of *ruAB* from *P. syringae*. A long-term single colony bottleneck experiment was conducted with 20 lineages of *P. syringae* pv. *syringae* B86-17 and GWS242 (B86-17 *ruB*:Km) with or without UVR exposure (80 total lineages/60 total cycles) to determine the effect of regular expression of MDR on ecological fitness and the accumulation of mutations. The maintenance of selected phenotypes was determined after every 10th cycle. Growth rate comparisons with ancestor

strains were done to assess reductions in fitness during the experiment. Mutations were only observed in UVR-exposed lineages, with similar numbers of mutations detected in B86-17 and GWS242 lineages.

A requirement for 2-methylcitrate synthase for Sterigmatocystin biosynthesis and sexual development in *Aspergillus nidulans*. Y. ZHANG (1), M. Brook (2), and N. P. Keller (1). (1) University of Wisconsin-Madison; (2) Philipps University. Phytopathology 92:S91. Publication no. P-2002-0658-AMA.

Sterigmatocystin (ST) is a carcinogenic polyketide produced by several *Aspergillus* species including *A. nidulans*. We have complemented a ST deficient mutant with the 2-methylcitrate synthase gene (*mcsA*). It catalyzes the conversion of propionyl CoA to 2-Methyl citrate, enabling the fungus to use propionate as a carbon source. Along with a decrease in ST production and loss of the ability to utilize propionate, the *mcsA* mutant strains are impaired in sexual development. A block at *mcsA* result in a build up of propionyl CoA, suggesting that the accumulating propionyl CoA in the mutant strains interferes with the function of certain ST biosynthetic enzyme(s). To examine this hypothesis, we disrupted the putative propionyl CoA synthase encoding gene *pcsA*, which is required for the conversion of propionate to propionyl CoA. The *pcsA* deletion strain still produces ST. Because it is likely that acetyl CoA synthase also produces some propionyl CoA, we will examine the phenotype of the double mutant, *delta pcsA/facA303* and the triple mutant *delta pcsA/facA303/delta mcsA* to help us determine the role of propionate and propionyl CoA in inhibition of ST biosynthesis.

Reaction of crucifer germplasm to bacterial pathogens. Y. F. Zhao, J. P. DAMICONE, M. E. Trent, and C. L. Bender. Department of Entomology and Plant Pathology, Oklahoma State University, Stillwater, OK 74078-3033. Phytopathology 92:S91. Publication no. P-2002-0659-AMA.

Selected accessions of *Brassica* spp. from the USDA germplasm collection were evaluated for their reaction to black rot (*Xanthomonas campestris* pv. *campestris*), Xanthomonas leaf spot (*X. campestris* pv. *armoraciae*), and bacterial leaf spot (*Pseudomonas syringae* pv. *maculicola*) by spray inoculation. Disease reaction was highly variable between plant species and pathogens. PI 418956 of *B. juncea* and PI 173861 of *B. nigra* were highly resistant to bacterial leaf spot. Several other accessions of *B. juncea* and *B. rapa* also showed resistance to bacterial leaf spot. PI 436606 of *B. oleracea* var. *capitata* was highly resistant to black rot as previously reported. However, the accessions PI 199947 and PI 199949 of *B. carinata*; and PI 173861, PI 367904, and PI 458981 of *B. nigra*; previously reported as resistant to black rot; were susceptible in this study. None of the accessions were resistant to Xanthomonas leaf spot. The results suggested that resistance to the different pathogens may be controlled by distinct host genes.

Gene content and organization of an 85-kbp DNA segment from the genome of *Spiroplasma kunkelii*. Y. ZHAO, R. E. Davis, I.-M. Lee, M. Martini, and R. Jomantiene. USDA-ARS Molecular Plant Pathology Laboratory, Beltsville, MD 20705. Phytopathology 92:S91. Publication no. P-2002-0660-AMA.

