ABSTRACTS OF PAPERS

Presented at Meetings of The American Phytopathological Society

The American Phytopathological Society

POTOMAC DIVISION

Annual Meeting March 19-21, 1980

ABSTRACTS

*Presented in the Graduate Student Award Competition of the Potomac Division of The American Phytopathological Society.

ANNOSUM ROOT ROT ASSOCIATED WITH REDUCED RADIAL GROWTH IN SOUTHERN PINE BEETLE INFESTED LOBLOLLY PINE. S. A. Alexander, J. M. Skelly and R. S. Webb. Department of Plant Pathology and Physiology, VPI & SU, Blacksburg, VA 24061.

Southern pine beetle (SPB) infested and noninfested (control) plots were established in thinned loblolly pine plantations. Root rot incidence was determined through root excavation and measurement of colonized and healthy roots. Annual radial growth was measured at a height of 1.4 m. Mean annual radial growth from 1968-77 for SPB and control plots was significantly different (P<0.01). Between SPB infested and noninfested trees in SPB plots, the average difference in radial growth was significant (P<0.01) with the SPB infested trees having 28% less radial growth than noninfested trees. Heterobasidion annosum colonized trees produced 39% and 33% less mean radial growth for 1973-77 and 1968-72, respectively, when compared to noncolonized trees (P<0.01). Average H. annosum colonization for SPB infested and noninfested trees was 54% and 11%, respectively (P<0.01).

EARLY, PREVISUAL, THERMAL IMAGERY DETECTION OF DUTCH ELM DISEASE. L. A. Alger, R. J. Stipes, H. J. Heikkenen and J. A. Daubert, VPI & SU, Blacksburg, VA. 24061.

Thermal imagery, with an AGA Thermovision 680 system, was used successfully to detect Dutch elm disease (DED) within 24 hours after artificial inoculation. One of two major leaders on each of four healthy *Ulmus americana* saplings was inoculated with ca. 3.3 X 10' conidia of *Cenatocystis ulmi* in July, 1979. Visual and thermal scanning observations of the trees revealed no symptoms of DED before inoculation, and no visible DED symptoms were observed during the thermal scanning observation period. Inoculated branches on three separate trees revealed elevated foliage temperatures 24, 48, and 144 hours after inoculation, but not on the fourth tree during this 144-hour observation period. Biopsies from all inoculated branches exhibited vascular discoloration and yielded *Cenatocystis ulmi* in culture.

CHEMICAL CONTROL OF MELOIDOGYNE HAPLA AND PRATYLENCHUS PENETRANS ON STRAWBERRY. D. E. Babineau and L. R. Krusberg. Dept. of Botany, Univ. of Maryland, College Park, MD 20742.*

The efficacy of nematicides in transplant water alone and in combination with other nematicides was evaluated for control of M. hapla and P. penetrans on strawberries grown for plant production. A sandy loam field was fumigated five weeks before nematode-infected plants were set. Standak and Vydate were applied in transplant water. Nemacur and Temik were applied in granular form in bands as postplant treatments and Fumazone was chisel-injected as a postplant, sidedress application. All treatments initially reduced soil populations of both nematode species. Single applications of nematicides in transplant water did not provide full-season nematode control. In tests of combinations, late-season control of $\underline{\text{M.}}$ hapla was best in plots treated with Nemacur while both Nemacur and Temik effectively reduced P. penetrans soil populations. Plant vigor and daughter-plant production were highest in Nemacur- and Temik-treated plots.

NUTRITIONAL STUDIES ON THE MYCOPARASITE SPORIDESMIUM SCLEROTIVORUM. E. A. Barnett, W. A. Ayers, and P. B. Adams, USDA, SEA, AR, Soilborne Diseases Lab., Beltsville, MD 20705.

Sporidesmium sclerotivorum, a mycoparasite of Sclerotinia spp. and certain other sclerotial fungi, grows and sporulates well on living sclerotia but poorly on synthetic culture media. An aqueous extract of sclerotia of Sclerotinia minor stimulated

the germination of macroconidia and the growth of \underline{S} . sclerotivorum. Glucose was the major carbohydrate and glutamate and citrulline the most abundant amino acids in the sclerotial extract. Optimum nutritional conditions for the growth of \underline{S} . sclerotivorum in culture include glucose or mannitol as a carbon source, casamino acids or glutamine as a nitrogen source, an initial pH of 5.0-5.5, and 10 $\mu g/1$ thiamine.

EVALUATION OF ULTRA-LOW VOLUME, LOW VOLUME AND DILUTE PESTICIDE APPLICATION TECHNIQUES ON APPLE. R. E. Barrat, J. L. Maas and R. E. Adams, 401 Brooks Hall, West Virginia University, Morgantown, WV 26506 and USDA, SEA, AR, Fruit Laboratory, Beltsville, MD 20705.*

Fifteen-year-old trees of four apple cultivars were sprayed at rates of 1 gallon per acre (gpa) for ultra-low volume (ULV) treatment, 20 gpa for low volume and 200 gpa for dilute treatment. Thirteen sprays were applied. Based on comparative pesticide deposits monitored biologically with Penicillium variabile and analytically with high-pressure liquid chromatography, there was increased foliar deposit and decreased pesticide drift with ULV technique, while using less pesticide. The mean diameters of spray droplets measured by optical array spectrometer were 41 nm for the ULV, 72 nm for the low volume and 147 nm for the dilute treatments. There was no significant difference in control of Venturia inequalis or Aphis pomi among the three application techniques, all of which were economically acceptable.

OZONE EFFECTS ON LONG-TERM RADIAL INCREMENT GROWTH AND REPRODUCTION OF EASTERN WHITE PINE. Larry F. Benoit. Dept. of Plant Pathology & Physiology, VPI&SU, Blacksburg, VA 24061.*

To determine ozone(03) effects on long-term growth of native eastern white pines, radial increment cores were taken on 10 plots of 3 trees each, with foliar ratings of trees tolerant, intermediate and sensitive to 03. The plots range 446 km from Skyline Dr. in Shenandoah Nat'l Pk. to Blue Ridge Pkwy. in VA. Average annual radial growth of the sensitive class was significantly less(P=.01) than that of the tolerant class for the period 1955-78. Growth increments in the intermediate class were between the tolerant and sensitive classes, while growth in all classes showed net declines for the period. The same 30 trees are being used to study the 0_3 effect on reproduction and possible differential effect between sensitivity classes. Pollen was exposed to 15 pphm 03 or charcoalfiltered air for 8 h periods. Exposure was dry, before germination, or in distilled water at the start of germination or both. After a 60 h germination period at 27C, % germination was recorded for 300 random pollen grains for each treatment. Significant (P= .01) reduction in pollen germination resulted from 03 treatments.

STOMATAL INFILTRATION OF INOCULUM AS A TECHNIQUE FOR INDUCING RESISTANCE TO ANTHRACNOSE IN CUCUMBER.

G. C. Bergstrom and S. Diachun. Department of Plant Pathology, University of Kentucky, Lexington, KY 40546.

Induction of disease resistance in cucurbits in the field is limited by the moisture requirement for infection by inducing organisms. Infection of cucumber and induction of systemic resistance to anthracnose, caused by Colletotrichum lagenarium, were accomplished at ambient greenhouse humidities by stomatal infiltration of inoculum. The inoculum was sprayed into leaf 1 from an atomizer held a few inches from the lower leaf surface. Infiltration of C. lagenarium race 1 spores into leaf 1 induced resistance in leaf 2 to surface challenge with race 1. Stomatal infiltration with an isolate of race 2, non-pathogenic on cucumber, induced systemic resistance to race 1. This

technique was also used to induce resistance to anthracnose using $\underline{\text{Pseudomonas lachrymans}}$ and $\underline{\text{Mycosphaerella}}$ $\underline{\text{melonis}}$ as the inducing organisms.

EXPRESSION OF TUBER RESISTANCE TO PHYTOPHTHORA INFESTANS IN RELATION TO PHYSIOLOGIC AGE OF TUBERS. S. K. Bhatia and R. J. Young, 401 Brooks Hall, West Virginia University, Morgantown, WV 26506.

Tuber resistance to Phytophthora infestans was studied in relation to physiologic age in the cultivars Atzimba, Katahdin, Kennebec, and Sebago. Resistance was evaluated in (a) freshly harvested tubers; (b) tubers stored at 6 C for 3 months; and (c) tubers kept at 6 C for 6 months. Tubers of Atzimba were highly resistant to <a href="Phi-10:16:ed-10:

PEROXIDASE, POLYPHENOL OXIDASE AND CATALASE ACTIVITY IN CERATOCYSTIS ULMI. G. F. Bills, J. L. Brooks and D. F. Hindal, 401 Brooks Hall and 1026 Agricultural Sciences Building, West Virginia University, Morgantown, WV 26506.

