

# Joint Meeting of the APS Potomac and Northeastern Divisions

## Abstracts

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Alphabetized by first author's last name

EFFECTS OF TOMATO MOSAIC VIRUS INFECTION ON THE GROWTH AND DEVELOPMENT OF RED SPRUCE SEEDLINGS. G.D. Bachand and J.D. Castello, State Univ. of New York, College of Env. Sci. and Forestry, Syracuse, 13210.

Tomato mosaic virus (ToMV) has been transmitted from red spruce (*Picea rubens* Sarg.) on Whiteface Mt., NY to herbaceous hosts in the greenhouse, purified, and transmitted to red spruce seedlings (Jacobi *et al.* 1992. Plant Dis. 76:518-522, and Jacobi and Castello 1992. Can. J. For. Res. 22:919-924). Our hypothesis was that ToMV infection adversely impacts the growth of red spruce seedlings. The objective of this study was to assess the impact of virus infection on the growth and development of red spruce seedlings. Two-year old red spruce seedlings were root inoculated with ToMV in July 1992. Significant reductions ( $p < 0.01$ ) in mean shoot length of infected versus control seedlings were observed six weeks into the second growth period. At the end of that growth period (12 weeks), significant reductions in root volume ( $p < 0.004$ ), seedling height ( $p < 0.002$ ), and needle length ( $p < 0.002$ ) were observed between infected and control seedlings. These results indicate that ToMV adversely impacts the growth and development of red spruce seedlings.

cDNA CLONING OF THE COAT PROTEIN AND A PORTION OF THE 30K PROTEIN REGION OF TOMATO MOSAIC VIRUS. G.D. Bachand, J.D. Castello, and S.O. Rogers, State Univ. of New York, College of Env. Sci. & Forestry, Syracuse, 13210.

Our objective was to develop a cDNA probe for detection of tomato mosaic virus (ToMV) by slot blot assay and to compare the sensitivity and specificity of this method with ELISA for detection of the virus in red spruce (*Picea rubens* Sarg.). Two synthetic oligonucleotide primers, based on the sequence of the type strain of ToMV (ToMV-L) were used to amplify a 1.1 kb cDNA fragment by reverse transcription and polymerase chain reaction (RT-PCR). The amplified fragment represents the coat protein and a portion of the 30k protein of ToMV. This fragment was digested with restriction enzymes and cloned into *E. coli*. The resulting 800 bp clone was used to detect ToMV RNA in red spruce using slot blot assays.

EFFECT OF IPRODIONE-RESISTANT STRAINS OF BOTRYTIS CINEREA ON EFFICACY OF IPRODIONE FOR CONTROL OF BUNCH ROT OF GRAPE. A.B.A.M. Boudoin, Dept. of Plant Pathology, Physiology and Weed Science, Virginia Tech, Blacksburg, VA 24061-0331

Iprodione-resistant isolates of *B. cinerea* (EC50 for radial growth 1.0-1.8 mg/L in potato dextrose agar, compared to wild-type EC50 of 0.15-0.25 mg/L) were detected in several Virginia vineyards in 1992. In 1993, iprodione efficacy was compared in a vineyard with less than 20% of the population resistant and in a vineyard with more than 90% of the population resis-

tant. Iprodione (2 pints Rovral 4F in 60 gallons per acre) was applied at bloom, prior to cluster closing, at early berry ripening, and 3 weeks later. Treatments included 2- and 4-spray combinations. In the vineyard with low resistance frequency, Botrytis bunch rot incidence and severity were not affected by iprodione treatments. In the vineyard with high resistance frequency, the first two sprays reduced bunch rot by over 50% while the last two sprays had no effect, presumably due to a dry second half of the season.

DETERMINING THE PATHOGENS PRESENT IN PRE-HARVEST APPLE FRUIT. Alan R. Biggs, West Virginia University, University Experiment Farm, P.O. Box 609, Kearneysville, WV, 25430

Symptomless apple fruit at various stages of maturity were washed in tap water, dipped in 95% ETOH for 2 min, then in 0.5% NaOCl for 7 min. Fruit were cored and sliced with a sterile metal template, and fruit sections were dipped for 60 sec. in 6000 µg/ml paraquat (1,1'-dimethyl-4,4'-bipyridinium dichloride (Gramoxone Extra 16.0 ml/L)), then rinsed in sterile, distilled water for 1 min. and placed in autoclaved jars with filter paper lids. Fruit were incubated in the dark for 5 d, then moved into the light and observed for 3 wk for rate of breakdown and appearance of fungi. Genera observed included *Colletotrichum*, *Botryosphaeria*, *Phoma*, *Phyllosticta*, *Penicillium*, and *Alternaria*. Samples taken at 3 wk prior to harvest revealed latent *Botryosphaeria* infections (73%), as well as *Phoma* and *Phyllosticta* (17%). Control fruit exhibited no signs of fungi. For fruit at optimum maturity, 40% showed latent *Botryosphaeria* infection, and 37% showed infections by other fungi. For fruit at 3 weeks after optimum maturity, 53% yielded species of *Phoma* and *Phyllosticta*, 23% showed latent *Botryosphaeria* infections, and 10% yielded *Penicillium*.

GENETIC AND PHENOTYPIC VARIABILITY IN BIALAPHOS RESISTANT TRANSFORMANTS OF *COLLETOTRICHUM GLOEOSPORIOIDES* F.SP. *AESCHYNOMENE*. N.L. Brooker, J. Lydon, and C. F. Mischke. Weed Science Laboratory, United States Department of Agriculture, BARC-West, Beltsville, MD. 20705.

*Colletotrichum gloeosporioides* f.sp. *aeschynomene* (C.g.a.) is a commercially available mycoherbicide used to control northern jointvetch, a leguminous weed found in rice and soybean fields in the Southern United States. Bialaphos is a naturally produced herbicide which is made by *Streptomyces hygroscopicus* and commercially marketed as Herbicac (20% bialaphos active ingredient). Bialaphos acts as a glutamine synthetase inhibitor in plants, bacteria and fungi. Using the bialaphos resistance gene isolated from *S. hygroscopicus* and cloned into a fungal expression vector, C.g.a. was transformed with bialaphos resistance. Bialaphos resistant transformants are being used to evaluate the co-application strategy of using mycoherbicides in combination with natural herbicides to control weeds. Five bialaphos resistant transformants of C.g.a. were evaluated on several phenotypic cultural characteristics, plant pathogenicity and virulence. The results of these studies indicate that phenotypically, many of the transformants are deficient in several important wild-type characteristics such as sporulation, growth rate, pigmentation and virulence. Transformant #48-5b appears to be identical with the wild-type parent, but also displays bialaphos resistance and stable integration of the bialaphos resistance gene after plant reisolation. Use of this co-application strategy for enhanced mycoherbicide weed control will be discussed.

Camera-ready abstracts are published as they were submitted by the Division. The abstracts are not edited or typed in the APS headquarters office.

UTILIZATION OF THE POLYMERASE CHAIN REACTION TO IDENTIFY *MAGNAPORTHE POAE*. Tracy E. Bunting, Karen A. Plumley, Bruce B. Clarke, and Bradley I. Hillman. Department of Plant Pathology, Rutgers University, P.O. Box 231, Martin Hall, New Brunswick, NJ 08903

*Magnaporthe poae* Landschoot & Jackson is a heterothallic, ascomycetous fungus which causes summer patch, a devastating disease of cool season turf. Definitive identification of the disease has been impractical, due to the length of time needed for the production of the sexual phase. We have designed primers which specifically amplify a 450 nucleotide fragment from the DNA of all *M. poae* isolates tested. In order to clarify the phylogenetic relationship of *M. poae* to *M. grisea*, *M. salvinii*, *M. rhizophila*, *Gaeumannomyces graminis* var. *graminis*, *Glomerella* (ana. *Colletotrichum*) *graminicola*, *Cryphonectria parasitica*, and *Leptosphaeria korrae*, we are currently determining the sequence of the first intragenic spacer region of the genomic ribosomal DNA.

#### EFFECT OF HOT WATER TREATMENTS ON ERADICATION OF *AGROBACTERIUM VITIS* FROM DORMANT GRAPE CUTTINGS.

Thomas L. Burr and Cheryl L. Reid, Department of Plant Pathology, Cornell University, NYSAES, Geneva, NY 14456.

*A. vitis*, the cause of crown gall on grape, can survive systemically in symptomless grapevines and may be spread in propagation material. The effectiveness of hot water treatments in eradicating the pathogen from dormant cuttings was tested. *In vitro*, *A. vitis* is highly sensitive at 50C with an approximate D-value of 2 min. (the time required to reduce the population of bacteria by 90% or 1 log). Naturally-infected cuttings often carried low levels of pathogen making it difficult to interpret the effect of treatments. Potted vines were inoculated in Jun.-Jul. and woody trunks with galls were harvested in Oct. when they were dormant. Cuttings of different cultivars were submerged in water for 30-90 min at temperatures ranging from 50-55C. Isolations were made from plant tissues just beneath the galls. More than 95% of the cuttings were free of *A. vitis* following a treatment of 50C for 60 or 90 min. None of the treatments completely eradicated the pathogen. Treatments at temperatures greater than 50C resulted in bud injury. Therefore *A. vitis* cells surviving in dormant cuttings are more heat tolerant than those at a stationary phase *in vitro*.

INFLUENCE OF PREVIOUS EXPOSURE TO OZONE ON SUSCEPTIBILITY OF TOBACCO AND TOMATO TO *ALTERNARIA* LEAF SPOTS. Yong Cao and William J. Manning. Department of Plant Pathology, Univ. of Mass/Amherst, MA 01003-2420.

Greenhouse and field experiments were conducted in 1992 and 1993 to investigate the influence of ozone on *Alternaria* leaf spots of tobacco (*Nicotiana tabacum* L.) and tomato (*Lycopersicon esculentum* Mill). In greenhouse experiments, tobacco plants (cv. Bel-W3 and Bel-B) were more susceptible to the A5 strain of *A. alternata* (Fries) Keissl after exposure to ozone at a concentration of 50 ppb or higher for 3 days or longer. Older leaves were more susceptible than younger leaves. Tomato plants (cv. Ramapo and Better Boy) were more susceptible to *A. solani* Sorauer after exposure to ozone at 80 ppb for 6 days or longer. Similar results were obtained in field experiments in open-top chambers.

EVALUATION OF POTATO CULTIVARS FOR RESISTANCE TO *VERTICILLIUM DAHLIAE*. R.B. Carroll and D.P. Whittington. Department of Plant and Soil Sciences, University of Delaware, Newark, DE 19717-1303.

Seven cultivars of potatoes were screened for resistance to *Verticillium dahliae* Kleb. in two greenhouse and two field experiments during 1993. Alleghany, Portage, Langlade, Snowden, Conestoga and CF 7523-2 were compared with Superior which is the predominant cultivar grown in DE. Both greenhouse tests utilized *V. dahliae*-infested grain. In the field, soil was fumigated and seed-pieces were inoculated, at planting, with either infested grain or a spore suspension. Appropriate controls were included in all experiments which were replicated and randomized. Greenhouse and field results were consistent with Alleghany showing the greatest resistance while Conestoga and Superior were most susceptible. There was no significant difference in disease incidence between the two inoculation methods.

CALCIUM OXALATE CRYSTAL PRODUCTION BY THREE BROWN ROT FUNGI. J.H. Connolly and J. Jellison, Dept. of Plant Biology and Pathology, University of Maine, Orono, ME 04469-5722.