Spiroplasma kunkelii is a cell wall-less phloem-inhabiting bacterium that causes corn stunt disease. As a part of the *S. kunkelii* Genome Sequencing Project, we analyzed an 85 kbp DNA segment from the pathogenic *S. kunkelii* strain CR2-3x. This genome segment contains 101 open reading frames (ORFs). A majority of the ORFs code for predicted proteins that can be assigned to respective clusters of orthologous groups (COG). Their suggested functions cover diverse functional categories including genetic information storage and processing, cellular processes, and metabolism. The most notable gene cluster found in this genome segment is a super-operon capable of encoding 30 proteins, of which 26 are ribosomal proteins. The organization of the genes in this operon reflects the unique evolutionary position of the spiroplasma. A region containing gene duplications, domain rearrangements, and frameshift mutations, which are indicators of phase variation, was also identified in the genome segment. In addition to the protein-encoding capabilities, this genome segment includes two tRNA genes as well.

Chromosome length polymorphism in *Cochliobolus sativus*. S. ZHONG and B. J. Steffenson. Dept. Plant Pathology, University of Minnesota, St. Paul, MN 55108. Phytopathology 92:S91. Publication no. P-2002-0661-AMA.

Using pulsed field gel electrophoresis (PFGE), we analyzed karyotypes of 16 isolates of *Cochliobolus sativus* (spot blotch pathogen of wheat and barley),

collected from North Dakota, Virginia, Canada, Japan, Brazil, Uruguay, and Poland. The number and size of chromosome bands resolved ranged from 8 to 13 and from 0.85 to 3.80 mega-bases (Mb), respectively, using two different running conditions. Each isolate exhibited a unique banding pattern, except for two from North Dakota (ND90Pr and ND91-Bowman), whose banding pattern was very similar. Hybridization of single-copy DNA probes, previously assigned to 15 chromosomes of isolates ND93-1 and ND90Pr, to Southern blots of PFGE-separated chromosomes revealed highly polymorphic chromosomes among isolates. The observed chromosome length polymorphisms are likely due to translocations, duplications and deletions. Based on PFGE and Southern hybridization analysis, 14 to 15 chromosomes were estimated in these isolates.

Differential regulation of chitinases and glucanases mediated by a CLP regulator in *Lyobacter enzymogenes*. J.-M. ZHOU (1), R. M. Reedy (1), G. Yuen (2), and D. Y. Kobayashi (1). (1) Dept. Plant Biology and Pathology, Rutgers University, New Brunswick, NJ 08901; (2) Dept. Plant Pathology, University of Nebraska, Lincoln, NE 68583. Phytopathology 92:S91. Publication no. P-2002-0662-AMA.

Lyobacter enzymogenes strain C3 produces several extracellular enzymes, including chitinases and beta-1,3-glucanases, that are thought to contribute to antifungal and biocontrol activities. Previous studies indicate that extracellular enzyme activities are globally regulated by a catabolite activator protein-like protein (CLP). Chitinase activity is subjected to catabolite repression by glucose, which can be reversed by the addition of cAMP. Conversely, glucanase activity is induced by both laminarin and glucose. Furthermore, *lacZ* reporter gene assays indicate *clp* is expressed at high levels in both rich and minimal salts media containing various carbon supplements. Expression of *clp* is induced at significantly higher levels with laminarin compared to all other carbon supplements, including chitin. Interestingly, chitinase activity is repressed in the presence of laminarin. These results suggest a dual mechanism for CLP regulation of enzyme activities.

Hairy vetch soil amendment: A new potential alternative for suppression of Fusarium wilt in watermelon. X. G. ZHOU (1) and K. L. Everts (1,2). (1) University of Maryland, Salisbury, 21801; (2) University of Delaware, Georgetown, 19947. Phytopathology 92:S91. Publication no. P-2002-0663-AMA.