Polyphenol oxidase or peroxidase activity was suspected of being associated with dark pigment formation and asexual reproduction in <u>Ceratocystis ulmi</u>. Spectrophotometric methods showed no polyphenol oxidase activity in mycelial extracts from a darkly pigmented isolate of <u>C. ulmi</u>, but faint bands of polyphenol oxidase were present after polyacrylamide-gel electrophoresis. Peroxidase and catalase activity from extracts of the darkly pigmented isolate was detected in spectrophotometric assays, but showed no relationship to the onset of pigmentation and sporulation during the first ten days of growth in potato-glucose broth. Peroxidase and catalase activity were similar among pigmented and non-pigmented isolates after ten days growth.

A LIPOPHILIC FRACTION FROM PHYTOPHTHORA INFESTANS ELICITS THE ACCUMULATION OF SESQUITERPENOID STRESS METABOLITES IN POTATO TUBBER. R. Bostock and J. Kuć, University of Kentucky, Lexington, KY 40546.

Based on the elicitor activity of an aqueous cell-free preparation of the mycelium of Phytophthora infestans, approximate-ly 60% of the activity was detected in a chloroform-methanol (2:1) extract of mycelium and 30% remained in the residue. Autoclaving the residue in water released 3% of the activity. Partitioning the extract (modified Folch) with pH 9-10 buffer drew more activity into the buffer than partitioning with pH 3.0 buffer or water. The specific activities of the high pH buffer and interfacial fluff were the highest of all fractions and extracts. Activity was detected in a protein-containing fraction of the fluff which eluted with chloroform on a Sephadex LH-20 column. The data indicate activity is associated with a proteo or glycoproteolipid.

INFLUENCE OF MELOIDOGYNE INCOGNITA AND GLOBODERA SOLANACEARUM ON DEVELOPMENT OF BLACK SHANK OF TOBACCO. Leslie A. Bower, J. A. Fox, and L. I. Miller. Dept. of Plant Pathology and Physiology, VPI & SU, Blacksburg, VA 24061.

The influence of Meloidogyne incognita (MI), and Globodera solanacearum (GS), on the development of stem lesions and wilting of Phytophthora parasitica var. nicotianae (PPN) infected greenhouse grown Nicotiana tabacum (cv. Virginia Gold) was studied. The treatments included I, PPN, II, PPN + MI; III, PPN + GS; IV, MI; V, GS; VI, Control (no PPN, MI or GS). Soil in 10.5 cm pots in which a single plant had grown for 4 wks was infested with 5000 eggs of either MI or GS. Ten wks after transplanting stab-wounded roots were inoculated with 2 ml of potato broth containing PPN mycelia. Twelve wks after transplanting there was an average of 283 and 369 GS females in treatments III and IV, respectively; and numerous root-knot galls in all MI treatments. Black shank lesion development on stems and wilting of PPN infected plants was greater in treatments II and III than in I and more wilting occurred in treatment II than in treatment III.

PHOSPHOLIPIDS OF MELOIDOGYNE JAVANICA. D. J. Chitwood and L. R. Krusberg, Botany Dept., University of Maryland, College Park,

Maryland 20742.*

Phospholipids comprised 5.4% of the dry wt of M. javanica. They consisted of 61.7% phosphatidylcholine (PC), 22.0% phosphatidylethanolamine (PE), and smaller quantities of 5 other lipids. Octadecenoic acid (18:1) comprised 61.3% of the phospholipid fatty acid; also abundant were 18:0 (8.0%) and 20:5 (9.5%). PE contained more 18:0 and 20:0 and less 20:5 than PC. Fatty acids esterified at the 1-position of PC or PE were shorter and more saturated than those esterified at the 2-position. The 1-alkyl-2-acyl and 1-alkenyl-2-acyl analogues of diacyl PE comprised 9.4% and 35.1%, respectively, of the total PE, whereas 98.9% of the PC was in the diacyl form. At least 95% of the alkenyl and alkyl moieties were 18:0. Fatty acid compositions of alkyl and alkenyl analogues of diacyl PE were similar to that of the 2-position of diacyl PE. Abundance of alkyl and alkenyl phospholipids is most likely a significant biochemical difference between M. javanica and its reported hosts.

REACTION OF ROOT CATION EXCHANGE CAPACITY OF PEACH SEEDLINGS TO XIPHENEMA AMERICANUM COBB. N. G. Conrad and R. E. Adams, 401 Brooks Hall, West Virginia University, Morgantown, WV 26506.

Root cation exchange capacity (CEC) of 3 selections of peach seedlings inoculated with Xiphenema americanum was compared to parameters used to assess host vigor and growth including; terminal growth, numbers of terminals and buds produced and the percentage of trees that retained their foliage for longer than 9 months. Seedlings were inoculated with 104 1.2 X. americanum in aliquots of infested Lehew sandy-loam soil or modified Baermann funnel extractions. Depending on the parameter used, host vigor or growth were reduced by 1.2-2 fold while inoculated root CEC showed a 1.7 to 2.3 fold reduction. Root CEC offers an accurate method of assessing host damage and may explain one mechanism by which nematodes damage their hosts.

EFFECT OF POSTHARVEST CALCIUM TREATMENT ON THE DECAY OF DELICIOUS APPLES. <u>William S. Conway</u>, Horticultural Crops Quality Laboratory, HSI, SEA-AR, U.S. Department of Agriculture, Beltsville, MD 20705.

Delicious apples, grown at the Beltsville Agricultural Research Center, were wounded on two sides and inoculated with a conidial suspension of Penicillium expansum. Twenty-four hours later the inoculated apples were treated with a 4% calcium chloride solution, either as a dip for 3 minutes or under pressure infiltration for 2 minutes at 1.03×10^5 Pa. (15 psi). After storage at 20 C for 7 days, the apples were rated for decay severity by measuring the diameter of the decayed area at the inoculation sites. When calculated on the basis of decayed area compared to the control, the apples treated by dipping had 10% less decay, while those apples pressure infiltrated with calcium chloride had 50% less decay than the nontreated fruit.

EPIDEMIOLOGY OF XANTHOMONAS CAMPESTRIS ON FIELD GROWN CABBAGE. L. E. Cytrynowicz and D. J. Fieldhouse, Univ. of Delaware, Newark, DE 19711.

Epiphytotic development of black rot of cabbage from a point source of $\underline{Xanthomonas}$ $\underline{campestris}$ and from contaminated seed was investigated. In a spring planting, five centrally located plants in each of four plots of transplanted Market Prize cabbage were spray-inoculated with a rifampicin-resistant strain of \underline{X} . $\underline{campestris}$ to determine pattern and rate of spread. A fall planting at a second location was direct-seeded with a mixture of Danish Ballhead cabbage and \underline{X} . $\underline{campestris}$ infested radish seed. Levels of infested seed were $\overline{0.0}$, 0.1 and 1.0%. All plots planted with a high level of infested seed had high disease incidence while infection in other plots varied with their position in the field. In both experiments, pathogen spread was in the direction of the prevailing wind.

VIRUS DISEASE OF THE CULTIVATED MUSHROOM TRANSMITTED BY CONTAMINATED SOIL. K. Deahl USDA-SEA-AR, Veg. Lab., Beltsville, MD 20705.

Trays containing a healthy strain of Agaricus bisporus were top-dressed with soil naturally or artifically infested with spores of several competitor fungi (weed molds). Pasteurized soil without weed molds was added to control trays. All treatments were maintained in a pest-free growth room.

Sporophore production was delayed 7-10 days on trays with the infested soil and sporophores that were produced exhibited symptoms typical of the virus disease known as LaFrance disease. Electron microscopy of the infected mushroom revealed two virus-like particles (VLPs), 26 and 35nm, commonly associated with LaFrance disease. Virus related double-stranded ribonucleic acid (dsRNA) was extracted from these tissues. Mushrooms from pasteurized casing soil displayed no symptoms and contained no detectable VLPs or dsRNA. Weed molds may be natural reservoirs for the mushroom virus(s).

THE IMPACT OF PHOTOCHEMICAL OXIDANT AIR POLLUTION ON BIOMASS DEVELOPMENT OF NATIVE VEGETATION AND SYMPTOM EXPRESSION OF ASCLEPIAS SPP. Stephen Duchelle, J. M. Skelly and L. W. Kress. Dept. of Pl. Path. & Phys., VPI & SU, Blacksburg, VA 24061.*

The effect of ambient ozone (03) on biomass production of vegetation native to the Big Meadows area of the Shenandoah Nat'l Park was examined. Emphasis was placed on studying Asclepias spp. as a possible bioindicator for 03. Twelve 10 ft diam. plots were randomly established with 4, each receiving charcoal-filtered and non-filtered air via open top chambers, and 4 receiving ambient air as open plots. The highest monthly average and the peak 1 h conc. of 0_3 for the 5 month study period was .051 and .095 ppm, respectively. Two collections of total above ground biomass yielded 9079g, 6171g, 3910g for the filtered, non-filtered and open plots, respectively. Chlorosis and purple stippling on the adaxial leaf surface were observed on Asclepias spp. grown in ambient air; those grown in charcoal-filtered air were asymptomatic and exhibited greater height growth. The results suggest 03 has reduced biomass and that Asclepias spp. could be used as a bioindicator of 03.