Oxalic acid production is widespread among the fungi and is often coincident with calcium oxalate crystal production. *Gloeophyllum trabeum* is a brown rot organism that was found to produce prototypical bipyramidal calcium oxalate dihydrate crystals in certain culture conditions, as well as in the wood environment. *Postia placenta* produced calcium oxalate crystals of a different habit in the wood environment from that which was found in malt agar culture. A putative *Fomitopsis* sp. isolate produced bipyramidal calcium oxalate dihydrate crystals in the wood environment and on malt, but produced prismatic crystals in certain batch culture conditions. Calcium is well known to stimulate calcium oxalate production in plants and fungi, but magnesium was also found to stimulate calcium oxalate production *in vitro*. No evidence for magnesium oxalate precipitation was found. Calcium oxalate crystals were observed in association with the putative hyphal sheath.

BIOLOGICAL CONTROL OF APPLE SCAB BY *PSEUDOMONAS SYRINGAE* STRAIN 508. M.R. Corral-Garcia, T.J. Burr, C.A. Smith and M. C. Matteson. Department of Plant Pathology, Cornell University, NYSAES, Geneva, NY 14456.

*Pseudomonas syringae* strain 508, isolated from an apple leaf in an abandoned orchard, significantly reduced severity of apple scab (*Venturia inaequalis*). Its activity was compared to several other bacterial strains from apple. Fifteen tests were done in which a suspension of each bacterial strain was sprayed on apple seedlings in the greenhouse. Strain 508 caused significant reduction of scab in all the tests, achieving the same level of control as the fungicide captan in 10 of them. Another five *P. syringae* strains from apple orchards showed variable results in the greenhouse, while 508 constantly controlled scab. In order to obtain this control, a suspension of 10<sup>9</sup> CFU/ml was required. The population of strain 508 was monitored for 4 days after being sprayed on leaves of apple seedlings. During the first 24 hours the number of bacteria on the leaves was quite constant, and similar on all leaves. After 24 hours, the number of bacteria progressively decreased, with increased variability among leaves. The persistence of the anti fungal activity of strain 508 on apple seedlings, and its post infection effect will be discussed.

EFFECTS OF TEMPERATURE ON GROWTH AND SURVIVAL OF *DISCULA DESTRUCTIVA* J. B. Crozier, and R. J. Stipes. Dept. Plant Pathol., Physiol., and Weed Sci., VA Tech, Blacksburg, VA 24061-0331.

Cardinal growth temperatures and thermal stress extremes were determined for isolates of *Discula destructiva*, causal agent of dogwood anthracnose. The optimum temperature was between 20.0 and 22.0 C, with 4 of 6 isolates growing best at 20.0 C. All isolates grew within 7 days at 1.0 C and 28.0 C, but no growth was noted after 7 days at 30.0 C; regrowth occurred after transfer to a lower temperature. All isolates were killed after 7 days at 35.0 C. The fungus was alive in 88% of 4-mm mycelium agar discs after 5 min. at 45 C in water, while only 51% were living after 10 min., and 0% after 15 min. The fungus was alive in 89% of 4-mm leaf discs from amended PDA plates (autoclaved dogwood leaf on which *D. destructiva* was allowed to grow) after 10 min. at 45 C. The thermal death point of conidia in water was 46-47 C, and the thermal death times for 45 and 55 C were 20 min. and 30 sec. respectively for conidia from oatmeal agar plates. Conidia from autoclaved dogwood leaves on amended PDA were killed within 5 min at 45 C. This information may lead to an understanding of possible climatic barriers, and the thermal treatment of plant material.

EFFECTS OF ACIDIC FOG ON LEAF SURFACE ANATOMY OF *CORNUS FLORIDA* AND *CORNUS KOUSA* SEEDLINGS J. B. Crozier, R. T. Thornham, and R. J. Stipes. Dept. Plant Pathol., Physiol., and Weed Sci., VA Tech, Blacksburg, VA 24061-0331.

Acidic precipitation reportedly enhances disease severity of dogwood anthracnose (DA) caused by *Discula destructiva* on *Cornus florida*, the flowering dogwood. Seedlings of *C. florida* and *C. kousa*, the Chinese dogwood, which is fairly resistant to dogwood anthracnose, were subjected to acidic fog episodes at pHs 2.5, 3.5, 4.5, and 5.5, using a simulated acidic rain solution. Leaf discs from these and non-treated plants were examined by scanning electron microscopy. Damage was noted at all pH levels and was primarily confined to the trichomes and stomata. Trichomes appeared dehydrated on both *C. florida* and *C. kousa* leaves, while the "lips" of *C. florida* stomata were increasingly eroded by decreasing pH; *Cornus kousa* stomata were relatively unharmed. At pH 2.5, trichomes of both species seemed to be brittle and fractured, causing deep holes in the lamina. *Discula destructiva* conidia may germinate at trichome bases where damage may cause leaching of nutrients. Also, the difference in stomatal damage may play a role in disease severity.

OCCURRENCE AND CONTROL OF POWDERY MILDEW ON POINSETTIAS IN U.S. GREENHOUSES. M. L. Daughtrey and M. T. Macksel. Dept. of Plant Pathology, Long Island Horticultural Research Laboratory, Cornell University, Riverhead, NY 11901

A new powdery mildew disease on poinsettia, *Euphorbia pulcherrima*, has been noted in commercial greenhouses in the U. S. since 1990. Although no cases were reported in 1991, in 1992 the disease occurred in many states, primarily in the northern U. S. In a New York survey, 30 growers reported mildew outbreaks during 1992. Mildew was observed as early as April, but was usually not detected until November. Mildew occurred on upper and lower leaf surfaces. Chlorotic spots appeared opposite mildew colonies on lower surfaces. Disease incidence and severity did not increase during the summer, but increased dramatically in the fall. Mildew continued to develop in florist's shops and purchaser's homes. Copper sulfate pentahydrate, insecticidal soap, oil + baking soda, an experimental bicarbonate formulation, thiophanate-methyl, triadimefon and triflumizole were effective at suppressing powdery mildew development on poinsettias in 1992-3 trials. A management program based on monitoring combined with prompt roguing and fungicide treatment is proposed.

DEVELOPMENT OF DIPLODIA EAR ROT IN MAIZE FOLLOWING INOCULATION WITH A *STENOCAPELLA MAYDIS* (*DIPLODIA MAYDIS*) BENOMYL RESISTANT MUTANT. A.E. Dorrance, H.L. Warren, and G.H. Lacy. Department of Plant Pathology, Physiology and Weed Science, Virginia Polytechnic Institute and State University, Blacksburg, VA 24061-0331

A spontaneous mutant of *Stenocarpella maydis* (Berk.) Sutton with resistance to benomyl was selected on PDA amended with 1 µg/ml of benomyl (99.18% a.i., in DMSO). Nine B73 maize inbreds (susceptible to diplodia ear rot) were inoculated (5 ml of 5,000 pycnidiospores/ml) in the whorl 10 d prior to tassel emergence. Three additional plants were inoculated with water. The mutant was reisolated after 6 wk from 7 of the 9 symptomatic plants. Segregation of benomyl resistance may occur because the fungus is heterokaryotic. Pycnidia formed in three inoculated plants. Other researchers have demonstrated that this fungus can directly penetrate leaves, but did not indicate if ear rot resulted. That the benomyl mutant penetrates via the whorl supports Durrell's hypothesis that this fungus penetrates at the shank base or the nodes. Colonization of host tissues is a slow process; observable symptoms may not appear until two weeks post anthesis. Inoculation methods which mimic this mode of entry may give the best results in testing maize germplasm for resistance to ear rot.

ISOLATION AND PATHOGENICITY OF *FUSARIUM* SPECIES ASSOCIATED WITH PUMPKIN FRUIT ROT. W. H. Elmer. CT Agr. Exp. Sta, Box 1106, New Haven, CT 06504.

Over 60% losses in 'Howden' pumpkins occurred in a grower's field in Connecticut due to fruit rot that was distinctly different from black rot. This disease was also found exclusively on 'Howden' pumpkins in several other areas in Connecticut. Three types of symptoms were noted. The most common symptom was a circular to oval lesion (0.5-2.0 cm diam.) with a depressed dry corky center (0.2-0.5 cm deep) that contained orange sporodochia of what was identified on carnation leaf agar (CLA) as *Fusarium acuminatum*. Another symptom was a white mycelium that would emerge from a lesion and colonize the rind of the fruit. Two fungi were isolated from these lesions which were identified as *F. graminearum* and *F. avenaceum*. A third symptom was a lesion which had soft rotting effect on the rind and was sometimes covered with white to grayish zonal rings filled with macroconidia of *F. equiseti*. When agar plugs colonized by these different *Fusaria* were inoculated into 'Howden' pumpkins in the greenhouse, these symptoms were observed and the same fungi were reisolated. On occasion *F. oxysporum*, *F. solani*, and *F. proliferatum* were isolated from symptomatic pumpkins, but these species did not cause symptoms in pathogenicity tests.

FUNGICIDAL ACTIVITY OF TAXANES FROM ORNAMENTAL YEWS W. H. Elmer, M. J. I. Matina, and G. J. MacEachern. The Connecticut Agricultural Experiment Station, Box 1106, New Haven, CT 06504.

Taxanes (diterpene alkaloids), including taxol and cephalomannine, from needles of several cultivars of ornamental yews were tested for activity in inhibiting soilborne fungi. Freshly ground needles from ornamental yews were agitated in methanol for 24 hr, loaded onto solid phase extraction cartridges and fractionally eluted. The 80% methanol/20% water fractions were dried to a solid residue on a roto-evaporator. The residue was reconstituted in 95% ethanol and quantified for taxol and cephalomannine by liquid chromatography with UV-diode array detection. The taxane residues were amended into molten potato dextrose agar (48-50 C) at rates that yielded taxol concentrations of 0.1, 0.5, 1.0, and 3.0 µg/ml. The radial growths of *Fusarium oxysporum*, *F. proliferatum*, *Rhizoctonia solani*, *Phytophthora cactorum*, and *Verticillium dahliae* were determined at 25 C and compared to ethanol controls. Inhibition was detected for all fungi except *V. dahliae* at 3.0 µg/ml. *P. cactorum* was the most sensitive to the taxane extracts and showed inhibition at 0.5 µg/ml. These data suggest that taxanes from yew needles may have potential as bio-fungicides.

VEGETATIVE COMPATIBILITY AMONG ISOLATES OF *FUSARIUM OXYSPORUM* F. SP. *BASILICUM* FROM BASIL SEEDS AND INFECTED PLANTS. W. H. Elmer<sup>1</sup>, R. L. Wick<sup>2</sup>, and P. Haviland<sup>2</sup>. <sup>1</sup>The Connecticut Agricultural Experiment Station, Box 1106, New Haven, CT 06504 and <sup>2</sup>Dept. of Plant Pathology, Univ. of Massachusetts, Amherst, MA 01003.

Over 35 isolates of *Fusarium oxysporum* f. sp. *basilicum* (FOB) were collected from diseased basil plants and seeds during 1991-1993 from California, Colorado, Connecticut, Massachusetts, New York, and South Carolina. An ATCC isolate of FOB (ATCC 38560) deposited by researchers from Italy in 1975 was also included. Nitrate non-utilizing mutants were selected from each isolate and used in heterokaryon tests to define vegetative compatibility groups (VCG). All isolates except one (NY63) were vegetatively compatible with the ATCC isolate. All isolates except NY63 were virulent on susceptible basil in the greenhouse. Since isolates of FOB may represent a unique VCG, heterokaryon tests could replace the lengthy pathogenicity tests and reduce the amount of time necessary to distinguish nonpathogenic isolates of *F. oxysporum* from isolates of FOB. The rapid spread of this disease in the U.S. is likely due to dissemination of infested seeds.

REDUCED PRODUCTION OF CLEISTOTHECIA BY *UNCINULA NECATOR* DUE TO THE MYCOPARASITE *AMPELOMYCES QUISQUALIS*. Stuart Falk and David Gadoury. Department of Plant Pathology, Cornell University, NYSAES, Geneva, NY 14456-0462.