Alternative management strategies for Fusarium wilt in watermelon [*Fusarium oxysporum* f. sp. *niveum* (FON)] are needed because methyl bromide use will be discontinued and other available methods are frequently ineffective. Crop residues, composts, aquacultural wastes and chemicals were incorporated into a loamy sand soil naturally or artificially infested with FON race 2 in a greenhouse to evaluate wilt suppression in the susceptible cultivar Sugar Baby. Hairy vetch, urea and crab shell provided 53 to 87% wilt reduction. Soybean, corn, and wheat residues, composted poultry litter, and pine sawdust were not suppressive. Hairy vetch added into field microplots in a repeated test resulted in an average 60% wilt reduction and 109% increase in plant weight. Hairy vetch, unlike urea and crab shell, did not dramatically affect FON levels in soil, chlamyospore germination or mycelia growth on agar plates in film-covered microplots. This is the first evidence that a soil amended with hairy vetch may induce Fusarium wilt suppressiveness and is a potential alternative for Fusarium wilt management.

Early Botrytis infections as sources of secondary inoculum for ripening grape berries. S. M. ZITTER and W. Wilcox. Department of Plant Pathology, Cornell University. Phytopathology 92:S91. Publication no. P-2002-0664-AMA.

Loose and tight clusters of Pinot noir clones were inoculated with *B. cinerea* at four stages from bloom through veraison (Ve). After each inoculation, the incidence of latent infections was determined, as was the incidence of diseased berries with active Botrytis bunch rot (BBR) at and after harvest. There was no relationship between the incidences of latent and active infections, which were highest following the inoculations at bloom and Ve respectively. Cluster tightness had no influence on the incidence of latent infections but dramatically influenced final BBR incidences (41 versus 6 and 16% following Ve inoculations of the tight versus two loose treatments, respectively, 4-15 versus 0-1%, respectively for all other inoculations). When Chardonnay berries were inoculated 3 wk preharvest, final BBR was proportional to the initial number of berries that were inoculated/cluster ($R^2 = 0.81$), i.e., 1, 12, 24, and 32% diseased berries following inoculation of 0, 1, 3, and 5 berries/cluster. Early latent infections may be most important as potential sources of secondary inoculum as berries become highly susceptible to BBR after Ve.

Identification of novel *hrp*-regulated genes through functional genomic analysis of the *Pseudomonas syringae* pv. *tomato* DC3000 genome. J. ZWIESLER-VOLLICK (1), A. E. Plovanich-Jones (1), K. Nomura (1), S. Bandyopadhyay (1), V. Joardar (2), B. N. Kunkel (2), and S. Y. He (1). (1) Dept. of Energy Plant Research Laboratory, Michigan State University, East Lansing, MI 48824, USA; (2) Dept. of Biology, Washington University, St. Louis, MO 63130, USA. *Phytopathology* 92:S92. Publication no. P-2002-0665-AMA.

Pseudomonas syringae pv. *tomato* (*Pst*) strain DC3000 depends upon the type III protein secretion system to infect *Arabidopsis thaliana*. This system

is thought to deliver multiple effector proteins to the plant cell, promoting pathogenesis. All known type III effectors of *P. syringae* are coordinately regulated and contain a conserved *cis* element (called the 'hrp box') in their promoters. In this study, we designed a search motif based on known *Pst* DC3000 'hrp box' sequences. Seventy-three predicted genes were retrieved from the *Pst* DC3000 genome sequence. Expression of the 73 genes was analyzed by microarray and northern blot, revealing 24 genes/operons (including 8 novel genes) whose expression was consistently *hrp*-regulated. An AvrRpt2-based type III translocation assay provides evidence that at least 4 of the novel *hrp*-regulated genes encode putative effector proteins.

Errata

Vol. 92, No. 6 (Suppl.), 2002

The following abstracts were submitted for presentation at the 2002 American Phytopathological Society Annual Meeting.

Characterization of a tobamovirus from hibiscus. S. ADKINS (1), D. Achor (2), and D. J. Lewandowski (2). (1) USDA-ARS-USHRL, Fort Pierce, FL 34945; (2) Univ. of Florida, CREC, Lake Alfred, FL 33850. Publication no. P-2002-0666-AMA.