STIMULATION OF UREDOSPORE GERMINATION AND INDUCTION OF VACUOLATION BY NICOTINE AND RELATED COMPOUNDS. <u>Richard C. French</u> and Charles L. Wilson. USDA, SEA-AR, Plant Disease Research Laboratory, P.O. Box 1209, Frederick, Maryland 21701.

Nicotine is used in greenhouses to control aphids and other insects. It has also been reported to affect carotene metabolism in plants and fungi. We were interested in the effects of nicotine on uredospores produced in the greenhouse for other research studies. Nicotine at 25-250 μ l/l stimulated germination of uredospores of Puccinia coronata, P. graminis var. tritici, P. punctiformis, Uromyces phaseoli, U. vignae, but not P. helianthi. Several cyano and vinyl pyridines, compounds related to nicotine, also stimulated certain rust species. The stimulation of germination was not as great as that of previously studied compounds such as nonanal and/or β -ionone. At high concentrations, 500 - 1000 μ l/l, nicotine inhibited germination and induced extensive vacuolation in uredospores of many of the species tested. These chemically-induced vacuoles differentially accumulated neutral red, indicating an active metabolic uptake of this vital stain.

INHIBITORY ACTIVITY OF MICROBIAL AND PLANT POLYSACCHARIDES ON INFECTION OF SWEET SORGHUM BY SUGARCANE MOSAIC VIRUS. A. G. Gillaspie, Jr., C. A. Thomas, and B. Prescott. USDA/SEA/AR, Applied Plant Pathology Laboratory, Beltsville, MD 20705 and National Institutes of Health, Bethesda, MD 20205.

Polysaccharides from 23 bacterial, fungal, and plant sources were tested for inhibition of sugarcane mosaic virus (SCMV)-infection of sweet sorghum (Sorghum bicolor 'Rio'). Extracts from virus infected leaves were mixed with the polysaccharides and inoculated onto plants with an artist's airbrush. Polysaccharides from yeast cell walls and from Bacillus subtilis and Streptococcus pneumoniae Type III cell-free culture liquids inhibited SCMV-infection by 90-100% (symptom formation on treated plants compared to control plants inoculated only with SCMV) at concentrations as low as 250 µg/ml. The other polysaccharides inhibited infection by 29% or less at concentrations as high as 2000 µg/ml. Although several of the plant and microbial polysaccharides we tested are antigenically related, no relationship could be shown between inhibitory activity and antigenic properties.

AN EFFICIENT TECHNIQUE FOR PROLONGED STORAGE OF PHYTOPHTHORA INFESTANS: Robert W. Goth, USDA, SEA, AR, NER, BARC, HSI Vegetable Laboratory, GH 13, Bldg. 011, Range 1, BARC-West, Beltsville, MD 20705

A decoction containing 10 Golden Bantam sweet corn seeds in $50\ \mathrm{ml}$ of deionized distilled water served as a suitable medium for storage of isolates of races 0; 1; 1,4; and 1,2,3,4,

respectively, of <u>Phytophthora infestans</u>. All cultures were stored in the dark at $14\pm1^{\circ}\mathrm{C}$ for as long as 14 months without transfer. This reduction in number of transfers required for their maintenance, in turn, reduced the opportunity for contamination, selection of less virulent cultural variants, and reduced the amount of time and materials. Preliminary results also indicate that these isolates may have less tendency to mutate on this medium. Since corn grain is readily available in many countries, this method should also prove to be a convenient, efficient method for prolonged storage of <u>Phytophthora infestans</u> even in areas with limited facilities.

ENHANCED PEROXIDASE ACTIVITY AND LIGNIFICATION IN THE INDUCED SYSTEMIC PROTECTION OF CUCUMBER. R. Hammerschmidt and J. Kuć, Dept. of Plant Pathology, Univ. of Kentucky, Lexington, KY 40546.

Inoculation of one leaf of cucumber with Colletotrichum lagenarium systemically protected plants against disease caused by subsequent challenge with the fungus or Cladosporium cucumerinum. Associated with the protection were increased peroxidase (PRO) activity, quantitative changes in PRO isozymes, and enhanced lignification in response to challenge. Significant protection and increases in PRO activity occurred with as few as one lesion on the inducer leaf, whereas injury did not elicit these responses. The onset of protection was correlated with the appearance of elevated PRO activity. Epidermal cells of protected plants lignified more frequently in response to challenge with C. lagenarium or C. cucumerinum than did non-protected plants. The localization of protection in the epidermis was supported by decreased penetration into isolated epidermis of protected plants.

FUNGITOXIC MECHANISM OF THE TRIAZOLE FUNGICIDE CGA-64251 IN USTILAGO MAYDIS.

M. J. Henry and H. D. Sisler. Dept. of Botany, University of Maryland, College Park, MD 20742.

The fungicide 1-[2-(2,4-dichloropheny1)-4-ethy1-1,3-dioxolan-2-y1-methy1]-1H-1,2,4-triazole (CGA-64251) at concentrations ranging from 0.1 - 5.0 µg/ml inhibited sporidial multiplication in liquid cultures of <u>Ustilago maydis</u> approximately 15% after 4 hr and 58-70% after 12 hr. Sporidia were swollen and highly branched in the presence of the fungicide. The morphological aberrations resembled those induced by fenarimol. Total lipid as percent of dry wt was not affected by 0.1 or 5.0 µg/ml of CGA-64251 after 4 hr, but desmethyl sterol biosynthesis was reduced 87-92%. Accumulation of C-14 methyl sterol precursors of ergosterol accompanied inhibition of ergosterol synthesis and resulted in total sterol levels exceeding those of the control. Large quantities of free fatty acids were also present after treatment for 4 hr. Fungitoxity of CGA-64251 is attributed to inhibition of ergosterol biosynthesis at the stage of sterol C-14 demethylation.

ISOLATION OF CYLINDROCLADIUM PARVUM FROM WALNUT SEEDLINGS AND SOIL IN PENNSYLVANIA, VIRGINIA AND WEST VIRGINIA. Barry B. Hunter and Donald A. Roth, Dept. of Biology, California State College, California, PA 15419, and Dept. of Plant Pathology, University of Wyoming, Laramie, Wyoming, 82071.

Numerous small-spored forms of Cylindrocladium were isolated from nursery soil and necrotic black walnut seedlings in West Virginia and Virginia. Additional isolates were recovered from white walnut seedling rhizospheres in Pennsylvania. Fungal recovery from roots was made employing standard microbiological procedures, and from soil with azalea or geranium baiting. Condiophore vesicle morphologies were variable, and some isolates had conidia with a size range similar to the small-spored species, C. parvum, C. peruvianum and C. parvum, C. parvianum and the conidial sizes are invalid criteria for species delineation. We concluded that small-spored forms of Cylindrocladium should all be called C. parvum.

SEROLOGICAL RELATEDNESS OF ELM AND OAK SCORCH-ASSOCIATED BACTERIA TO THE PIERCE'S DISEASE (PD) BACTERIUM. S.J.Kostka, J.L.Sherald, S.S.Hearon, and J.F.Rissler. ESL,NCR, National Park Service, Wash,D.C. 20242; USDA,ARS,BARC, Beltsville,MD 20705; and Botany Dept., Univ. of MD, College Park, MD 20742.*

Rod-shaped, Gram negative bacteria measuring 0.4-0.5 μ m x 1.5-4.0 μ m were vacuum extracted in 0.01M phosphate buffer (pH 7.0) from stem sections of leaf scorch-affected American elms and

red oaks but not from symptomless trees. Indirect fluorescent antibody staining (IFAS) using antiserum to the PD bacterium demonstrated a positive serological relatedness between elm and oak bacteria and the PD bacterium. Extracts from symptom-less trees and smears of the elm wetwood agent (Enterobacter cloacae) did not react with the antiserum using IFAS. Elm and oak scorch-associated bacteria are serologically related to a xylem-limited bacterium that causes a similar leaf scorch disease in grape and almond. Consistent extraction of bacteria serologically related to the PD bacterium indicates the possible involvement of these organisms with elm and oak leaf

REDUCTION OF EASTERN FILBERT BLIGHT ON CORYLUS AVELLANA. Michael Kosztarab, M. K. Roane and Charles R. Drake. Department of Entomology and Department of Plant Pathology and Physiology, VPI & SU, Blacksburg, VA 24061.