Cleistothecia of *Uncinula necator* are susceptible to infection by *Ampelomyces quisqualis* (AQ) during their early development, but are immune to infection once matured. In field trials, grapevines were treated with AQ by suspending cotton twine colonized by the mycoparasite above vines to encourage early and more effective parasitism. In 1992, increased parasitism and reduced production of cleistothecia on leaves of treated vines was demonstrated early in the season. However, parasitism by AQ was high in all plots at the end of the season, possibly due to ideal conditions for natural parasitism created by frequent and heavy rainfall in 1992. Numbers of cleistothecia overwintering on bark were low on all vines, but parasitized cleistothecia could only be found on bark of AQ-treated vines. In 1993, treatment of vines with AQ increased the parasitism of cleistothecia throughout the season. Increased parasitism reduced the number of viable cleistothecia removed from leaves during the fall rains. Monitoring is under way to determine whether treatment with AQ will result in a reduction of the number of cleistothecia that survive winter on bark.

A RHIZOCTONIA BLIGHT SEVERITY MODEL FOR PERENNIAL RYEGRASS M.A. Fidanza and P.H. Dernoeden. Department of Agronomy, The University of Maryland at College Park, MD 20742.

A field study was initiated in 1991 to determine environmental conditions favorable for *Rhizoctonia* blight (*R. solani* Kuhn) in perennial ryegrass (*Lolium perenne*), and to develop a model to predict disease severity. Environmental conditions monitored were ambient air temperature, relative humidity (RH), precipitation, leaf wetness duration, soil temperature, soil moisture, and solar irradiance. A visual estimate of percent plot area covered with *R. solani* mycelium was used to determine moderate, high, and very high disease severity levels. A very high disease severity index required  $\geq 9$  hours of RH  $\geq 95\%$  with a minimum and average air temperature of  $\geq 22$  and  $\geq 24^\circ\text{C}$ , respectively. Rain ( $>8\text{mm}$ ) within 72 hr also was required for very high disease. Minimum and average air temperatures associated with high disease severity were  $\geq 21$  and  $\geq 24^\circ\text{C}$ , respectively, with  $\geq 6$  hours of RH  $\geq 95\%$  or  $\geq 6$  hours of RH  $\geq 90\%$  plus precipitation  $\geq 8$  mm. A moderate disease severity index was associated with minimum and average air temperatures of  $\geq 17$  and  $\geq 19^\circ\text{C}$ , respectively, with  $\geq 3$  hours of RH  $\geq 95\%$  or  $\geq 3$  hours of RH  $\geq 90\%$  plus precipitation  $\geq 8$  mm. Disease severity was predicted with  $\geq 83\%$  accuracy. In 1993, the model was field evaluated and a 30% reduction in fungicide use was achieved when compared to applying a fungicide on a 14-day schedule.

LEVELS OF VARIATION IN FOLIAR OZONE INJURY AMONG BLACK CHERRY TREES. T.S. Fredericksen, J.M. Skelly, K.B. Kouterick, B. J. Joyce, T.E. Kolb, K. C. Steiner, J. E. Savage, and K. R. Snyder. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Black cherry (*Prunus serotina* Ehrh.) is a valuable timber species which is highly sensitive to ozone. The response of black cherry trees to ozone is complex and understanding the sources of variation and the mechanisms of this response is essential for determining the impact of this air pollutant on black cherry in eastern forests. Studies were conducted in 1993 to investigate the extent of variation in foliar injury between and within different-sized black cherry trees in northcentral Pennsylvania. Foliar ozone injury, observed as a dark adaxial stipple, varied among tree size classes, individuals within a size class, and leaves located at different crown positions within an individual tree. Measurements of physiological and morphological traits revealed that the amount of foliar injury was significantly related to stomatal conductance, shoot phenology, and leaf thickness.



FACTORS ASSOCIATED WITH REGENERATION FAILURES IN BIGTOOTH ASPEN (*POPULUS GRANDIDENTATA* MICHX.) STANDS. T. Frontz and D. D. Davis. Department of Plant Pathology, The Pennsylvania State University, 210 Buckhout Laboratory, University Park, PA, 16802.

Selected forests in north-central Pennsylvania, which have a high basal area of bigtooth aspen, currently are being managed as bigtooth aspen stands by the use of clearcutting. Clearcutting stimulates aspen to reproduce as root suckers. However, following clearcutting, mortality of resulting aspen root suckers has been a limiting factor in maintaining adequate regeneration of this species. During the summer of 1993, two surveys were conducted to determine the overall incidence of aspen root sucker mortality within selected clearcuts of various ages; 55 permanent plots were established; 60 stump excavations were made; and roots of 142 pairs of adjacent live/dead suckers were excavated. In each of six residual stands, 15 bigtooth aspen trees were felled, and disks removed at breast height to identify stress periods using tree-ring analysis. Small disks for analysis were also removed from live and dead root suckers in the clearcuts. Results to date indicate that the mortality may be related to the interaction of gypsy moth defoliation and *Armillaria* root rot.

TRACKING SOYBEAN MOSAIC VIRUS MOVEMENT BY IMMUNOASSAY OF LEAF IMPRINTS. T. Gera and S. A. Tolin. Dept. of Plant Pathology, Physiology & Weed Science, Virginia Polytechnic Institute & State University, Blacksburg, VA 24061.

Soybean mosaic virus (SMV) induces mosaic symptoms on susceptible cultivars of soybean (*Glycine max*) and either no symptoms or local and/or systemic necrosis on resistant cultivars. Resistance is strain specific and mediated by a single host gene, but the mechanism is unknown. Our objective was to assess the roles of restriction of replication and movement, together with hypersensitivity, in the expression of resistance. We developed a method whereby the rate and extent of invasion could be tracked from an inoculation site on the tip of a primary, unifoliate leaf throughout that leaf. Whole leaves were placed on hardened filter paper, covered with plain paper, and imprinted with sharp hammer blows onto a covering acrylic sheet. Viral antigens were detected by immunoassays using SMV-specific primary antibody, alkaline phosphatase-labeled, goat anti-rabbit secondary antibody, and NBT/BCIP. In susceptible responses, SMV was at the site of inoculation in 6-7 days and in the entire mid-rib in 8-9 days. In necrotic responses, SMV was at and surrounding the inoculation site in 8-9 days and in veins and the mid-rib by 17 days but was restricted to necrotic areas. No virus was detected at any time in resistant responses. These results demonstrate that resistance is manifested by reduced and delayed replication and movement of virus.

CHARACTERIZATION OF AN RNA-DEPENDENT RNA POLYMERASE ACTIVITY ASSOCIATED WITH LA FRANCE ISOMETRIC VIRUS. Michael M. Goodin, Beth Schlagnhauer, and C. Peter Romaine, Dept. of Plant Pathology, The Pennsylvania State University, University Park PA, 16802.

Nine double-stranded RNAs (dsRNAs), ranging in size from 3.8-0.8 kbp, are diagnostic of button mushrooms, *Agaricus bisporus*, afflicted with La France disease. These dsRNAs are encapsidated by 36 nm virus-like particles that we have named La France isometric virus (LIV). We believe that LIV plays a major role in the etiology of La France disease. Therefore, we have initiated studies aimed at the molecular and biochemical characterization of this virus, which has led to the identification of a virion-associated RNA-dependent RNA polymerase (RDRP). The polymerase activity was resistant to  $\alpha$ -amanitin and actinomycin D and was dependent on magnesium, a requirement that could not be replaced by manganese. Polymerase products synthesized *in vitro* were determined to be single-stranded RNA based on sensitivity to RNase A and CF-11 cellulose column chromatography. Further, the RNA product hybridized to all nine LIV genomic dsRNAs and to one, but not both, strands of single-stranded DNA copies of a 2 kbp cDNA corresponding to the 3.8 kbp dsRNA which putatively encodes the polymerase. These data suggest that the LIV-associated RDRP is responsible for the *in vitro* synthesis of viral mRNAs.

OZONE-INDUCED FOLIAR INJURY ON TREES IN SHENANDOAH NATIONAL PARK, VA., 1991 - 1993. E. S. Hildebrand and J.M. Skelly. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

During late summer of 1991, '92, and '93, ozone-induced foliar injury was evaluated on mature canopy hardwoods within three Trend Plots adjacent to three air quality monitoring stations in the park. Plots consisted of 30 trees each of three ozone-sensitive species, black cherry (*Prunus serotina* Ehrh.), yellow-poplar (*Liriodendron tulipifera* L.), and white ash (*Fraxinus americana* L.). Six branchlets were taken from each tree and foliage was evaluated using the Horsfall-Barratt scale. In addition, 10 black cherry at each site were evaluated every two weeks from June through September 1992 to quantify symptom progression. Ozone exposures were greater in 1991 than in 1992; data from 1993 are not yet available. Incidence of injury was greater in 1991 than in 1992, and more moderate in 1993 on all three species evaluated. Cumulative ozone exposures were highest at Big Meadows, the high elevation site, and lowest at Sawmill Run, the low elevation site. Black cherry and white ash trees were most symptomatic at Big Meadows and the least symptomatic at Sawmill Run for all evaluations.

THE TRANSFER AND DISTRIBUTION OF STREPTOMYCIN-RESISTANCE GENE IN PLANT EPIPHYTIC AND PATHOGENIC BACTERIA. Tze-chung Huang, Thomas J. Burr, Cheryl L. Reid, Rodney G. Roberts\*. Department of Plant Pathology, Cornell University, NYSAES, Geneva, N.Y. 14456, and \*USDA/ARS, Wenatchee, WA

Streptomycin-resistant strains of a yellow-pigmented, plant epiphytic bacterium (YP) (taxonomically related to *Pseudomonas syringae*) were commonly detected in apple and pear orchards where blister spot and/or fire blight occurred in NY and WA. Resistance determinants of YP (21 strains examined) are carried on plasmids of various sizes which hybridize with SMP3, a specific DNA probe derived from *P. syringae* pv. *papulans* (Psp). Most strains of YP are very competent in transferring resistance to Psp with frequencies as high as  $\sim 10^{-1}$  to  $\sim 10^{-2}$  *in vitro* but not to *Erwinia amylovora* (Ea), *E. chrysanthemi* (Ech), or *Pantoea agglomerans* (Pa). The transjugation to Psp was also detected *in planta*. Although some strains of Pa hybridized with SMP3, they did not transfer resistance to Psp or Ea at detectable levels. None of the streptomycin-resistant strains of Ea from NY (11 strains) or from WA (94 strains) hybridized with SMP3, nor did Ech (6 strains) from Taiwan. The taxonomy of YP and its role in transferring resistance to pathogenic bacteria will be discussed.

MORPHOLOGY OF POWDERY MILDEW ON POINSETTIAS. S. H. Kim and T. N. Olson. Plant Disease Diagnostic Laboratory, Pennsylvania Department of Agriculture, Harrisburg 17110-9408.

Powdery mildew (PM) on Poinsettia (*Euphorbia pulcherrima*), a new disease in Pennsylvania in 1990, had an identical morphology to the PM of 1992 and 1993. Mycelium, 4 - 7  $\mu$ m in diam, persisted in circular patches on leaves. Conidiophores, 20 - 120  $\mu$ m long with up to 4 septations, often had a bent basal cell. Up to 9 conidia formed in chains. Oval to cylindrical conidia, 15 - 20 X 25 - 50  $\mu$ m, lacked fibrosin bodies, and contained up to 9 clear vacuoles, 3 - 20  $\mu$ m in diam, per conidium. Conidia on water agar had 34 % germination in 24 hr of continuous fluorescent light at 19 C. Germ tubes were simple or branched. The PM lacked cleistothecia.