An unknown rod-shaped virus was isolated from landscape plantings of hibiscus (*Hibiscus rosa-sinensis*) in Florida. Viral-associated double-stranded RNA profiles and electron microscopy were consistent with it being a tobamovirus. Cloning and sequencing of the viral genome indicate that it is most likely a novel tobamovirus. The deduced amino acid sequence from a contiguous approximately 1300 nucleotide sequence was 45-50% identical to the helicase domain of the replicase proteins of recognized tobamovirus species. The deduced coat protein was 35-46% identical at the amino acid level with recognized tobamovirus species, and was most similar to *Tobacco mosaic virus* and *Sunn-hemp mosaic virus*. Sequences within the movement protein, methyltransferase domain of the replicase and the 3' untranslated region also had similarity to known tobamoviruses. An experimental host range comprised of 33 species in eleven families was tested. Most systemic hosts were in the Malvaceae.

Ribotyping as a means to delineate groups within the biocontrol bacterium *Bacillus mojavensis*. C. W. BACON and D. M. Hinton. USDA, ARS, Toxicology and Mycotoxin Research Unit, Athens, GA 30613. Publication no. P-2002-0667-AMA.

Bacillus mojavensis is a recently described bacterium in the *B. subtilis* group that is isolated from desert soils. We discovered that strains of this species are endophytic and offer potentials as biocontrol agents. However, this species is difficult to rapidly distinguish from the closely related bacillus species using various phenotypic and molecular methods. A robust tool is needed to rapidly and correctly identify this species, and if possible the subtypes. Ribotyping is a rapid, and convenient molecular typing method that separates species and subtypes. Automated ribotyping is based on restriction digestion of bacterial chromosomal DNA, followed by Southern hybridization with a ribosomal operon probe. Results are presented that ribotyping distinguished all isolates of *B. mojavensis* and established subgroups (ribogroups). Results are presented based on correlations between the ribogroups, the degree and type of fungal inhibition, and plant responses with the ultimate aim of facilitating grouping of this biocontrol bacterium.

Bleeding cankers caused by *Phytophthora* spp. on European beech. G. W. HUDLER, H. M. Griffiths, C. D Smart, and S. Jensen-Tracy. Dept. Plant Pathology, Cornell University, Ithaca, NY 14853. Publication no. P-2002-0668-AMA.

For at least 30 years, arborists in the northeast U.S. have tried to preserve mature (e.g. 80-150 yr old) European beech trees that begin to decline and quickly die for no apparent reason. In 1999, at several sites on Long Island, New York we began to record the fate of trees with bleeding cankers on the trunks but with no other symptoms of ill health to learn whether the cankers might be precursors to eventual death. We also used Neogen Alert™ kits to test necrotic tissue from these and other cankers found later for *Phytophthora* spp., and 10 of 11 cases tested positive. In subsequent years we have corroborated those results with cultures of *Phytophthora* spp. from all symptomatic bark. PCR products were obtained for the ITS sequences of genomic rDNA from the cultures, and the sequences were compared to those in the BLAST database. Results indicate that the isolates are equally closely related but not identical to *P. inflata* and *P. citricola*. Of six trees we first observed in 1999, all are dead.

Molecular control of the rice blast disease. Y. JIA. USDA-ARS Dale Bumpers National Rice Research Center, Stuttgart, Arkansas. Publication no. P-2002-0669-AMA.

Rice blast disease caused by *Magnaporthe grisea* is a major constraint to rice production worldwide. The rice blast system is one of the best-characterized monocot model systems. The goal of this project is to understand molecular mechanisms of disease resistance using rice blast as a model system. A combination of genetic, biochemical and pathological approaches to understand the host defense response was undertaken. Current efforts on development of the dominant marker for the rice blast resistance *Pi-ta* gene and its utilization for new cultivar development will be presented.