Eastern filbert blight incidence in a small orchard of selected wild filbert (hazelnut) seedlings, Corylus avellana, was reduced over several years by use of an annual program involving the removal of diseased branches by pruning and burning, and five applications of Manzate 200 (4.5 kg/ha). Six to eight applications of Sevin 50 WP (4.5 kg/ha) were used for Japanese beetle control. Annual loss of untreated shrubs was as high as 19% while that of treated shrubs was 3%. Six asymptomatic shrubs may be possible sources of resistance and worthy of propagation.

NUTRITION AND PARASITISM OF THE CONTACT MYCOPARASITE ACLADIUM TENELLUM. W. R. Kuykendall, D. F. Hindal and H. L. Barnett, 401 Brooks Hall, West Virginia University, Morgantown, WV

Acladium tenellum, a contact biotrophic mycoparasite, has been found in association with Fusarium moniliforme diseased corn ears. It requires thiamine and biotin for normal axenic growth but does not require mycotrophein, a growth factor required by most contact mycoparasites. A medium containing alanine, asparagine or glutamic acid as the nitrogen source and glucose as the carbon source best supported growth and asexual sporulation by <u>A.</u> tenellum. Several darkly pigmented Deuteromycetes, including <u>Drechslera</u> carbonum, <u>Diplocarpon</u> rosae, <u>Diplodia</u> <u>zeae</u>, <u>Bispora</u> sp., and <u>Alternaria</u> alternata, were good hosts using the criteria of: 1, the formation of contact cells and 2, growth of the parasite in association with the host on media not supporting axenic growth of A. tenellum.

"BORDE BLANCO" DISEASE OF MAIZE IN LATIN AMERICA. Frances M. Latterell and Albert E. Rossi, USDA/SEA Plant Disease Research Laboratory, P.O. Box 1209, Frederick, Maryland 21701.

A Basidiomycete identified as a "reduced Agaric" is associated consistently with a distinctive disease of maize in Mexico, Nicaragua, and Costa Rica. Symptoms include the blanching of elongate marginal areas of leaves, lesions often extending to the midrib, but always beginning at the margin. Severe stalk lesions extend downward from sheaths infected at the juncture of the blade. Leaf lesions range in length from ca 5 to 25 cm, and from 1 to 3 cm in width. Lesions have concentric zones, each of a slightly different degree of decolorization. Minute basidiocarps resembling miniature agarics form amphigenously in the center "zone" of larger leaf lesions. We propose the Spanish name "borde blanco" (-white border) for this disease because of its prevalence in Latin America, and because of the characteristic white border lesions that glisten in sunlight in severely infected fields. Stalk lesions are numerous in fields showing a high percentage of infected leaves, and stalk rot is severe where foliage is dense and humidity is high.

MILD FORM OF HYDRANGEA VIRESCENCE ASSOCIATED WITH A MYCOPLASMALIKE ORGANISM (MLO) SUPPRESSES SYMPTOMS OF INFECTION BY THE SEVERE VIRESCENCE MLO. R. H. Lawson and F. F. Smith, U.S. Dept. of Agriculture, Beltsville Agricultural Research Center, Beltsville, MD 20705.

Hydrangea virescence (mild and severe) is associated with bud graft inocula that produce characteristic and stable symptoms with repeated transmission. Plants with severe symptoms (S) are stunted and have small leaves with vein yellowing and produce small green cymes. Plants with mild (M) disease symptoms have large symptomless leaves and cymes with large green florets. Sieve cells of plants with S symptoms con-

tained a high concentration of MLO's but the organism was difficult to detect in plants with M symptoms. Plants with M disease that were challenge inoculated with buds from S diseased plants showed mild vein yellowing that disappeared. Symptoms of S disease are greatly suppressed in plants previously infected with the M disease agent. Severe and mild virescence are apparently caused by related MLO's.

POTATO CULTIVAR SUSCEPTIBILITY TO OZONE AND SOIL-BORNE PATHOGENS. C. Lawton, A. L. Morehart, and R. B. Carroll Plant Science Dept., Univ. of Delaware, Newark, De. 19711.

In 1978 and 1979 studies were made of potato cultivar susceptibility to ozone damage, basal stem lesions and pink rot of Ozone caused severe damage on some cultivars. Sustubers. Ozone caused severe damage on some cultivars. Susceptibility to stem lesions, caused by Fusarium spp. and Rhizoctonia solani, was variable. Pink rot symptoms could not be produced in the field by direct inoculation with Phytophthora erythroseptica. Tuber infections were obtained on plots in which diseased tubers from commercial fields were incorporated the previous fall. In laboratory tests, pink rot symptoms were evident in tubers of all cultivars directly inoculated with P. erythroseptica. inoculated with P. erythroseptica.

A STUDY OF ROOT-GRAFT TRANSMISSION OF CERATOCYSTIS FAGACEARUM IN RED OAK SEEDLINGS. S. R. Leong and W. L. MacDonald, 401 Brooks Hall, West Virginia University, Morgantown, WV

A major lateral root from each individual of paired two-yearold red oaks was selected for grafting or tying. For grafted pairs an incision was made on each root, and the cut surfaces were placed perpendicular to one another and fastened with a grafting band. Root ties were made in a similar fashion with uninjured roots. After one season's growth, no union between grafted or tied roots was observed. However, callus formation around the incision was recorded for the grafted roots and an indented area at the point of contact occurred between tied roots. One seedling in each pair was inoculated in May and are currently being conducted to determine whether the lack of symptom development can be explained by a change of pathogenicity of C. fagacearum.

CALCIUM OXIDE FOR THE REDUCTION OF PREEMERGENCE DAMPING-OFF OF PEAS. J. A. Lewis and R. D. Lumsden. Soilborne Diseases Laboratory, PPI, SEA, USDA, Beltsville, MD 20705.

Preemergence damping-off of peas (Pisum sativum) in soil naturally infested with Pythium ultimum was reduced in the greenhouse and in the field by amendment with calcium oxide (CaO). Addition to soil of 0.2% CaO (w/w), 0, 2, 4 or 8 wk before planting resulted in a greenhouse stand of 86% which was comparable to that obtained with thiram-treated seed. CaO reduced disease in five cultivars in three soils infested with either of three $\frac{P}{P}$. ultimum isolates. Application of CaO to soil infurrow (31 g/m of row) in the field resulted in a 63% stand compared to 34% in the control. The mechanism for disease control is associated with an alkaling $\frac{P}{P}$. Which decreases control is associated with an alkaling $\frac{P}{P}$. trol is associated with an alkaline pH which decreases germination, growth, and survival of P. ultimum. After 1 wk, CaO (0.4% w/w) reduced pathogen propagules from 73/g of soil in the control to 16/g of soil.

NYLON SCREEN FOR STUDY OF THE ECOLOGY OF PYTHIUM SPP. IN SOIL. R. D. Lumsden, USDA, SEA, AR, Beltsville, Maryland 20705.

Oospores of Pythium spp., from V8-cholesterol cultures, were mounted in nylon screen fabric (Nitex $^{\overline{R}}$, Tetko, Inc., Elmsford, NY). Spores of P. ultimum were trapped in 20 µm-, P. aphanidermatum in 25 µm-, and P. myriotylum in 30 µm-pore size fabric by vacuum filtration and by gently rubbing them into the mesh. The fabric with trapped spores was cut into 1x3 cm pieces, buried in soil, incubated, retrieved and microscopically examined. Spores were dislodged from the mesh into H2O and were tested on agar media for germinability or presence of antagonistic microorganisms. In this way, mycoparasitism and antibiosis, including fungistasis, could be assessed for different soils. Lysis of mycelium, grown on 100 μ m-pore size fabric on agar medium, was determined by counting mycelial strands in a $400~\mu\text{m}^2$ area after burial in soil for intervals of incubation. The nylon mesh can also be used to view Pythium structures by scanning electron microscopy.

VERIFICATION OF AN EARLY BLIGHT FORECASTING SYSTEM ON FOUR TOMATO CULTIVARS. L. V. Madden, S. P. Pennypacker, and A. A. MacNab, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

A forecasting system for <u>Alternaria solani</u> on tomato (FAST) was verified during the 1979 season. To verify the system three treatments were applied to each of the following processing tomato cultivars: Merit, Dorchester, Red Rock, and 77B38, a new USDA cultivar released as being resistant to early blight. These cultivars were subjected to the following spray schedules: no spray (OD), 7-day (7D), and according to FAST. The 7D treatment rows were sprayed nine times; FAST treatments received only five sprays. Analysis of variance indicated no cultivarspray schedule interaction for either final disease severity or apparent infection rate. There was no significant difference in disease severity or infection rate for the FAST and 7D treatments; the amount of disease resulting from these two treatments was, however, significantly different from the check plots (OD). In summary, the spray schedule produced by the forecasting system was an efficient control technique.