IDENTIFICATION OF A MATING POPULATION WITHIN THE *FUSARIUM NYGAMAI* ANAMORPH. J. A. Klaasen and P. E. Nelson. Fusarium Research Center, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

*Fusarium nygamai* has morphological characteristics similar to *Fusarium* species currently included in section *Liseola* and *Elegans* (Burgess & Trimboli, 1986, Mycologia 78:223). This species forms short chains of microconidia similar to species in the section *Liseola*, and also forms chlamydospores, similar to species in section *Elegans*. Isolates of *F. nygamai* from Australia and South Africa were mated, and produced perithecia on carrot agar. Successful crosses yield typical *Gibberella* species teleomorphs. Two hermaphroditic isolates, one from Australia and the other from South Africa, of opposite mating types were identified. One hundred and twenty random ascospore progeny produced short conidial chains and chlamydospores on carnation leaf agar. Sixty progeny of a cross between the two hermaphrodites were checked for female fertility in order to develop female fertile tester strains for more reliable detection of other members of this mating population. Only 15 progeny were female fertile.

RELATIONSHIPS BETWEEN FOLIAR OZONE INJURY AND PHYSIOLOGY AMONG BLACK CHERRY GENOTYPES. K. B. Kouterick, J. M. Skelly, T. S. Fredericksen, J. E. Savage, and K. R. Snyder. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

The effect of ambient ozone ( $O_3$ ) exposure on seedlings of black cherry genotypes was investigated in northcentral Pennsylvania. Ozone exclusion treatments were administered to half-sib (HS) families R-12 and MO-7, and wild-type (WT) grown in open-top chambers. Foliar  $O_3$  injury was related to leaf gas exchange and stem volume over a growing season. Half-sib R-12 exhibited the most severe foliar injury, and WT seedlings had slightly less injury. Half-sib MO-7 had the least foliar injury. Leaf injury increased with increasing  $O_3$  exposures. Stomatal conductance values were greatest in HS R-12, followed by WT and HS MO-7. No consistent pattern was observed for photosynthesis and respiration rates. Differing  $O_3$  exposure did not affect stem volume, but overall HS R-12 had higher stem volume than MO-7 and WT seedlings. Foliar response to  $O_3$  among genotypes exhibits a relationship with stomatal conductance.

STRUCTURAL AND FUNCTIONAL ANALYSIS OF *ERWINIA AMYLOVORA* HARPIN, AN ELICITOR OF THE PLANT HYPERSENSITIVE RESPONSE. Ron J. Laby and Steven V. Beer. Department of Plant Pathology, Cornell University Ithaca, NY 14853.

The deduced amino acid sequence of harpin<sub>Ea</sub> and harpin<sub>Ear</sub>, of *E. amylovora* Ea321 (a pathogen of pomaceous plants) and Ea246 (a pathogen of *Rubus* spp.), respectively, were compared. The proteins are >97% identical; the only differences occurred in an N-terminal glycine-rich portion of each protein. Deletion analysis indicates that the C-terminal portion of each protein is not required for HR-eliciting activity on non-host plants. However, the C-terminal portion of each protein is required for secretion by bacteria.

WARMER AUTUMN TEMPERATURES REDUCE FREEZING TOLERANCE OF *PICEA ABIES* AND *PINUS SYLVESTRIS*. W. H. Livingston, Dept. For. Bio., Univ. Maine, Orono, ME 04469; and A. Lindström, Inst. f. skogsprod., Sveriges lantbruksuniv., S776 98 Garpenberg, Sweden

How climate change will influence freezing tolerance will determine the survival of many trees in the future. This study examined how warmer autumn temperatures could affect the freezing tolerance of spruce (*P. abies*) and pine (*P. sylvestris*) in Sweden. At the end of August, 3 mo. old seedlings were placed in two chambers differing in minimum (+5°C and +10°C) and maximum temperatures (0°C and 5°C above the average for Garpenberg; 60°15'N, 16°15'E). Seedlings were sampled in October, November, and December. Damage to the tissue was estimated by using relative conductivity. For November samples, warmer air temperatures resulted in increased damage at -20°C for spruce shoots, -25°C for pine shoots, and at -5°C or colder for roots of spruce and pine. Within the warmer chamber, seedlings growing in ca. 5°C colder soil had reduced freezing damage in shoots and roots. Warmer autumn temperatures will reduce freezing tolerance in roots and shoots of spruce and pine in Sweden. Cooler soil temperatures could minimize the effects of warm periods.

MAPLE HEALTH AND NUTRIENT STATUS IN OHIO'S SUGARBUSHES. R.P. Long, T.C. Weidensaul, and C.L. Capek. USDA Forest Service, 359 Main Road, Delaware, OH 43015, and Ohio Agricultural Research and Development Center, Wooster, OH 44691.

The health status of sugar (*Acer saccharum* Marsh.) and red (*A. rubrum* L.) maples in 35 sugarbushes was evaluated in 1991 and 1992. Injury and/or disease incidence and severity on roots and boles, and number of open tap holes were recorded for each of 700 trees in 1991. Percent crown dieback and foliage transparency were estimated for each tree in both years. From 175 trees, foliage, soil, and increment cores were sampled to evaluate nutrient status, age, and radial growth rates. Median wound severity was greater in bucket-collection than in tubing-collection sugarbushes. While most trees were healthy, in 1991 bucket-collection sugarbushes had higher proportions of trees with thin crowns (>15% transparency) compared to tubing-collection sugarbushes. Most foliar nutrients were within the range reported for "healthy" maples, but anomalously high foliar Al concentrations averaged >100 ppm in four sugarbushes.

FREEZING INJURY IN RED SPRUCE NEEDLES: INTEGRATING ELECTRICAL CONDUCTIVITY, GAS EXCHANGE AND FUNGAL INFECTIONS. Daniel K. Manter and William H. Livingston, Forest Biology Dept., University of Maine, Orono, Maine, 04469

Measurements of gas exchange, electrical conductivity and chlorophyll content were performed pre- and post-freezing (bi-weekly for six weeks beginning in November) in order to understand how the biology and physiology of damaged needles are affected by different levels of freezing injury. Foliage of forty-eight red spruce seedlings was exposed to a range of test temperatures (5, -5, -10, -15, -20, -25, -30 & -35°C); six seedlings per temperature. In addition, one branch per seedling was inoculated with *Rhizosphaera kalkhoffii* after freezing. *R. kalkhoffii* infections had the strongest correlation with decreasing freezing temperatures. These preliminary results suggest that these other measures in conjunction with electrical conductivity may be necessary to understand how red spruce needle's biology and physiology are affected by freezing damage.

ISOLATION, SELECTION AND SCREENING OF BACTERIA AS POTENTIAL BIOLOGICAL CONTROL AGENTS FOR *VENTURIA INAEQUALIS*. M. C. Matteson, T.J. Burr, C.A. Smith, M.R. Corral-Garcia. Dept. of Plant Pathology, Cornell University, NYSAES, Geneva, NY 14456.

Pure cultures of 1146 bacteria isolated from various sites in apple orchards were evaluated for their ability to prevent germination of conidia of *V. inaequalis* (Vi) *in vitro*. Eleven percent of the bacteria showed *in vitro* antibiosis and were further tested on apple seedlings for control of scab in the greenhouse. Bacteria from 24 h cultures on PDA were suspended in PO<sub>4</sub> buffer, adjusted to ~OD<sub>1.0</sub> and applied 24 h prior to inoculation with conidia of Vi. Seedlings were incubated in a mist chamber at 20°C for 48 h. Sixty seven percent of the strains showing activity *in vitro* reduced seedling infection. However, 42% of the 307 original bacteria not exhibiting *in vitro* activity currently screened in greenhouse tests also reduced seedling infection. One bacterium (strain 508) provided control equal to the fungicide captan. Strain 508 may be a non-pathogenic *Pseudomonas syringae*. We conclude that *in vitro* screening should not be the sole criterion in selecting biological control agents. A significant percentage of isolates tested to date exhibited no *in vitro* activity, but reduced disease on seedlings.

AN IPM PROGRAM FOR POWDERY MILDEW IN PUMPKIN THAT INCLUDES TIMING OF CHEMICAL CONTROL AND FUNGICIDE RESISTANCE CONSIDERATIONS. M. T. McGrath and H. Staniszevska, Department of Plant Pathology, Long Island Horticultural Research Laboratory, Cornell University, Riverhead, NY 11901.

Development of *Sphaerotheca fuliginea* in pumpkin was suppressed effectively with applications of triadimefon (14-day schedule) plus chlorothalonil (7-day) initiated after disease detection in 1989-1993. The decision criterion used was 1 leaf with symptoms out of 50 older leaves examined. There is ample time for a management response in this program because fungicide treatment was effective even when the actual mean disease incidence at the first application was as high as 33 affected leaves of 50 sampled leaves. However, control was inadequate when treatment was initiated 1 or 2 weeks later. One or two applications of triadimefon provided commercially acceptable control of powdery mildew and protected yield, but affected the pathogen population: the proportion of triadimefon-resistant isolates was 3% on 11 Aug 93 and 71% on 1 Sept 93. Myclobutanil and propiconazole (combined with chlorothalonil) were more effective than triadimefon; however, most triadimefon-resistant isolates also were less sensitive to these fungicides. Based on our results, we recommend applying a systemic fungicide (in combination with a contact fungicide) only at the start of powdery mildew development, as determined by an IPM scouting procedure.

GENETICALLY CONTROLLED NEEDLE RETENTION OF SCOTS PINE IN RELATION TO CYCLANEUSMA NEEDLECAST. Merrill W., and N. G. Wenner, 211 Buckhout Laboratory, University Park, PA. 16802.

Repeated applications of Bravo® 720 fungicide were made for three years to blocks of Scots pine Christmas trees in Centre and Clearfield Counties, PA, to prevent infection of the 1990, 1991 and 1992 needle complements by *Cyclaneusma minus*. In Oct 1992 infection of the fungicide-treated 1990 and 1991 complements were 9.0 and 0.5%, and 7.0 and 0.3%, respectively, in the two locations. In comparison, infection of the same complements in untreated trees were 99.0 and 89.5%, and 92.0 and 75.3%, respectively, in the two locations. Counts of remaining needles and fascicle scars showed that the fungicide applications significantly increased retention of third-year needles (P = 0.04 and 0.02), but not second-year needles (P = 0.54 and 0.86), respectively. However, some treated trees with very low levels of needle infection still cast virtually all third-year needles, and high percentages of second-year needles. This indicates a host genetic factor that affects needle retention independent of the hosts susceptibility to *C. minus*, and which has confounded all previous studies on the relationship of *Cyclaneusma* needlecast to needle retention in Scots pine.

DEVELOPMENT OF FOUR ISOLATES AND TWO INTRASPECIFIC HYBRIDS OF HETERODERA SCHACHTII ON THREE SUGARBEET X BETA PROCUMBENS INTERSPECIFIC HYBRIDS. L. J. Miller, Dept. of Plant Path., Phys. & Weed Sci., Virginia Tech, Blacksburg, VA 24061

Four physiologically different isolates of *H. schachtii* (C1 from tomato and C2 from sugarbeet in California, N1 from cabbage in New York, M1 from sugarbeet in Michigan and two intraspecific nematode hybrids derived from C1 x N1 and C2 x M1) were tested to determine their ability to develop egg-bearing females in interaction with USH11 sugarbeet (susceptible check), *B. procumbens* (resistant check) and three S4 progeny of Lewellen's *H. schachtii* interspecific hybrids derived in crosses of sugarbeet lines x *B. procumbens* (N 103, N 206, and N 207). All nematode isolates and nematode hybrids developed numerous females on USH11; C1, M1 and C2 x M1 on N 207; and C2 x M1 on N 206. No nematode isolate or nematode hybrid reproduced on *B. procumbens*; C1 on N103 and N 206; N1 and C1 x N1 on N 103, N 206, and N 207; and M1 and C2 x M1 on N 103. N 103 and N 206 were poor hosts of C2. N 206 was a moderately susceptible host of M1 as was N 207 of the C2 isolate.