The relationship of the rice blast resistance genes *Pi-ta* and *Pi-ta²*. Y. Jia (1), Z. WANG (1,2) and R. Fjellstrom (3). (1) USDA-ARS DB NRRC, Stuttgart, AR 72160; (2) INAS, Zhejiang University, Hangzhou, P.R. China 310029; (3) USDA-ARS Rice Research Unit, Beaumont, TX 77713. Publication no. P-2002-0670-AMA.

Rice blast, caused by *Magnaporthe grisea*, is a serious disease worldwide. The rice *Pi-ta* gene is effective against *M. grisea* strains in a gene-for-gene manner and has been cloned. *Pi-ta* gene dominant markers were developed to determine whether the *Pi-ta²* blast resistance gene is at the *Pi-ta* locus. Historically, IB-49 resistance is conferred by the *Pi-ta²* gene in the cultivar Kaybonnet. A ratio of 3 resistant (R):1 susceptible (S) was observed in an F₂ population of 349 individuals of Kaybonnet and Maybelle using blast race IB-49 that is presumed to contain *AVR-Pita²*. The *Pi-ta* gene was only identified in resistant individuals. A ratio of 3 R:1 S was also observed in another F₂ population of 280 individuals of Kaybonnet and M-204. Again, only resistant individuals contain *Pi-ta*. We suggest that *Pi-ta* and *Pi-ta²* may be the same gene.

Protection from tomato late blight conferred through prosystemin-antimicrobial-peptide fusions. R. W. Jones (1), M. Ospina-Giraldo (1), and T. Clemente (2). (1) USDA-ARS, Vegetable Laboratory, Beltsville, MD 20705; (2) Dept. of Agronomy, University of Nebraska, Lincoln, NE 68588. Publication no. P-2002-0671-AMA.

The late blight pathogen *Phytophthora infestans* continues to elude current control strategies. The highly variable pathogen has developed many new races that limit the deployment of resistant varieties of tomato and potato. For non-race-specific resistance to late blight, we have developed a unique method for delivering very small antimicrobial peptides. The antimicrobial peptide is placed at the carboxyl end of a larger protein, and later released by proteolysis, a mechanism similar to that occurring in nature. In order to use this approach, the potato prosystemin (potpro2) transcript was cloned and the systemin-encoding region replaced with sequence encoding an eleven amino acid antimicrobial peptide (pep11). Tomato (Rutgers) was transformed with the binary vector pBI121 carrying the potpro2-pep11 construct and regenerates were screened by detached leaf assay. Six lines showed lesion expansion rates less than 20% that of wild type. Significant levels of powdery mildew resistance were also observed in greenhouse grown plants. Resistance was mediated through peptide induced growth reduction and apparent host response.

Redox regulation of *Monilinia fructicola* cutinase expression and germ tube development. M. H. LEE, S. M. Marek, and R. M. Bostock. Dept. Plant Pathology, University of California, Davis, CA 95616. Publication no. P-2002-0672-AMA.

Monilinia fructicola causes brown rot blossom blight and fruit rot in stone fruits. Immature fruit are highly resistant to brown rot but can become infected. These infections typically remain superficial and quiescent until they become active upon maturation of the fruit. High levels of chlorogenic acid (CGA) and related compounds occur in the peel of immature fruit but these levels decline during ripening. CGA inhibits cutinase expression, a putative virulence factor, with little or no effect on spore germination or hyphal growth. To better understand the regulation of cutinase expression by

fruit phenolics, we examined the effect of CGA, caffeic acid (CA) and related compounds on the redox potential of the growth medium and intracellular glutathione (GSH) levels. The presence of CA in the medium initially lowered the electrochemical redox potential of the medium, increased GSH levels and inhibited cutinase expression. Conidia germinated in the presence of CA, CGA, or GSH produced fewer appressoria and had elongated germ tubes compared to the controls. Host redox compounds seem to regulate fungal infectivity.

Agrobacterium tumefaciens-mediated transformation of *Monilinia fruticola* with green fluorescent protein. S. M. MAREK (1), Z. Pan (1), L. M. Ciuffetti (2), and R. M. Bostock (1). (1) Dept. Plant Pathology, University of California, Davis, CA 95616; (2) Dept. Botany & Plant Pathology, Oregon State University, Corvallis, OR 97331. Publication no. P-2002-0673-AMA.