OCCURRENCE AND DEGRADATION OF AFLATOXINS IN COMMERCIAL MUSHROOM COMPOST. J. Milburn, K. Deahl, and G. Bean. USDA-SEA-AR, Veg. Lab., Beltsville, MD 20705, and Botany Dept. Univ. of Md., College Park, MD 20742.

Aflatoxins may be introduced into compost either through natural production by Aspergillus spp. during mushroom cultivation processes or through the addition of aflatoxincontaminated nutrient supplements. Samples of compost taken from commercial houses contained aflatoxins B1, B2 and G1. Several substrates commonly used in mushroom growing media supported the growth of two Aspergillus spp. isolates, and aflatoxin production at 24 C. Pasteurized compost that was commercially prepared also supported growth of Aspergillus spp. and toxin production at 30 C. Corn supplemented with 150 ppb aflatoxin B₁ showed about a 60% reduction in aflatoxin concentration when subjected to commercial compost pasteurizing procedures. Compost supplemented with 24 ppb aflatoxin B at spawning showed no trace of aflatoxins 6 days later. No toxin was detected in mushrooms grown in the substrates.

EFFECTS OF VACUUM INFILTRATION OF CALCIUM CHLORIDE ON RIPENING RATE AND CHILLING INJURY OF TOMATO FRUIT. H.E. Moline, Hort. Crops Quality Lab., HSI, SEA-AR, USDA, Beltsville, MD 20705.

Two concentrations of CaCl2 (either 4 or 6%) were applied to fruit of 3 maturities and ripening rates were compared to fruit treated with distilled water. Sorting of fruit into maturity class and internal and external color change during ripening was recorded by optical measurement. Fruit were ripened at 200 for 14 days; chilled fruit were first held at 2C for 14 days, then ripened. Fruit color change was dependent upon the amount of vac. applied, concn. of CaCl2 used, maturity of fruit selected, and temperature. Calcium applied at 380 mm vac. had no significant effect on internal color change but reduced external color change of nonchilled fruit; chilled fruit had more internal color development while 6% CaCl2 increased external color and 4% reduced it. Calcium applied at 190 mm vac. did not affect color of nonchilled fruit and had variable effects on internal and external color of chilled fruit as affected by maturity. Calcium uptake can affect ripening rate and reduce chilling injury; 380 mm vac. damaged fruit.

PATHOGENIC POTENTIAL OF VERTICILLIUM ALBO-ATRUM PROTOPLASTS.
A. L. Morehart and G. L. Melchior, Univ. of Delaware, Newark,

Release of protoplasts from Verticillium albo-atrum mycelia treated with strepzyme and the pathogenic potential of these protoplasts in susceptible plants were studied. Thirty-nine Streptomyces spp. isolates were obtained from local soils on dilution-plates of a casein-glycerol medium. The isolates were grown on strepzyme production medium and the culture fluid assayed for strepzyme activity against \underline{V} . albo-atrum mycelia by agar-gel diffusion. Maximum strepzyme activity occurred in 5-7 days. Treatment of one-day-old \underline{V} . albo-atrum mycelia with 7 days. Treatment of one-day-old v, albo-actum mycella with partially purified strepzyme stabilized in $0.7\,\mathrm{M}$ MgSO $_4$ yielded maximum protoplast release in 20 hr. Test plants inoculated by root dip with 4.5 x $10^4/\mathrm{ml}$ protoplasts or conidia showed leaf chlorosis in five days and wilt in 10-14 days. v0 alboatrum was reisolated from all inoculated plants. These findatrum was reisolated from all inoculated plants. These ings suggest possible involvement of protoplasts in the systemic spread of fungi in vascular disease.

APPLICATION OF THE ENZYME-LINKED IMMUNOSORBENT ASSAY TO THE DE-TECTION OF TOBACCO RINGSPOT VIRUS IN GERANIUM. S. R. Newhart

and C. P. Romaine, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

The ability of the enzyme-linked immunosorbent assay (ELISA) to detect purified tobacco ringspot virus, diluted in leaf homogenates of the geranium (Pelargonium x hortorum), is markedly influenced by the composition of the extraction buffer. Greatest sensitivity was obtained when leaf homogenates were prepared in 0.1 M phosphate, pH 7.2, with 4% polyethylene glycol, or in PBS-T $(\overline{0}.02~\underline{M}$ phosphate, pH 7.4, 0.15 \underline{M} NaCl, 0.05% Tween-20) with 2% polyvinyl pyrrolidone (PVP). PBS-T with 0.5% 2-mercaptoethanol or 0.5% sodium diethyldithiocarbamate, used alone or in combination, provided lower sensitivities. Reliable detection was possible in PBS-T-PVP homogenates of cuttings, seedlings and seed of several geranium cultivars, and of plants latently infected for 6 months.

THE UTILIZATION OF SELECTIVE AGAR MEDIA IN CONJUNCTION WITH BAITING AND ROOT ISOLATIONS OF CYLINDROCLADIUM AND FUSARIUM. Joseph R. Newhouse and Barry B. Hunter, Dept. of Biology, California State College, California, PA 15419 *

Geranium and azalea baiting was used to assay naturally and artificially infested soils in Pennsylvania and West Virginia for Fusarium spp. and Cylindrocladium spp. Several selective agar media were used with the baiting techniques and root isolations to ascertain their efficacy for recovering Cylindrocladium and Fusarium. Glucose-lima bean agar (100:23:15 g/l) with antibiotics and varying amounts of rose bengal or malachite green/Tergitol was excellent for recovering pathogenic cylindrocladia. When yeast extract was substituted for lima bean extract the number of Cylindrocladium isolates decreased, whereas Fusarium spp. recovery was greatly enhanced. Geranium as bait for Cylindrocladium was superior to azalea because conidia were more abundant, internal hyphal colonization of the leaf was increased and more species were isolated. This bait aided earlier identification of the fungi and a greater incidence of recovery from the leaf tissue.

SOUTHERN BLIGHT OF COOL SEASON GRASSES. N. R. O'Neill, Field Crops Lab., SEA, USDA, Beltsville, MD 20705.

A new disease in annual bluegrass, characterized by circular and crescent-shaped diseased brown and yellow areas (20 to 140 cm), was observed on a golf course during the summers of 1978 and 1979. The symptoms progressed during hot and humid weather, causing the diseased areas to enlarge. Two types of sclerotia were isolated from the thatch in the advancing edge of diseased areas. Mature sclerotia were either small (1.5 mm), round, and black, or large (3.2 mm), irregularly shaped, and reddish-brown. Both isolations yielded <u>Sclerotium rolfsii</u>. The virulence, host range, growth rates, and growth temperature optima of these isolates were compared with 11 isolates of <u>S</u>. rolfsii from other hosts and locations. Isolates from annual bluegrass and bentgrass were more virulent to these hosts and to tobacco, tomato, peanut, and sugarbeet than isolates from non-grass hosts. Growth rates were positively correlated with virulence. Of the cool season grasses tested, perennial ryegrass, annual bluegrass and bentgrass were most susceptible, while red fescue and tall fescue were nearly immune.

PHYSIOLOGIC RACES OF COLLETOTRICHUM TRIFOLII. S. A. Ostazeski and J. H. Elgin, Jr., Field Crops Lab., SEA, USDA, Beltsville,

In 1979 we reported anthracnose on resistant 'Arc' alfalfa on Maryland's Eastern Shore. The Eastern Shore (ES) isolates have been compared with our standard (S) isolates and with new standards (NS) collected from Delaware, Maryland, Pennsylvania, and Virginia in 1979. ES, S, and NS isolates could not be separated by differences in growth at 10, 15, 20, 25, 30 or 35°C. There were no apparent differences among the S, NS, and ES isolates in spore size. ES and NS isolates were identical in agar culture. Mycelia of S isolates were darker than ES or NS isolates, probably a result of repeated culture transfer. pathogenicity tests using resistant Arc and 'Saranac AR' and susceptible 'Saranac' alfalfa cultivars, the S and NS isolates were virulent on Saranac only, but ES isolates were virulent also on Arc. Saranac AR was equally resistant to all isolates. Therefore, we propose that S and NS isolates be recognized as race 1, and ES isolates as race 2 of \underline{C} . $\underline{trifolii}$. The cultivars Arc, Saranac, and Saranac AR differentiate the two races.