ESTABLISHMENT AND PERSISTENCE OF *Gliocladium virens* IN TURF. A. S. Mintz, S. M. Poch, and C. P. Baker. W. R. Grace & Co.-Conn., Washington Research Center, Columbia, MD 21044.

*GlioGard*<sup>™</sup>, a dry product formulation of *Gliocladium virens* for controlling *Rhizoctonia* and *Pythium* diseases in greenhouses, and a water-dispersible formulation of *G. virens* were used to inoculate plots of Apache tall fescue. Grass, thatch and soil samples were collected from each plot for nine weeks and assayed on selective media. Samples from *Gliocladium*-treated plots were compared to samples from untreated plots and from plots treated with the chemical fungicide, Vorlan. *Gliocladium* soil populations in the treated plots remained unchanged for the duration of the test, while thatch populations were moderately increased. *Gliocladium* treatments significantly increased grass populations and persisted on the blades for seven weeks after inoculation. Colonization and persistence of *Gliocladium* on grass was more effective from the dry formulation than the water-dispersible formulation.

SPECIFICITY OF GERMINATION OF A BIOCONTROL FUNGUS. Sue Mischke, USDA, ARS, Systematic Botany and Mycology Laboratory, Beltsville MD 20705.

*Sporidesmium sclerotivorum* is a beneficial fungus that in nature behaves as an obligate parasite of sclerotia of *Sclerotinia minor* and related plant pathogenic fungi. Isolates are known to differ in their ability to infect and destroy sclerotia in the soil. Fungal hosts of *Sp. sclerotivorum* produce one or more germination activators that initiate macroconidial germination. These activators are associated with the melanized rind or cortex of sclerotia. Germination of *Sp. sclerotivorum* is followed by infection of the sclerotium, and living sclerotia are known to be more quickly colonized than heat-killed sclerotia. However even non-viable sclerotia can initiate macroconidial germination of *Sp. sclerotivorum*. Germination has not been stimulated by viable sclerotia of non-host plant pathogens. The factor(s) stimulating germination have properties of neutral lipids with low water solubility.

REACTION OF HETERODERA GLYCINES TO APPLICATION OF POULTRY MANURE ON THE DELMARVA PENINSULA. M.A. Morant, and C.B. Brooks, Department of Agriculture, University of Maryland Eastern Shore, Princess Anne, MD 21853

This study investigated the relationship between application of Poultry Manure (PM) and Soybean Cyst Nematode (SCN), the major pest of soybean on the Delmarva Peninsula. There was a significant ( $P=0.01$ ) negative correlation between cysts and PM, with a 50% reduction at 4 t/ha compared to 2 t/ha PM for the susceptible CV Essex. Seed yield was significantly ( $P=0.1$ ) affected by PM with highest yield for Essex and Manokin, (resistant) respectively, occurring when 2 t/ha PM was applied. Population of juveniles increased by approximately 50% when soil adjacent to Essex was sampled at 1 and 2 mo, respectively after application of PM. The opposite was true for Manokin. PM may stimulate SCN reproduction.

POTATO PINK ROT IN DELAWARE AND NEW JERSEY CAUSED BY PHYTOPHTHORA NICOTIANAE. R. P. Mulrooney, R. B. Carroll, A. L. Morehart and D. P. Whittington, University of Delaware, Newark, DE 19717-1303; S. H. Kim and T. O. Olson, Pennsylvania Dept. of Agriculture, Harrisburg, PA 17110-9408, and S. A. Johnston, Rutgers Univ., Bridgeton, NJ 08302-9499.

Potatoes with pink rot were collected from seven fields in DE and NJ during 1993. Cultures of *Phytophthora* were obtained on V-8 agar. Pathogenicity was assessed via inoculation of healthy tubers. Microscopic features of the pathogen included: heterothallism; nearly spherical sporangia with conspicuous papillae; uniform hyphae with swellings; oogonia with thick (2  $\mu$ m) rough walls, and nearly spherical, amphigynous antheridia. The fungus grew above 35°C. Based on these characteristics and mating studies, the causal agent was identified as *Phytophthora nicotianae* A2 (*P. parasitica* A2). This is the first report of this fungus causing pink rot of potatoes in the mid-Atlantic region.

IDENTIFICATION OF POSSIBLE VECTORS OF OAK LEAF SCORCH, CAUSED BY THE BACTERIUM *XYLELLA FASTIDIOSA*. M. W. Olszewski and A. B. Gould, Department of Plant Pathology, Rutgers University, NJ 08903.

*Xylella fastidiosa* is vectored by xylem-feeding insects in the Cicadellinae and in the Cercopidae. To identify possible vectors of this bacterium in oaks, yellow sticky traps were positioned at three different heights in each of 10 mature red oak trees in Moorestown, New Jersey. The traps were collected every ten days from May to October 1993, and insects retained on the traps were removed, tentatively sorted into different species, and counted. To isolate bacteria from collected insects, representative insects from each group were surface sterilized, and the heads were removed and ground in phosphate buffer. The resulting suspension from four individual insects within a single treehopper species tested positive for *Xylella* using ELISA, and the bacterium was isolated from these insects using XF-26 and CS-20 media.

ANTIBIOTIC PRODUCING ISRAELI STRAINS OF *ERWINIA HERBICOLA* AND CONTROL OF FIRE BLIGHT. Yakir Ophir and Steven V. Beer. Department of Plant Pathology, Cornell University, Ithaca, NY 14853

Of more than 900 Israeli strains of *E. herbicola* isolated from the surfaces of symptomless plants, 45% produced antibiotics on glycerol-ammonium medium that were inhibitory to *E. amylovora*. Based on the cultural conditions under which they inhibited the fire blight pathogen and their biological spectra of activity, the 405 strains produced at least fifteen different antibiotics. The ability of selected strains of *E. herbicola* to suppress the development of fire blight symptoms in immature pear fruits and on apple blossoms in a controlled-environment chamber was tested. Three strains, Eh423, Eh424 and Eh425, proved particularly effective and also showed promise under orchard conditions. *Tn5*-induced mutants of Eh423 and Eh425, which had lost their ability to inhibit *E. amylovora* *in vitro*, also lost their ability to suppress fire blight in immature pear fruits and on apple blossoms. These results suggest that antibiotic production is important to the biological control ability of these strains.

A TEMPORAL STUDY OF CATION CONCENTRATIONS IN RED SPRUCE WOOD DECAYED BY WHITE ROT AND BROWN ROT FUNGI. A. Ostrofsky and J. Jellison, Dept. of Plant Biology and Pathology, Univ. of Maine, Orono, ME 04469 and K.T. Smith and W.C. Shortle, USDA Forest Service, Durham, NH 03824.

Changes in cation concentrations in red spruce wood degraded by eleven white and brown rot fungi were monitored over time. Wood was degraded by fungi in modified ASTM soil block assays. Cation concentrations were determined by Inductively Coupled Plasma Spectroscopy. Changes in cation concentrations varied with fungal species and with extent of decay. Decay often resulted in a progressive increase in concentrations of calcium and magnesium. For example, calcium concentrations increased from 21.1  $\mu$ mol/g to from 30.6 to 92.2  $\mu$ mol/g in 8 months. Iron concentrations were high in wood decayed by *Postia placenta*. A better understanding of cation changes associated with the degradative process can yield insight into the basic processes involved in fungal decay of wood.

BICARBONATES AND BOTRYTIS: I. EFFECTS OF SALTS ON *IN VITRO* COLONY GROWTH OF *BOTRYTIS CINEREA*. C. L. Palmer, R. K. Horst, R. W. Langhans, and H. W. Israel. Departments of Floriculture & Ornamental Horticulture and of Plant Pathology, Cornell University, Ithaca, NY 14853.

To determine whether the bicarbonate anion or its cation affects *Botrytis cinerea* Pers., radial growth of spore-initiated colonies was examined on PDA supplemented to concentrations of 0.05 M with various ammonium, potassium, and sodium salts. Additionally, the effects of pH on growth were examined on PDA containing 0.05 M  $\text{KH}_2\text{PO}_4$ , 0.05 M  $\text{K}_2\text{HPO}_4$ , 0.01 M  $(\text{NH}_4)_2\text{SO}_4$ , 0.02 M  $\text{NH}_4\text{HCO}_3$ , or no supplement. Colony diameters were measured daily from 0 to 144 h. Chloride, nitrate, sulfate, and thiosulfate salts did not affect *B. cinerea* growth. Strong bases and reducing agents inhibited *B. cinerea* growth, as did bicarbonates, carbonates, and phosphates. Except for  $\text{NH}_4\text{HCO}_3$ , cations did not affect colony growth. Throughout examined pH range, phosphate inhibited *B. cinerea* growth. At pH > 8.0,  $(\text{NH}_4)_2\text{SO}_4$  decreased growth slightly, but  $\text{NH}_4\text{HCO}_3$  completely prevented growth at pH 7.0-8.5. While ammonium or ammonia gas may contribute to *B. cinerea* growth inhibition, anions primarily determined effect on growth. Although high pH alone reduces growth, bicarbonate and phosphate anions negatively affect *B. cinerea* colony growth *in vitro*. (Supported by H&I Agritech, Ithaca, NY 14850.)

EFFECTS OF POLYGALACTURONASE AND ENDOGLUCANASE FROM *AGROBACTERIUM VITIS* ON ROOT DECAY AND TRANSFORMATION OF GRAPE. X. Pu, P. Rodriguez-Palenzuela\*, T. Herlache\*, T. J. Burr, and A. Collmer\*. Department of Plant Pathology, Cornell University, Geneva, and \*Ithaca, N.Y. 14856.

*Agrobacterium vitis* (formerly *A. tumefaciens* biovar 3), the causal agent of grape crown gall, is unique among *Agrobacterium* species in its high degree of host specificity, its ability to infect grapevine systemically, and its ability to cause decay of grape juvenile roots. The production of two cell-wall degrading enzymes, polygalacturonase (PG) and endoglucanase (EG), was reportedly associated with this root decay. The genes that encoding these two pectic enzymes have been cloned from *A. vitis* strains. Transferring plasmids encoding PG or EG or both enzymes into *A. tumefaciens* biovar 1 strains resulted in detectable enzyme activities *in vitro*, however, root decay of grape seedlings was not observed. This suggests that the pectic enzymes, PG and EG, may not be the only factor that involved in root decay of grapes. The role of PG and EG in *Agrobacterium*-mediated transformation of grapevine is currently being studied.

WILD GRAPE (*VITIS RIPARIA*) AS A SOURCE OF *AGROBACTERIUM VITIS*, THE PATHOGEN CAUSING GRAPE CROWN GALL. Cheryl L. Reid and Thomas J. Burr, Department of Plant Pathology, Cornell University, NYSAES, Geneva, NY 14856.

Symptomless *Vitis* cuttings may be systemically infected with *A. vitis*, the cause of grape crown gall. We investigated *Vitis riparia* for the presence of *A. vitis*, since it is a widespread weed and possible host for this pathogen. Roots from 66 wild grape vines were collected from non-vineyard areas for the isolation of *A. vitis*. Roots were ground in selective Roy-Sasser (RS) broth and plated on selective RS medium. Colonies typical of *A. vitis* were then tested for reaction to a species specific monoclonal antibody. Those strains which reacted positively with the monoclonal antibody were screened for pathogenicity on 3 hosts. Polygalacturonase, endoglucanase, and seedling decay tests were performed. We recovered 547 *A. vitis* strains from 41 of the 66 vines. Of the 238 strains tested thus far, none are tumorigenic. Should *A. vitis* strains from *V. riparia* be able to acquire the tumor inducing (Ti) plasmid from tumorigenic strains, the management of *V. riparia* as a host for *A. vitis* around commercial vineyards will become important.