Monilinia fruticola causes brown rot of many stone fruits. In order to genetically manipulate this fungus, a transformation system using *Agrobacterium* was developed. The binary transformation vector, pPTGFPH, was constructed from the plasmid pCT74, carrying green fluorescent protein (GFP) driven by the *ToxA* promoter of *Pyrenophora tritici-repentis* and hygromycin B phosphotransferase (*hph*) under control of the *trpC* promoter of from *Aspergillus nidulans*, and the binary vector pCB403.2, carrying neomycin phosphotransferase (*nptII*) between the T-DNA borders. Macroconidia of *M. fruticola* were co-incubated with *A. tumefaciens* strain LBA 4404(pPTGFPH) on media containing acetosyringone for two days. Hygromycin- and G418-resistant *M. fruticola* transformants were selected while inhibiting *A. tumefaciens* with cefotaxime. Transformants expressing GFP fluoresced brightly. Transformation efficiency, T-DNA integration frequency, and *in situ* studies on stone fruit tissues will be presented.

Septoria leaf spot intensity, defoliation, and yield loss relationships in southern blueberries. P. S. OJIAMBO, H. Scherm, and P. M. Brannen. Dept. of Plant Pathology, University of Georgia, Athens, GA 30602. Publication no. P-2002-0674-AMA.

Septoria leaf spot, caused by *Septoria albopunctata*, is prevalent on most southern highbush (*Vaccinium corymbosum* hybrids) and some rabbiteye (*V. ashei*) blueberry cultivars in Georgia. The disease has been observed to cause premature defoliation in the fall, but its quantitative effects on defoliation and return bloom have not been determined. In field plots of 'Premier' rabbiteye blueberry during fall of 2001, shoots with varying levels of leaf spot intensity were selected and assessed regularly for disease progress, defoliation, and flower bud set. A significant linear relationship ($R=0.787$; $P=0.001$; $n=50$) was observed between area under the leaf spot progress curve (AULSPC) and area under the defoliation progress curve (AUDFPC). A hyperbolic decay curve significantly described the relationship between AULSPC ($R=0.683$; $P=0.001$; $n=50$) or AUDFPC ($R=0.657$; $P=0.001$; $n=50$) and the number of flower buds formed on each shoot during the fall. Similar results were obtained in 'Bluecrisp' southern highbush blueberry. Further aspects related to disease assessment and multi-stage sampling size optimization are discussed.

A novel tomato cDNA with homology to lipid transfer proteins is rapidly induced by arachidonic acid and pathogen infection. Z. Pan (1), P. D. WEYMAN (2), Q. Feng (2), D. G. Gilchrist (1,2), and R. M. Bostock (1,2). (1) Ctr. for Engineering Plants for Resistance Against Pathogens, and (2)

Dept. Plant Pathology, University of California, Davis, CA 95616. Publication no. P-2002-0675-AMA.

A cDNA clone, *Da14*, isolated by mRNA differential display from arachidonic acid (AA)-treated leaves of tomato (*Lycopersicon esculentum*), encodes a protein homologous to a family of plant lipid transfer proteins. *Da14* transcript is expressed within 15 min of AA infiltration, is induced systemically, and accumulates in response to infection by *Phytophthora infestans* and to *Pseudomonas syringae* pv. *tomato*. No expression was observed in response to *Helicoverpa zea* larvae, AAL-toxin, or other elicitors. The *Da14* protein is targeted to the cell periphery, as observed by expression of a *Da14::GFP* fusion protein. Transgenic tomato plants overexpressing sense and antisense copies of *Da14* and vector-only transformed plants were constructed. The effect of the transgenes on the expression of genes regulated by salicylic and jasmonic acids and on disease resistance will be presented.