INDUCED TOLERANCE OF TRICHODERMA HARZIANUM TO FUNGICIDES. G. C. Papavizas, USDA, SÈA-AR, Beltsville, Maryland 20705. Although wild strains of $\underline{\mathbf{T}}$. $\underline{\text{harzianum}}$ are being used experimentally for biological control of certain soil-borne plant diseases, very few attempts have been made to select or form new biotypes tolerant to fungicides and with enhanced biological control and survival ability in soil. Biotypes of $\underline{\mathbf{T}}$. $\underline{\text{harzianum}}$ tolerant to captan, captafol, chlorothalonil, iprodione, BAS 352, and DPX 4424 were developed by exposing conidia to increasing concentrations of the fungicides in V8 juice agar and selecting surviving colonies for further exposure to higher concentrations. Conidia of selected biotypes survived for at least 3 wk in fungicide suspensions (0.1% active ingredient) and for longer periods of time in soil amended with fungicides. Certain biotypes tolerant to fungicides possessed an increased ability for biological control of white rot of onion caused by Sclerotium cepivorum.

LATENT INFECTION OF RED RASPBERRIES BY TOBACCO STREAK VIRUS.
K. E. Peterson and M. K. Corbett, Dept. of Botany, University of Maryland, College Park, MD 20742 *

Indexing of Maryland grown Rubus spp. by mechanical transmission to herbaceous hosts Chenopodium quinoa, Cucumis sativus, Vigna sinensis and Nicotiana tabacum indicated that many cultivars were infected with a latent virus. An isolate from R. idaeus 'Sentry' had a dilution end point of 10^{-1} , thermal inactivation of 46-49 C and a longevity in vitro of 27 C of one day. Partially purified preparations obtained by chloroform and activated charcoal clarification and three cycles of differential centrifugation from infected Gomphrena globosa gave 3 light-scattering zones in sucrose density-gradient centrifugation. The zones contained isometric particles and had sedimentation coefficients of 57, 84, and 92 S. Material from the middle and bottom zones injected intramuscularly and intravenously in rabbits produced antiserum which reacted in gel-diffusion with precipitin lines of identity with black raspberry latent virus and with lines of relatedness to tobacco streak virus.

SOIL PLATE AND FIELD EVALUATION OF FUNGICIDES FOR CONTROL OF SCLEROTINIA BLIGHT OF PEANUT. P. M. Phipps, Tidewater Research and Continuing Education Center, VPI & SU, Suffolk, Va. 23437.

Growth of Sclerotinia minor on a soil-corn meal medium (5% corn meal) in 9 cm petri plates was measured before and after spraying the medium with fungicides. Each soil plate received 5 ml of spray containing a fungicide in sterile distilled water. In tests of over 20 fungicides, growth by S. minor on the surface of the medium was prevented only by DPX4424, BAS352, RP26019, Botran and Terraclor. Counts of sclerotia formed in the medium indicated that DPX4424 was most active in suppressing growth below the soil surface. In field evaluations, fungicides were sprayed on peanuts at or about the time of initial infection. Fungicide rates in these tests were identical in most cases to rates used in soil plates. DPX4424 at rates of 0.56 to 2.24 kg ai/ha was the only fungicide that resulted in significant (P = .05) control of Sclerotinia blight. BAS352 and RP26019 at rates of 0.56 to 2.24 kg ai/ha, Botran at 4.2 to 8.4 kg ai/ha, and Terraclor at 11.22 kg ai/ha provided only partial control.

THE ASSOCIATION OF TOBACCO RINGSPOT VIRUS WITH DECLINE OF MARYLAND GROWN GRAPEVINES. <u>S. L. Reynolds</u> and M. K. Corbett, Dept. of Botany, Univ. of Maryland, College Park, MD. 20742*

Tobacco ringspot virus (TRSV) was isolated from young leaves of declining Chardonnay grapevines in a 12 year old planting from northern Maryland. Declining vines had sparse growth, shortened internodes, poor fruit set, leaf mottle and open leaf sinuses. Partially purified preparations were obtained from infected cucumber plants by butanol extraction, polyethylene glycol precipitation and differential centrifugation. Density-gradient centrifugation of such preparations gave 3 light-scattering zones which contained isometric particles. The bottom zone was injected intramuscularly and intravenously into rabbits; the antiserum reacted in gel-diffusion with precipitin zones of identity in reciprocal tests with the N. Y. grape isolate of TRSV. The antiserum did not react with grapevine fanleaf or tomato ringspot viruses. Electron microscopy of infected tissue showed the presence of electron dense viral-like particles in cytoplasmic vesicles and plasmodesmata.

INHERITANCE OF REACTIONS TO THREE VIRUSES IN SOYBEAN. C. W. Roane, S. A. Tolin and G. R. Buss. Department of Plant Pathology and Physiology and Department of Agronomy, VPI & SU, Blacksburg, VA 24061.

'York' soybean, resistant to soybean mosaic virus (SMV) and

peanut mottle virus (PMV), was crossed with 'Lee 68', resistant to peanut stunt virus (PSV). Progenies from 10 $\rm F_1$ plants were advanced to $\rm F_3$ and data from inoculated $\rm F_3$ plants were used to determine genotypes of 605 $\rm F_2$ plants. This determination was made possible by planting equal portions of seed from each $\rm F_2$ plant in rows in 3 separate nurseries and inoculating resulting seedlings ($\rm F_3$) with PMV, SMV, or PSV followed by classification of $\rm F_3$ plants for virus reactions and pubescence color. Good fits to monogenic ratios were obtained in all families for reactions to SMV and PMV and for pubescence color. Acceptable fits to a monogenic ratio for reaction to PSV were obtained in 6/10 of the families. Independent segregation occurred for all characters except SMV and PMV reactions, which were linked with an estimated 3.7% recombination.

DETECTION OF PLANT VIRUSES USING INTACT LEAF DISKS BY THE ENZYME-LINKED IMMUNOSORBENT ASSAY. C. P. Romaine, S. R. Newhart, and D. Anzola, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

The enzyme-linked immunosorbent assay (ELISA) detected tobaccoringspot virus (TRSV) and maize dwarf mosaic virus (MDMV) when intact leaf disks were substituted for leaf extracts in the test. The procedure involved placing a disk of tissue, removed from a leaf with a paper hole-punch, in a microtiter plate well containing PBS-T-PVP (20 mM phosphate, pH 7.4, 150 mM NaCL, 0.05% Tween-20, and 2% polyvinyl pyrrolidone). In a 1-2 hr substrate reaction, the absorbance (405 nm) obtained with a single leaf disk was only 10-30% of that obtained with an extract of the same leaf. However, a more comparable absorbance (50-100%) resulted by either increasing the substrate reaction time to 24 hr or by using several leaf disks in a plate well. The test was sufficiently sensitive to detect MDMV in corn up to 4 weeks postinoculation and TRSV in latently-infected geraniums. The disk method should prove valuable for the rapid qualitative evaluation of virus infection when dealing with large plant populations.

PROTEASE INHIBITION OF THE BACTERIALLY INDUCED HYPERSENSITIVE REACTION. M. Sasser, Univ. of Delaware, Newark, DE 19711.

Bacterial and plant protein synthesis are required for the plant hypersensitive reaction (HR) to infection by incompatible bacteria. The proteins formed can be intra- or intercellular, but if the latter, then proteolytic enzymes may inhibit the HR. Screening of fourteen proteases indicated that subtilisin, trypsin or ficin added to the inoculum prevented the HR in tobacco or cowpea caused by Pseudomonas pisi or Xanthomonas vesicatoria. The enzymes did not inhibit bacterial growth in vitro nor did pretreatment of the plants with the enzymes cause a delay in the HR; suggesting that the effect is not on preformed proteins. When areas inoculated with bacteria and protease were subsequently infiltrated with protease inhibitor, the HR developed rapidly. These results suggest involvement of extracellular protein(s) in the bacterially-induced HR.

EPIDEMIOLOGY OF PYTHIUM ULTIMUM PREEMERGENCE DAMPING-OFF OF SOYBEAN IN MICHIGAN. R. L. Schlub and J. L. Lockwood, Soilborne Diseases Lab., Beltsville, MD 20705 and Dept. Botany and Plant Path., Mich. State Univ., E. Lansing, Michigan 48824.

Multiple regression using the stepwise deletion method was used to analyze data on soybean seed emergence in soil containing $\underline{P}.$ ultimum (100-300 ppg). The dependent variable, seedling emergence, was based on the mean of two 3 m rows of 100 seeds sown 6.5 cm deep, on 10 separate dates. Soil matric potential, soil temperature and rainfall were continuously monitored during the first week of each planting. The derived prediction equation [emergence = 87.6 - 5.32 (SM) - 17.76 (Rain)] had a R^2 of 0.84. The soil moisture variable (SM) was the number of days continuously after planting that the maximum matric potential at planting depth was > -0.5 bar plus the number of days in the first 3 days that it was < -3 bars. The (Rain) variable was the total rainfall in cm within the first 3 days after planting plus the number of days in the first 3 days that the soil was dry (< -3 bars).