EVIDENCE FOR THE INDEPENDENT REPLICATION OF THE VIRUSES ASSOCIATED WITH LA FRANCE DISEASE OF *AGARICUS BISPORUS*. C. P. Romaine and B. Schlaghauser, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

La France disease of *Agaricus bisporus* is associated with a dsRNA isometric virus (LIV) infecting alone or in concert with a ssRNA bacilliform virus (MBV). MBV is thought to occur only as a coinfection with LIV, which raises the possibility of a satellite-helper virus relationship. Using PCR amplification of sequences targeted to the LIV and MBV genomes (sensitivity to <10 fg of viral RNA), we found that mushrooms having symptoms characteristic of La France disease were either infected singly by LIV or doubly by LIV and MBV. Mushrooms having atypical symptoms were either noninfected or infected solely by MBV. A partial cDNA clone of MBV RNA shared extensive homology in amino acid sequence with the putative RNA polymerases of ssRNA plant viruses, suggesting that MBV RNA encodes an enzyme for its replication. Our data support the hypothesis that LIV and MBV replicate independently of each other and that MBV is not required for pathogenesis. A broader survey of mushroom germplasms is needed to disclose whether infection by MBV alone is related to a pathology.

ECONOMIC IMPACTS OF FAILURE TO CONTROL SUMMER DISEASES ON 'LIBERTY' APPLES. D. A. Rosenberger, F. W. Meyer, and C. A. Engle, Cornell's Hudson Valley Lab, P. O. Box 727, Highland, NY 12528.

'Liberty' apple trees propagated on M.9 EMLA and planted in 1987 were used to determine the economic impacts of failure to control sooty blotch and fly-speck (SBFS). The three treatments were a control (no fungicides), a summer-only program comprised of three applications of Topsin M plus Captan, and a full-season fungicide program which included five applications of Nova plus Dithane followed by two applications of Topsin M plus Captan. Fruit from all plots were harvested, weighed, and graded. Sales value of the crop was estimated using average prices paid for the various grades and sizes of 'Empire' apples during the 1990-91 and 1991-92 marketing years. Development of summer diseases was favored by the unusually cool, wet summer in 1992. The percentage of fruit down-graded because of SBFS was 73, 32, and 11% for the control, summer program, and full spray program, respectively. For the same treatments, production measured as number of 42-lb bushels per acre was 915, 1046 and 1094, and total return per acre was \$3,486, \$5,847, and \$7,568. Control of SBFS is essential for maximizing returns with 'Liberty' if this cultivar is to be marketed like other commercial cultivars.

INHIBITION OF SCLEROTIAL GROWTH OF *COLLETOTRICHUM COCCODES* BY SIMULATED "SUNLIGHT". S. Sanogo, S.P. Pennypacker, and R. Stevenson. Department of Plant Pathology, The Pennsylvania State University, PA 16802.

The effect of light intensity on sclerotial growth of *Colletotrichum coccodes* was assessed by use of a 1000-watt solar simulator operated in a temperature-controlled chamber. One sclerotium was placed onto the center of 5-cm glass petri plates containing 2% water agar medium. The plates were subjected to five intensity levels (260, 460, 660, 860, and 1060 w.m<sup>-2</sup>) of electromagnetic radiation (>300 nm). At each light intensity level, a set of four plates was irradiated daily for 14 hrs followed by 10 hrs of darkness for a period of 5 days. Another set of four plates, serving as a control treatment, was maintained over the 5-day period in continuous darkness. Temperature at the surface of the water agar medium was controlled at 28±1°C. At the end of the 5-day period, the mean radial growth of sclerotia in irradiated (I) and non-irradiated (N) plates was determined and the proportion of inhibition in sclerotial growth (Y) was assessed as Y=1-I/N. Regression analysis indicated that the inhibition in sclerotial growth of *C. coccodes* was inversely related to the square of light intensity (P<0.001, Ra<sup>2</sup>=0.72).

INTEGRATING A HOST RESISTANCE FACTOR INTO A POTATO EARLY BLIGHT FORECASTING MODEL. Joel Shuman, B.J. Christ and S.P. Pennypacker, Dept. of Plant Pathology, Pennsylvania State University, University Park, PA 16802

The FAST (Forecast of *Alternaria solani* on Tomatoes) system originally developed to schedule fungicide sprays to control early blight on tomato was utilized to control early blight on potato. Our objective was to incorporate into FAST critical severity values that reflect differential cultivar resistance to early blight. The cultivars Atlantic, Katahdin, Norchip, and Norwis which range from susceptible to moderately resistant were each treated with the fungicide Bravo 720. Five fungicide treatments included a 7-day schedule initiated after flowering, a non-sprayed control, and three schedules based on various levels of FAST severity values. A 4x5 factorial in randomized complete block design with four replicates was utilized. Disease severity and yield components were evaluated for all plots. There were significant differences in disease severity and yield among cultivars and among spray schedules. There were cultivar by spray schedule interactions. Preliminary results indicate that critical levels of severity values can be established to account for cultivar resistance when scheduling fungicide applications for the control of potato early blight.

RESPONSE OF OPEN POLLINATED BLACK CHERRY FAMILIES TO AMBIENT OZONE UNDER OPEN-TOP CHAMBER CONDITIONS. J.M. Skelly, J. E. Savage, K.R. Snyder, T.S. Fredericksen, and K.B. Kouterick. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Eight half-sib (HS; open-pollinated) black cherry (*Prunus serotina* Ehrh.) families were established as 1-yr-old seedlings within open-top chambers in northcentral Pennsylvania. Seedlings were planted in 1990 and 1991 into chambers receiving charcoal-filtered, half-filtered, and nonfiltered air, as well as open plots representing 49, 74, 97, and 100% of ambient ozone exposures. Following 3 seasons of exposure (1991-1993), HS-R12 proved to be most sensitive to ambient ozone based upon foliar adaxial stippling; HS-MO7 was most tolerant, being asymptomatic even in open plots. Five families (MO 17, MO 23, MO 5, MO 22, and M 15) were intermediate in sensitivity. HS-R12 proved to be most sensitive in a second set of treatment chambers established in 1992 as compared to HS-MO7 and wildtype seedlings. Results are applicable to the further development of a native tree species for use in ozone injury detection surveys.

EFFECT OF UREA AND SELECTED BIOLOGICAL CONTROL ORGANISMS ON THE OVERWINTERING OF *VENTURIA INAEQUALIS* ON APPLE. Charles Smith, Thomas Burr, Mary Catherine Matteson, Matt Forsline. Department of Plant Pathology, Cornell University, NYSAES, Geneva, N.Y. 14856.

Apple leaves with visible scab lesions were collected in November and dipped in different concentrations of urea or suspensions of fungi and bacteria that showed *in vitro* antibiosis to *Venturia inaequalis*. The leaves were placed in open-mesh onion bags on the ground in the orchard for overwintering. Apple trees were sprayed with 5% and 10% (w/v) solutions of urea. Sprayed leaves were also placed in onion bags on the orchard floor. In May of the following year leaves were evaluated for percent decomposition and numbers of ascospores of *V. inaequalis* produced. Numbers of ascospores that were released from 20 leaf discs (37.5mm<sup>2</sup>) were recorded for each treatment. Urea and *Athelia bombacina* were the most effective treatments for reducing ascospores. Ascospore numbers were significantly less from leaves dipped in urea as compared to leaves sprayed while on trees.



SUCCESSFUL MANAGEMENT OF POWDERY MILDEW IN SQUASH WITH GENETIC AND CHEMICAL CONTROLS. H. Staniszewska and M. T. McGrath, Department of Plant Pathology, Long Island Horticultural Research Laboratory, Cornell University, Riverhead, NY 11901.

Management strategies for *Sphaerotheca fuliginea* in summer squash were evaluated in two experiments in 1993 comparing resistant PSX 2287 with susceptible Goldbar and resistant HMX 1707 with susceptible Supersett. Powdery mildew was suppressed in all hybrids with a fungicide program (triadimefon, benomyl and chlorothalonil) initiated after disease detection. These fungicides were not as effective on the abaxial leaf surface of the susceptible hybrids (average severities on 30 Sep on adaxial/abaxial leaf surfaces of non-treated and fungicide-treated Supersett were 26%/52% and 0%/49%, respectively). Genetic control performed well compared with chemical control in terms of disease suppression (average severities on 30 Sep for non-treated HMX 1707 were 3%/14%), but not in terms of yield (29% higher for treated Supersett than for non-treated HMX 1707). An integrated management program (fungicides applied to a resistant hybrid) provided exceptional powdery mildew control (0%/0.1% for treated HMX 1707) but did not result in a significant increase in yield for HMX 1707.

PREVENTIVE AND THERAPEUTIC MANAGEMENT OF DUTCH ELM DISEASE WITH FENPROPIMORPH AND PROPICONAZOLE FUNGICIDES. R. J. Stipes\*, Dept. of Plant Pathology, Physiology and Weed Science, VPI&SU, Blacksburg, VA 24061-0331.

Some newer elm injectables are fascinatingly promising for the prevention and/or therapeutic management of Dutch elm disease (DED). We evaluated fenpropimorph and propiconazole, ergosterol biosynthesis inhibitors. Fenpropimorph sulfate was injected preventively or therapeutically into large landscape American elms (*Ulmus americana*) at 0.83 or 2.0 g a. i./DBH inch. Trees were artificially inoculated in 5 random crown sites @ ca.13X10 spores/incision. Two years after treatment and injection, 14%, 42% and 100% of preventive, curative and inoculated control trees, respectively, exhibited DED symptoms. In companion studies with propiconazole (Banner, Alamo) at rates of 0.8 - 1.4 gm a. i./DBH inch, DED averaged 5% or less in preventive trees and up to 50% in curative trees. Both compounds are highly promising for DED control, but dosage elevations seem strongly indicated. Translocation and residue profiles must be defined.

CHARACTERIZATION OF *FUSARIUM OXYSPORUM* f.sp. *LYCOPERSICI* FROM MARYLAND AND EGYPT. Patrice Suleman, Abdel Mohsen Tohamy, and David Straney, Dept. of Botany, University of Maryland, College Park; second author, AGERI, Giza, Egypt.

Fusarium isolates were collected from wilted tomato plants in Egypt and from soils in Maryland with a history of tomato cultivation. Five Egyptian and seven Maryland isolates were identified as *Fusarium oxysporum* f.sp. *lycopersici* race 1 by morphological features, pathogenicity on differential hosts, and symptoms on tomato. The virulence of these isolates was measured on the tomato cultivars Super Marmande and Bonny Best by disease index, vascular discoloration, and colonization. A wide variation in virulence was observed in strains from both locations. All isolates displayed limited colonization and discoloration in Race1-resistant cultivars. Phytoalexins, rishitin and tomatine at 50 µg/ml reduced spore germination by 18 and 22%, respectively, and germ tube elongation by 60-75%. The variation in tolerance to these tomato phytoalexins are being compared to determine whether this correlates with variation in virulence between isolates.

*CRYPHONECTRIA PARASITICA* CANKERS ON SCARLET OAK (*QUERCUS COCCINEA*) IN PENNSYLVANIA. M. L. Torsello, D. D. Davis, and B. L. Nash. Department of Plant Pathology, The Pennsylvania State University, 210 Buckhout Laboratory, University Park, PA, 16802.

Field surveys revealed that basal and/or bole cankers caused by *C. parasitica* (= *Endothia parasitica*) occurred on approximately 15% of the scarlet oak throughout the natural range of this species in Pennsylvania. Macroscopic signs (pycnidia or stroma) of *C. parasitica* were associated with 67% of the typical cankers, and *C. parasitica* was recovered from ca. 70% of attempted isolations. Radial growth at breast height (1.4 m, BH) was less on scarlet oaks having light-to-moderate basal cankers as compared to apparently healthy trees. In contrast, scarlet oaks with severe basal cankers exhibited greater radial growth at BH, as compared to healthy trees, an anomaly possibly related to apparent growth stimulation at BH due to callus tissue. Severely infected trees also showed greater high-frequency variation in the annual growth pattern, as compared to healthy trees. This is the first report of *C. parasitica* cankers on scarlet oak in Pennsylvania.