Molecular and pathological characterization of rice sheath blight pathogen isolates from Arkansas using rDNA-internal transcribed spacer sequences. P. SINGH (1,2), Y. Jia (1), R. Cartwright (2), F. N. Lee (2), and G. C. Eizenga (1). (1) USDA-ARS DB NRR, Stuttgart, AR 72160; (2) RREC, University of Arkansas, Stuttgart, AR 72160. Publication no. P-2002-0676-AMA.

Rice sheath blight, caused by *Rhizoctonia solani* Kühn (anastomosis group AG1-IA), is a serious disease worldwide. *R. solani* has a broad host range and no complete genetic resistance is available among cultivated rice. As first step to identify sheath blight resistance gene(s), molecular characterization of *R. solani* was performed to identify the most virulent isolate using DNA sequences of the ribosomal internal transcribed spacer (rDNA-ITS). Fourteen *R. solani* isolates were collected from 10 rice fields in Arkansas during the 2001 growing season. The rDNA-ITS region from these isolates was amplified by PCR, cloned, and sequenced. Resulting sequences were analyzed using Informax Vector NTI Suite V.6 and their phylogenetic relationship was established. To date, three genetically distinct classes of the pathogen were discovered. Progress in pathological characterization of these isolates will be presented.

PDR5-like transporters in the soybean pathogen *Phytophthora sojae*. M. S. Connolly, K. Allsion, A. Zhao, and P. F. Morris. Department of Biological Sciences, Bowling Green State University, Bowling Green, OH 43403. Publication no. P-2002-0677-AMA.

A family of proteins termed ATP Binding Cassette (ABC) proteins are found in all organisms and are responsible for the active transport of a wide variety of compounds including ions, phospholipids, sterols, organic acids, and xenobiotics. In plant pathogens, ABC transporters may contribute to biological fitness by controlling lipid asymmetry of membranes, providing protection against plant phytoalexins, or delivering toxins to host tissues. The primary focus of our interest is the Pleiotropic Drug Resistance family (*PDR5*) of ABC transporters in the soybean pathogen, *Phytophthora sojae*. *PsABC1*, the first member of this sub-cluster to be cloned from *P. sojae*, encodes a 1310 aa protein that is constitutively expressed at high levels by the free swimming zoospores stage and is the only *PDR5*-like transporter expressed by this stage. This transporter is not expressed by hyphae grown on V8 plates. Nor does it appear to be expressed during infection of soybean tissues. Progress in cloning related members of the *PDR5* family is described, and a strategy to assign function to individual members is described.

Erratum

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The following abstract replaces the abstract by Abad et al. (92[suppl.]:S1) submitted for presentation at the 2002 American Phytopathological Society Annual Meeting.

Advances in the integration of morphological and molecular characterization in *Phytophthora* genus: The case of *P. kelmania* and other putative new species. Z. G. ABAD (1,3), J. A. Abad (3), and T. Creswell (2,3). (1) Plant Pathogen ID. Lab.; (2) Plant Disease and Insect Clinic; (3) Dept. Plant Pathology, North Carolina State Univ. Phytopathology 92:S1. Publication no. P-2002-0001-AMA.

Phytophthora with over 70 spp is one of the most important Genera of plant pathogens. Considerable advances in the taxonomy of this stramenopile using morphological and molecular characterization have been accomplished. Yet, correct identification to species levels and characterization of new taxa is still a major challenge. Using our innovative morphological/molecular pictorial key new hosts and 3 new species have been determined including: *P. glovera*, *P. bisheria* (semipapillated), and *P. kelmania* (non papillated, named to honor Dr. A. Kelman). Additional unnamed new species are presented, including: Ph2471, Ph3552, (papillated), Ph9-001, Ph2529, Ph5212 (non papillated). An updated phylogenetic tree (ITS1-2 rDNA) including the recently described *P. europaea*, *P. pistaciae*, *P. psychrophila*, *P. ramorum*, and *P. uliginosa* maintains the consistency of 2 main groups (papillated and non-papillated) and verifies the identity of our eight putative new *Phytophthora* species.