A RAPID METHOD OF SCREENING FOR RESISTANCE TO RHIZOCTONIA ROOT ROT IN SNAP BEANS. F. M. Shao, E. S. Elliott and R. J. Young, 401 Brooks Hall, West Virginia University, Morgantown, WV 26506.

Twenty-five bean lines and some F1 progenies of crosses between Tenderette (susceptible parent) and NY 2114-12 and N203 (resistant parents) were evaluated for resistance to Rhizoctonia solani. Five to seven-day-old seedlings, germi-

nated in steamed vermiculite, were uprooted before the cotyledons emerged. Seedlings were washed and surface disinfected in 0.5% NaClO solution for five minutes and rinsed in sterile distilled water. They were then placed on sterile moist filter paper resting on a glass triangle in a 9 cm petri dish. Seedlings were then inoculated with 3 mm dia.—mycelial plugs of a virulent strain of the pathogen and were incubated at 26 C under continuous light for 48 hours. Root rot ratings were then made on a scale of 0 = healthy to 4 = dead. Results compared well with data obtained in a greenhouse test. This technique may be a useful method for rapid screening for resistance to Rhizoctonia root rot.

THE EFFECT OF PARAQUAT, SEVIN AND CAPTAN ON THE SOIL POPULATION OF SPORIDESMIUM SCLEROTIVORUM. S. M. Sherren and W. L. Klarman, Dept. of Botany, Univ. of MD, College Park, MD 20742 $\boldsymbol{\star}$

Three pesticides were tested to determine effect on Sporidesmium sclerotivorum, a mycoparasite of Sclerotinia spp. Unsteamed soil treated with pesticides at 0, 0.01, 0.1, 1.0, 10 and 100% rates recommended for Maryland soybeans was infested with Sclerotinia minor sclerotia and mycoparasite macroconidia. Pesticide effects were determined by examining sclerotia at weekly intervals for survival and colonization. Paraquat had no effect. Sevin at 10% and captan at 1% temporarily suppressed mycoparasite activity and captan at 100% completely prevented colonization. Where significant inhibition was noted, pesticide effect on spore germination was tested. Millipore filters on which mycoparasite macroconidia had been collected were placed on soil containing pesticide and sclerotia. Germination was completely prevented by captan at 1X and temporarily suppressed at 0.1%. Germination was completely prevented by the pesticides in combination at 1X, temporarily suppressed at 0.1% and stimulated at 0.01%.

GROWTH IMPACT OF A PERIODIC SOURCE OF INDUSTRIAL AIR POLLUTANT EMISSION ON EASTERN WHITE PINE: AN UPDATE AND RE-EVALUATION OF PREVIOUS REPORTS. J. M. Skelly and Yaw-Shing Yang. Dept. of Pl. Path. & Phys., VPI&SU, Blacksburg, VA 24061.

A re-examination of the radial increment growth of 50 eastern white pines growing near a periodic source of high $\rm SO_2$ and $\rm NO_2$ was completed as a follow up to the previously reported growth period of 1935 to 1971. During the period 1971-1975, pollution concentrations decreased from previous highs experienced in 1964-1970 but then increased significantly during the 1975-1978 period. Multiple linear regression analysis was carried out using annual increment growth as the dependent variable and annual seasonal rainfall, total annual rainfall, tree age, and annual coal consumption (as a surrogate for pollution) as the independent variables. A significant (p=0.05) negative relationship was found between radial increment growth and production history. Thus, the previously reported relationship of growth response to changes in industrial pollution emission has continued to function.

INTERACTION OF THE HERBICIDES ALACHLOR AND LINURON WITH FUSARIUM OXYSPORUM INFECTION OF SOYBEANS. J. P. Smith and R. B. Carroll, Univ. of Delaware, Newark, De. 19711.

In 1979 Essex and Williams soybeans were planted no-till in a split plot design in naturally infested field soil and in soil artificially infested with Fusarium oxysporum. Plots were then sprayed preemergence with five combinations of alachlor plus linuron at three rates. Plant and rhizosphere samples were collected 11 times from planting to harvest. Plants were rated for disease incidence, and isolations were made from root and basal stem sections. Soil samples were evaluated for Fusarium by dilution plate method. Root disease ratings and root isolations showed there was no effect from any of the combinations of alachlor plus linuron on F. oxysporum infection. When the same herbicide treatments were tested on soybeans grown in the greenhouse, results showed an effect on predisposition to infection by F. oxysporum.

OBSERVATIONS OF THE EXTENSIVE 1979 TOBACCO BLUE MOLD EPIDEMIC, Harvey W. Spurr, Jr., North Carolina State Univ. and USDA-AR Tobacco Research Lab., Oxford, NC 27565, and F. A. Todd, N. C. State University, Raleigh, NC 27650.

The worst North American tobacco blue mold field epidemic occurred during 1979 resulting in a 252 million dollar loss to growers. The epidemic spread generally from south to north favored by ample inoculum and unseasonably cool, wet weather. In North Carolina, burley losses were 40% with many fields

completely destroyed and flue-cured tobacco losses were 4%. Systemic infections were abundant in burley which resulted in severe stunting. Conidia produced by the fungal pathogen *Peronospora tabacina* were typical. Oospores were abundant in leaves and systemically infected stems of burley. They were less abundant in flue-cured tobacco especially when local leaf infections were arrested.

A DEFINED AGAR MEDIUM FOR ISOLATING <u>VERTICICLADIELLA</u> <u>PROCERA</u> FROM NATURALLY INFESTED SOILS. <u>I. S. Swai</u> and D. F. Hindal, 401 Brooks Hall, West Virginia University, Morgantown, WV 26506.

Verticicladiella procera, which causes white pine root decline (WPRD), is difficult to isolate from naturally infested soils. A defined agar medium was developed which is suitable for isolating this fungus from such soils. The medium contains: 2.0 g glucose, 0.2 mg Fe++, 0.2 mg Zn++, 0.1 mg Mn++, 50 mg chlorotetracycline, 50 mg streptomycin sulfate, 50 mg cycloheximide, 15 g Difco Bacto agar and 990 ml distilled water. The pH of this medium is adjusted so that it is in a range from 4.5 to 5.0 before autoclaving. The antibiotics, dissolved in 10 ml sterile distilled water, are added to the cooled medium before dispensing into Petri plates. For isolation of V. procera, a sample of soil is collected from the rhizosphere of white pines with WPRD. After soil dilutions are made, a 1 ml aliquot from the 10-2 and 10-3 dilutions is spread on the medium and following 7 to 15 days incubation at 20 C, colonies of V. procera can be distinguished.

ALKANE CONTENT OF THE EPICUTICULAR WAX OF TWO PINUS STROBUS L. CLONES DIFFERING IN SENSITIVITY TO OZONE. Judy L. Trimble and David M. Orcutt, Department of Plant Pathology and Physiology, Virginia Tech, Blacksburg, VA 24061.*

The epicuticular waxes of 3-yr-old grafted eastern white pines from two clones, one sensitive and one tolerant to 03, were selected for analysis of alkane content. Fascicles were harvested 10 wk after exposure to 0 and 0.30 ppm 03 6h/day for 7 consecutive days. Fascicles were cut 5.0 cm from the tips and weighed. Wax was extracted by dipping fascicles in chloroform for 30 s. The extract was filtered, evaporated to dryness, weighed, and redissolved in hexane. Alkane content was determined by gas liquid chromatography. Hentriacontane (C31) and tritriacontane (C33) were the predominant alkanes. Alkane concentration was greater for the tolerant clone on both a fascicle surface area and fascicle weight basis regardless of exposure. In both clones, exposure to 03 resulted in higher alkane concentration. It was concluded that the alkane content of epicuticular wax is related to 03 sensitivity of eastern white pine.

SURVIVAL OF PAECILOMYCES BUXI IN NURSERY SOIL AND IN DECOMPOSING ENGLISH BOXWOOD ROOTS. M. A. Vizvary, W. H. Wills & R. C. Lambe, Dept. of Plant Path. & Phys., VPI & SU, Blacksburg, VA 24061. First, the survival of Paecilomyces buxi (Pb) in nursery soil as spores was studied. The initial population of 10,000 conidia/g soil of 3 Pb isolates was reduced by 50% after 2 mo in soil incubated at 22-23°C and cycled between -0.33 and -15 bars water potential. Populations of all isolates were reduced 70-80% between 6-8 mo and nearly 100% at 24 mo. Second, the survival of Pb was studied in roots excised from declining English boxwoods and buried in nursery soil. The initial isolation frequency from root segments was 40%. This increased to 78.5% at 6 mo in spite of severe root decay. By 30 mo, only 14% of the root segments contained Pb. Third, in <u>situ</u> recolonization of nursery soil was studied after fumigation with Vapam® and Vorlex®. Both chemicals reduced Pb to <10 conidia/g soil. Seven mo later, populations increased to 10,200 and 7,175 conidia/g soil and diminished to 3,050 and 1,125 conidia/g soil at 25 mo in the Vapam® and Vorlex® treated plots, respectively. These experiments show that Pb can survive in soil a min of 24 mo without host substrate and a min of 30 mo with substrate.