SUSCEPTIBILITY OF SCAB-RESISTANT APPLE CULTIVARS TO THREE SUMMER FRUIT ROTS, LEAFSPOTS, AND SIX INSECT PESTS IN PA. J. W. Travis, J. L. Rytter, and E. G. Rajotte, Depts. of Plant Pathology and Entomology, Penn State University, Univ. Park, PA 16802.

Fruit of seven scab-resistant apple cultivars were wound-inoculated with *Botryosphaeria obtusa*, *B. dothidea*, and *Glomerella cingulata* under laboratory conditions. Significant differences among cultivars and the amount of fruit rot caused by these pathogens were observed. Redfree, Nova Easygro, and Williams Pride were most susceptible to fruit rot caused by all pathogens when fruit was inoculated immediately after harvest and after being in cold storage for 30 days. The other cultivars varied in their susceptibility to each pathogen. In 1992, a planting of four scab-resistant cultivars on Mark rootstock was established. On 4 May, scouting for the presence of disease and insect pests began and continued on a weekly basis until 10 Aug. Observations for the entire season showed Freedom to be most susceptible to leafspot infection. Susceptibility to insect pests varied. Golden Delicious controls were most susceptible to green apple aphids and leafrollers. Liberty was most susceptible to potato leafhopper damage with Freedom being the most resistant. Freedom, however, was most susceptible to leafminer feeding and Liberty the most resistant. Liberty was the most susceptible cultivar to Japanese beetle damage.

VARIABILITY ASSOCIATED WITH LESIONS ON GRAPE LEAVES CAUSED BY *GUIGNARDIA BIDWELLII*. D. L. Truxall and J. W. Travis. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802

In order to improve risk assessment for scouting black rot in the field, the amount of inoculum associated with leaf lesions needs to be defined. Leaves of *Vitis labrusca*, cultivar 'Niagra', infected with *Guignardia bidwellii* were collected from unsprayed vineyards in Erie County, Pa. The leaves were assessed for the number of lesions per leaf, the number of pycnidia per lesion, and the area of the lesions. The number of lesions per leaf ranged from 1 - 17, but more than 50% of the infected leaves surveyed contained only 1 - 5 lesions. The average size of lesions was 0.2 cm<sup>2</sup> or less in more than 70% of the leaves surveyed. Fewer than 50 pycnidia were observed in more than 70% of the lesions surveyed, but some lesions were found to contain up to 140 pycnidia. A significant relationship was found between the size of the lesions and the number of pycnidia per lesion at a level of P = 0.001. The linear relationship between these variables was not strong, resulting in an r<sup>2</sup> of less than 0.5. Therefore, assessments of the amount of secondary inoculum produced should utilize the size of the lesions, as well as the number of lesions on the leaf.

EFFECT OF ORCHARD FLOOR MANAGEMENT PRACTICES ON THE SEVERITY OF FIRE BLIGHT ON APPLE AND PEAR. T. van der Zwet and D. M. Glenn, USDA, ARS, Appalachian Fruit Research Station, Kearneysville, WV 25430.

Bartlett pear and Jonathan apple trees were planted in 1990 in a randomized block design with three orchard floor management schemes: narrow (0.3 m) or wide (3.0 m) herbicide strips, or root pruning trees 1.2 m from the trunks. In the third season (1993), tree trunk diameters were significantly smaller in narrow strip (47.4 mm) rows than in wide strip (57.5 mm) or root pruned (56.7 mm) rows. Similarly, leaf analysis (1992) indicated 2.26%, 2.46% and 2.47% nitrogen, respectively. After a few strikes of fire blight (*Erwinia amylovora*) in 1992, the trees became severely infected in 1993. By late August, the number of blighted shoots/trees was significantly lower on trees in narrow strip (3.9) rows than in wide strip (9.2) or root pruned (8.8) rows. The above data were calculated for pear and apple combined. Integrated orchard management practices that reduce shoot growth may become an effective way to minimize or control fire blight.

HRPO REGULATES EXPRESSION OF MULTIPLE HRP GENES AND HARPIN SYNTHESIS. Zhong-Min Wei and Steven V. Beer. Department of Plant Pathology, Cornell University, Ithaca, NY 14853

*hrpO*, one of the two regulatory genes, is located in a 3.8 kb *EcoRI* fragment of the *hrp* gene cluster of *E. amylovora*. It is an independent transcriptional unit with 540 base pairs, capable of encoding a 20 kD regulatory protein. HrpO is essential both for HR and for pathogenicity by *E. amylovora*. Merodiploids of strain Ea321 containing cloned *hrpO* (in pCPP1078) elicit the HR without *de novo* mRNA and protein synthesis, whereas the wild-type strain does not. The expression of five of six *hrp* loci fused with Tn5gusA1 are dependent on the presence of a functional HrpO. Synthesis of harpin is also regulated by HrpO. Western blots showed that harpin production by Ea321 directly results from the presence of HrpO. No significant DNA or protein sequence similarities have been found between *hrpO* and other sequences deposited in Genbank. Thus, HrpO may represent a new class of regulatory proteins essential for pathogenicity or virulence by bacteria.



CHEMICAL CONTROL OF *CHRYSOMYXA WEIRII* ON *PICEA PUNGENS*. Wenner, N. G., and Merrill, W., 211 Buckhout Laboratory, University Park, PA 16802.

Bravo® 720 and ASC 66792 fungicides were applied at rates of 6.51 and 9.4 liter f.p./H, respectively, to blocks of severely affected *Picea pungens* Christmas trees in Lackawanna County, PA to prevent infection of the 1992 needle complement by the autoecious needle rust *Chrysomyxa weirii*. Applications were made with a Solo® backpack mist blower on 23 and 29 May and 1 June 1992, beginning when some buds had broken on about 10% of the trees. In mid-May 1993, after symptom development but before needle casting, four twigs were removed, one at each cardinal direction, at 0.5 m above ground from each of ten permanent sample trees in each treatment block. The proportion of infected needles on the 1992 internode of each twig was determined and averaged by tree. Average needle infection was 35.4% in non-treated check trees, 0.5% in Bravo-treated trees, and 0.00% in ASC 66792-treated trees. Both treatments differed significantly from the check at  $P < 0.0005$ . Because Bravo® 720 is registered for control of *Rhizosphaera needlecast* on spruce, a spray schedule initiated 4 to 6 days earlier than normal for *Rhizosphaera needlecast* should control both pathogens.

IDENTIFICATION OF ECTOTROPHIC ROOT-INFECTING (ERI) FUNGI OF TURFGRASSES WITH THE RANDOM AMPLIFIED POLYMORPHIC DNA (RAPD) ASSAY. H.C. Wetzel, P.H. Demoen and P.D. Millner.\* Department of Agronomy, The University of Maryland, College Park, MD 20742. \*ARS-BARC Soil Microbial Systems Lab, Beltsville, MD 20705.

Turfgrass patch diseases caused by ERI fungi are among the most devastating in the U.S. The precise identification of ERI fungi can be difficult and time consuming because it requires laboratory isolation and culture to produce ascocarps. Production of the sexual stage in the laboratory is frequently unsuccessful because incompatible mating types may have been isolated or specific inducers of the sexual stage are unknown. Since these fungi can be isolated from their hosts, tentative identifications are currently made on the basis of cultural morphological characteristics. To reduce the dependence on phenotypic characteristics of ERI fungi, establishment of a new technique of identification that relies on inherent genotypic characteristics of these organisms is desirable. We investigated the use of RAPD for the identification of the ERI fungi. Initial RAPD reactions consisted of screening 37 reference isolates of five genera against 16 primers (10-base oligonucleotides). Characterization of the RAPD fingerprints of the 37 reference isolates allowed the comparison of the RAPD fingerprints of 20 unknown ERI fungi isolated from symptomatic turfgrass plants. Comparison of RAPD fingerprints between the reference and unknown isolates showed that ERI fungi could be distinguished at the species and variety levels.

METALAXYL SENSITIVITY OF ISOLATES OF PHYTOPHTHORA NICOTIANAE OBTAINED FROM DELAWARE POTATOES. D. P. Whittington, R. B. Carroll, R. P. Mulrooney, and A. L. Morehart, University of Delaware, Newark, DE 19717-1303; S. H. Kim and T. N. Olson, Pennsylvania Dept. of Agriculture, Harrisburg, PA 17110-9408.

Six isolates of *Phytophthora nicotianae* and one of *P. erythrosetpica* were assessed for sensitivity to metalaxyl. Isolates were grown on PARP medium for 7-8 days. Mycelial plugs were placed on PARP amended with metalaxyl at 0.0, 0.3, 0.6, 1.0, 10, 50, and 100 ppm. Radial growth was measured after 7 days at 28°C. All *P. nicotianae* isolates grew at 0.3, 0.6, and 1.0 ppm, only one grew slightly at 10 ppm and none above 50 ppm. For *P. erythrosetpica*, there was no growth at 0.3 ppm, only slight growth at 0.6 ppm, and none above this rate. All isolates of *P. nicotianae* were less sensitive to metalaxyl than *P. erythrosetpica*. This may impact the effectiveness of metalaxyl for pink rot control.

NEW HOSTS AND ASSOCIATIONS FOR PHYTOPHTHORA SPECIES. R. L. Wick, P. Haviland and T. Tattar. Department of Plant Pathology, University of Massachusetts, Amherst, MA 01003.

All host/*Phytophthora* associations listed here were naturally-occurring. Koch's postulates have been carried out for the following hosts and associated *Phytophthora* species: *Taxus baccata*, *P. citrophthora* and *P. citricola*; both species were weakly pathogenic to roots. *Gardenia jasminoides*, *P. parasitica*, A<sub>1</sub> and A<sub>2</sub> mating types; some isolates were highly pathogenic to gardenia roots and some were weakly pathogenic. *Lamium maculatum*, *P. citrophthora*, highly pathogenic to crowns and stems. *Spathiphyllum*, *P. drechsleri*, root rot. Koch's postulates have not been carried out for the following hosts: *Ilex opaca*, *P. cinnamomi*, *P. citricola*, crown and root rot. *Pachysandra terminalis*, *P. citrophthora*, root rot. *Camellia japonica*, *P. citricola*, root rot. *Arctostaphylos uva-ursi*, *P. cactorum*, crown rot.

MUTATIONAL ANALYSIS OF *GLIOCLADIUM VIRENS* TO DETERMINE THE ROLE OF GLIOTOXIN IN SUPPRESSION OF *PYTHIUM* DAMPING-OFF. Stephen E. Wilhite, David C. Straney, and Robert D. Lumsden. Department of Botany, University of Maryland, College Park, MD 20742. Third author: Biocontrol of Plant Disease Laboratory, USDA/ARS, Beltsville, MD 20705.

Certain strains of *G. virens* produce the antimicrobial metabolite, gliotoxin. Seven UV-mutants of strain G20-4VIB (WT) lacking gliotoxin production were isolated using selection-based enrichment and screening procedures. Colony appearance, sporulation, and mutants' ability to grow on minimal medium were comparable to G20-4VIB. However, mutants lost an average of 45% of the *in vivo* disease-suppressive activity of the WT strain, and experienced a near-total loss of *in vitro* antibiosis towards *P. ultimum*. This supports a major role for gliotoxin in *G. virens*' suppression of *P. ultimum*, and represents conclusive genetic evidence that antibiosis acts in fungal-biocontrol of plant disease.

A NON-ISOTOPIC PCR- COUPLED LCR REACTION ASSAY FOR THE IDENTIFICATION OF *ERWINIA STEWARTII*. W. J. Wilson<sup>1</sup>, C. A. Batt<sup>2</sup>, and H. R. Dillard<sup>1</sup>. Department of Plant Pathology, Cornell University, Geneva, NY 14456<sup>1</sup> and Department of Food Science, Cornell University, Ithaca, NY 14853<sup>2</sup>.

A non-isotopic polymerase chain reaction (PCR)- coupled ligase chain reaction (LCR) assay was developed to directly detect the plant pathogenic bacterium *Erwinia stewartii* in plant and vector material. The technique allows identification to species level based on a single-base-pair difference in the 16S rRNA gene which is unique to *E. stewartii*. A portion of the 16S rRNA gene of *E. stewartii* and of the closely related bacterium *E. herbicola* was sequenced. From comparison of the two 16S rDNA regions, two primer pairs were constructed such that only *E. stewartii* DNA gave a product in the LCR assay. An initial PCR amplification of the 16S rRNA gene increased sensitivity of the LCR to 10<sup>3</sup> CFU/ml without isolation or enrichment of the *E. stewartii*.

VECTORING OF THE BIOCONTROL AGENT *GLIOCLADIUM ROSEUM* TO RASPBERRY FLOWERS BY BUMBLE BEES AND HONEY BEES. H. Yu and J. C. Sutton. Dept. of Environmental Biology, University of Guelph, Guelph, Ontario, Canada N1G 2W1

The ability of bumble bees (*Bombus impatiens*) and honey bees (*Apis mellifera*) to vector *G. roseum* to flowers of Boyne and Redwing raspberries was investigated in field plots enclosed with polypropylene shade cloth to confine the bees. Hives of bumble bees and of honey bees were positioned in the enclosures (one hive/plot) after flowering began. A powder formulation of *G. roseum* (10<sup>9</sup> cfu/g) was placed in an inoculum dispenser on each hive. When emerging from the dispensers, bumble bees and honey bees carried a mean of 2.7 x 10<sup>5</sup> cfu and 3.0 x 10<sup>5</sup> cfu of *G. roseum* per bee respectively. Inoculum density of *G. roseum* in raspberries foraged by bumble bees, honey bees, and by spraying (5 x 10<sup>6</sup> conidia of *G. roseum*/mL water plus 0.05% Triton XR-100) averaged 2400, 800, and 5700 cfu/flower, respectively in Boyne, and 2200, 2100, and 3100 cfu/flower, respectively in Redwing. Vectored inoculum effectively suppressed *Botrytis cinerea* on the stamens and stigmas of each cultivar. Both bumble bees and honey bees were effective vectors in warm temperatures, but bumble bees were superior under cool temperatures in the Boyne plots.

RHIZOCTONIA SOLANI ANASTOMOSIS GROUPS AND FUNGI ASSOCIATED WITH TURFGRASSES AFFECTED BY BROWN PATCH. M. Zhang and P.H. Demoen, Department of Agronomy, The University of Maryland at College Park, MD 20742.

Determining anastomosis groups may provide valuable information relative to host range, pathogenicity and fungicide sensitivity of *Rhizoctonia solani* Kuhn. *R. solani* isolates were obtained from turfgrasses affected with brown patch in Maryland in 1992 and 1993. Three methods (i.e., cellophane strip, clean glass slide, and petri dish) were compared for their ease in identifying *R. solani* anastomosis groups. Growing isolates in a petri dish with a thin layer of water agar was the most simple and reliable method. *R. solani* isolates belonging to AG-1 were most frequently obtained from perennial ryegrass (*Lolium perenne* L.) and AG-2 isolates were commonly identified from bentgrass (*Agrostis* spp.). Lower frequencies of AG-2 and AG-4 also were isolated from perennial ryegrass. From a total of 810 samples of crown or root tissue obtained from diseased perennial ryegrass or colonial bentgrass (*A. tenuis* Sibth), only two isolates of *R. solani* were obtained from crowns of colonial bentgrass and perennial ryegrass and one from root tissue of perennial ryegrass. All of these isolates belonged to AG-2. *Bipolaris* sp., *Curvularia* spp., *Fusarium* spp., *Phoma* sp., *Acremonium* sp., *Gaeumannomyces* sp., and *Phialophora* sp. were frequently isolated from leaves, crowns or roots of perennial ryegrass plants affected with brown patch.

POPULATION DYNAMICS OF *GLIOCLADIUM ROSEUM* ON BLACK SPRUCE SEEDLINGS IN RELATION TO BIOCONTROL OF *BOTRYTIS CINEREA*. P.G. Zhang, A.A. Hopkin, and J.C. Sutton\*, Great Lakes Forestry Center, Sault Ste. Marie, Ontario P6A 5M7, Canada, and \*Dept. of Environmental Biology, University of Guelph, Ontario N1G 2W1, Canada.

Seedlings of black spruce (*Picea mariana*) were kept under normal growing conditions or stressed by keeping them in darkness at 40 C for 44 h, and then inoculated with conidia of *G. roseum*. The seedlings were then maintained at 12, 20, and 28 C for 0, 6, 12, 24, 48, 72, 96, 120, and 168 h, after which conidia were recovered from the needles, counted, and assessed for ability to germinate. Sporulation incidence of *G. roseum* was estimated on needles that were surface sterilized and then incubated for 8 days. The postinoculation temperature treatments did not significantly affect numbers of conidia recovered or sporulation incidence of *G. roseum* on the needles. However, percent germination of recovered *G. roseum* conidia declined with increase in temperature and treatment period in both stressed and non-stressed seedlings; these relationships were adequately described by a Weibull regression model. In a separate experiment, stressed seedlings were inoculated with *G. roseum* 0, 1, 2, 4, and 6 days before challenge-inoculation with *B. cinerea*. Results showed that ability of *G. roseum* to suppress *B. cinerea* decreased with time between inoculation with *G. roseum* and challenge inoculation with *B. cinerea*. The decrease in biocontrol effectiveness correlated with decrease in percent germination of *G. roseum* conidia.

LOW LIGHT INTENSITY PREDISPOSES BLACK SPRUCE SEEDLINGS TO INFECTION BY *BOTRYTIS CINEREA*. P.G. Zhang, J.C. Sutton\*, B. He\*, and A.A. Hopkin, Great Lakes Forestry Center, Sault Ste. Marie, Ontario P6A 5M7, Canada, and \*Dept. of Environmental Biology, University of Guelph, Ontario N1G 2W1, Canada.

Low light condition was examined as a possible factor predisposing seedlings of black spruce (*Picea mariana*) to infection by *B. cinerea*. Four-month-old seedlings were kept in low light intensities of 0, 7, 15 and 30  $\mu\text{mol m}^{-2}\text{s}^{-1}$  for 0 - 45 days at 3-day intervals, inoculated with the pathogen ( $10^6$  spores  $\text{ml}^{-1}$ ), and then placed in high humidity at 20 C for 36 h. Sporulation incidence in needles from seedlings kept in light intensities of 0 and 7  $\mu\text{mol m}^{-2}\text{s}^{-1}$  increased with preinoculation treatment period to 45.6% and 32.2%, respectively, after 27 days, but decreased after longer periods. Sporulation incidence was < 6.3% when the preinoculation light intensity was 15 and 30  $\mu\text{mol m}^{-2}\text{s}^{-1}$ . In greenhouse tests, light intensity near the lower needles of seedlings growing in containers in the greenhouse decreased to < 20 and < 8  $\mu\text{mol m}^{-2}\text{s}^{-1}$  at about 40 and 80 days, respectively, after the seedling canopy closure. With the decrease in light intensity, sporulation incidence of *B. cinerea* in the lower needles of naturally and artificially inoculated seedlings increased. We conclude that low light intensities predisposed black spruce seedlings to infection by *B. cinerea*.

A STUDY ON THE CHROMOSOMAL HOMOLOGS OF THE NATIVE PLASMID pCXC100 IN *CLAVIBACTER XYLI* SSP. *CYNODONTIS*. Y.P. Zhang, T.Y. Li, & T.A. Chen. Department of Plant Pathology, Rutgers University, New Brunswick, NJ 08903.

Two repeated elements, H3 and X2, were identified on the 51-kb native plasmid, pCXC100, of *Clavibacter xyli* subsp. *cynodontis* (Cxc), which is a non-pathogenic, gram-positive bacterium inhabiting the xylem of Bermudagrass. Their chromosomal homologs, CH3 and CX2, were cloned from the genome of Cxc, respectively. Only one copy each of H3 and X2 was observed on pCXC100 when analyzing the entire plasmid DNA with the

repeated element probes. However, five CX2 copies and more than fifteen CH3 copies were observed on the Cxc genome. H3 and X2 were two independent repeated elements because no cross-hybridizations were detected between H3 and X2, H3 and CX2, X2 and CH3, and CH3 and CX2. According to DNA deletion analyses, a 0.6-kb fragment of H3 and a 1.0-kb fragment of X2 were responsible for the hybridization with Cxc chromosome. H3 was then sequenced and a pair of inverted repeat was found by comparing with the DNA database. DNA sequencing of CH3, X2 and CX2 is now in process.

COMPLEMENTATION OF *hrp* MUTANTS OF *Erwinia amylovora* BY *Escherichia coli* DH5 $\alpha$ (pCPP430). L. P. Zhao, Z.-M. Wei, and S. V. Beer. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

The *hrp* mutants of *E. amylovora* do not multiply in host tissues. Three days after vacuum infiltration of apple (host) leaf segments, the population of an *hrpN* mutant of *E. amylovora* remained at the initial level ( $2.2 \times 10^4$  CFU/cm $^2$ ), while the wild-type strain reached  $4 \times 10^7$  CFU/cm $^2$ . Coinoculation of either apple leaf sections or immature pear fruits with *E. amylovora hrp* mutants and *E. coli* DH5 $\alpha$  (pCPP430) resulted in multiplication to wild-type levels by all the *hrp* mutants tested. (pCPP430 is a cosmid harboring the entire functional *hrp* gene cluster of *E. amylovora*.) Coinoculation with the *hrpN* mutant version of pCPP430, did not affect the growth of *hrp* mutants in host tissues. *E. coli* DH5 $\alpha$ (pCPP430) did not increase the multiplication ability of *Xanthomonas campestris* and *Pseudomonas syringae hrp* mutants in apple or pear tissues. Since extracellular complementation was *hrpN* dependent, harpin may play a role in stimulating the multiplication of *hrp* mutants of *E. amylovora* in host tissues.

IMPROVED DISEASE CONTROL IN VEGETABLES WITH ELECTROSTATIC VS. HYDRAULIC SPRAYERS. T. A. Zitter, and R. C. Derksen. Department of Plant Pathology, and Department of Agricultural and Biological Engineering, Cornell University, Ithaca, NY 14853.

A prototype version of the Electrostatic Spraying Systems (ESS) air-assisted sprayer was compared with a hydraulic boom sprayer (STD) for disease control in 1992 and 1993 on Waltham butternut squash, and in 1993 on Celebrity tomato. Control of powdery mildew, Septoria leaf and fruit spot, and gummy stem blight/black rot on squash, and of early and late blight on tomato was addressed. The experimental design was a split plot with three reps of two spray systems as main plots and fungicide treatments as subplots. For squash, triadimefon, benomyl, and chlorothalonil, and potassium bicarbonate (KHCO $_3$ ) combined with benomyl were applied weekly with initiation based upon IPM scouting. For tomato, sprays of chlorothalonil, chlorothalonil + copper oxychloride + maneb, and KHCO $_3$  were initiated following CUFAST. Fungicides were used at labeled rates and KHCO $_3$  at 1% (w/v). Air delivery and small droplet technology (40 $\mu$  for ESS and 175 $\mu$  for STD) provided improved spray coverage with all materials despite lower spray volumes (4.5 gpa for ESS and 27 gpa for STD). This resulted in significantly less foliar and fruit infection and higher marketable yields in both crops. KHCO $_3$  with good spray coverage controlled early blight but not late blight.