VARIATION IN RESPONSE AMONG 33 WIND-POLLINATED FAMILIES OF PINUS TAEDA L. Marsha Ward and L. W. Kress. Department of Plant Pathology and Physiology, VPI & SU, Blacksburg, VA 24061

Five week-old seedlings representing wind-pollinated families of $\underline{\text{Pinus}}$ taeda L. were exposed to 40 pphm ozone for 8 h at approximately 32 C and 74% RH. Response was evaluated 7 days after exposure for 4 symptom types: % needle surface area with chlorotic mottle (Y1) and with necrosis (Y2), and % needles with chlorotic mottle (Y3) and with necrosis (Y4). Analysis of data revealed 4 significantly different sensitivity classes in the Y1, Y2 and Y3 symptom types and 3 classes in Y4.

Approximately 20% of the families partitioned into the most sensitive class and 35% into the least sensitive class. Most families partitioned into the same relative sensitivity class in the 4 symptom types. For example, if a family showed low sensitivity for a symptom type, then it showed low sensitivity for the other 3 symptom types. Environmental variance was minimized in the execution of the experiment and differences between families are considered to be genetic in origin.

EFFECTS OF ROOT-KNOT NEMATODES ON PHYTOPHTHORA ROOT ROT OF AL-FALFA. R.E. Welty, USDA, SEA, AR and K.R. Barker, Dept. of Plant Pathology, N.C. State Univ., Raleigh 27650; and D.L. Lindsey, Dept. of Botany and Ent., New Mexico State Univ., Las Cruces 88003.

Phytopthora megasperma f. sp. medicaginis causes a serious root rot of alfalfa (Medicago sativa) and generally limits yield and persistence. Root-knot nematodes parasitizing alfalfa include Meloidogyne hapla and M. incognita. This study was done to determine if root-rot was influenced by the presence of rootknot nematodes. Root rot and root necrosis was more severe in cultivars resistant (Agate and Apollo) or susceptible (Saranac) to Phytophthora root rot when inoculated with the fungus and M. hapla or M. incognita. M. hapla increased the incidence of root rot more than M. incognita, but both nematode species suppressed top and root growth, increased chlorosis, and reduced vigor based on general plant appearance. Inoculation with \underline{P} . megasperma suppressed reproduction of both nematodes.

HISTOLOGY OF ILEX CRENATA ENDOMYCORRHIZAE. Robert L. Wick and Laurence D. Moore. Dept. of Plant Pathology and Physiology, VPI & SU, Blacksburg, VA. 24061 \bigstar

A histological study of holly (Ilex crenata) roots was undertaken to determine the nature of the mycorrhizae. Rooted cuttings were inoculated with spores of the endomycorrhizal fungi (mostly Glomus and Gigaspora spp.) recovered from soil in which landscape holly had been grown. Six months after inoculation, 82% of 1 cm long feeder roots were found to be mycorrhizal. Cortical cells of the root including the hypomycorrhizal. Cortical cells of the root including the hypodermis were colonized while the vascular system and endodermis were not. The fungi moved intracellularly through the root, forming many arbuscules and few vesicles. Average diameter of hyphae, arbuscules and vesicles were 4.41 μm , 17.5 x 23.2 μm and 20.38 x 25.53 μm , respectively. Nuclei of colonized cells were 20% larger and had different staining properties than those of uncolonized cells. No other anatomical changes of the root were observed. root were observed.

GROWTH AND SPORULATION OF VIRULENT AND HYPOVIRULENT ISOLATES OF ENDOTHIA PARASITICA. R. L. Willey and W. L. MacDonald, 401 Brooks Hall, West Virginia University, Morgantown, WV 26506.

Two virulent and seven hypovirulent isolates of Endothia parasitica were inoculated in American chestnut trees to compare their growth and sporulation. Canker area comparisons showed that virulent isolates invaded host tissue extensively while colonization by hypovirulent isolates was highly variable. Stromata presence on cankers indicated the virulent isolates and all but one hypovirulent isolate sporulated asexually. However, stromata production did not correlate directly to the amount of host tissue invaded. Sexual sporulation was present only on cankers caused by virulent isolates. Laboratory crosses between sexually compatible virulent and hypovirulent isolates resulted in perithecial production by the virulent and two of the hypovirulent isolates.

MORTIERELLA ANTAGONISM TO OOMYCETES. W. H. Wills and R. C. Lambe. Department of Plant Pathology and Physiology, VPI & SU, Blacksburg, VA 24061.

Mortierella spp. isolated from azalea and boxwood roots were inhibitory to growth of Phytophthora cinnamomi (PC) and P. parasitica (PP), pathogens of azalea and boxwood respectively, in paired culture on corn meal agar. Both isolates inhibited growth of several other isolates of Phytophthora, Pythium ultimum, P. debaryanum, P. splendens, P. myriotylum and P. aphanidermatum. Neither isolate inhibited several non-oomycetous fungi, including Rhizoctonia solani and several Fusaria. Rooted cuttings of azalea and boxwood were inoculated with PC and PP respectively, and challenge inoculated with azalea and boxwood isolates of Mortierella, respectively. Challenge inoculation one week prior to inoculation with the pathogen resulted in a high level of protection of azalea from PC, as shown by low root

rot ratings and low reisolation of PC. Protection of boxwood from PP was not as well defined as that of azalea from PC, but a potential for biological control by means of Mortierella isolates was demonstrated in both systems.

A COMPARISON OF THE ACTION OF THREE SYSTEMIC FUNGICIDES ON PYRICULARIA ORYZAE. P. M. Wolkow, C. P. Woloshuk, & H. D. Sisler. Department of Botany, University of Maryland, College Park, Maryland 20742.

Tricyclazole (EL 291), 1,2,5,6-tetrahydro-4H-pyrol-lo-[3,2,1-i,j]-quinolin-4-one (CGA 49104), and 4,5dihydro-4-methyltetrazolo (1,5-a) quinazolin-5-one (pp389) specifically control rice blast disease caused by Pyricularia oryzae. None of the compounds are toxic to growth of P. oryzae at 10 $\mu g/ml$, but all selectively block melanin biosynthesis at this concentration. The concentrations of tricyclazole, CGA 49104 and pp389 which completely block melanization are respectively 0.1, 1 and 10 µg/ml. Inhibition of melanin biosynthesis results in excretion of 2-hydroxyjuglone and flaviolin. These studies indicate that the three compounds have a similar or identical mechanism in P. oryzae. Control of rice blast disease probably results from a change in the host parasite interaction rather than direct fungitoxicity.

EFFECTS OF OZONE, SULFUR DIOXIDE AND NITROGEN OXIDES ON THE GROWTH OF EASTERN WHITE PINE. $\underline{Yaw-Shing\ Yang}$, Dept. of Plant Pathology and Physiology, Virginia Tech, Blacksburg, VA 24061.*

Eight clones of eastern white pine (Pinus strobus L.) with predetermined sensitivity to air pollution were fumigated with $\boldsymbol{5}$ pphm and 10 pphm ozone (03), sulfur dioxide (SO2), and nitrogen oxides (NO_X) singly, and in combination for 4h/day for 35 consecutive days. During the fumigation period, needle length, visible symptoms, and dry wt. were evaluated weekly. The carbon dioxide (CO2) exchange rate was measured during the 10 pphm 0_3 and 10 pphm 80_2 fumigation for 4h/day for 7 consecutive days. Needle length was not significantly different among different clones and treatments, whereas, there was a correlation between needle dry wt. (mg/mm of fascicle) and air pollution injury. Ten pphm O_3 and 10 pphm SO_2 slightly decreased CO_2 exchange rate during the first lh of fumigation. Recovery phenomenon occurred after the end of each day's 4h fumigation. There was a trend of continuous decrease in relative CO2 exchange rates over the 7 day period, however.

EFFECT OF APPLICATION INTERVAL ON CONTROL OF APPLE POWDERY MILDEW BY DINOCAP-BENOMYL COMBINATIONS. K. S. Yoder, Dept. of Plant Pathology and Physiology, VPI & SU Fruit Research Lab, Winchester, VA 22601.

Dinocap (Dikar 76.7% WP) and benomyl (Benlate 50% WP) were applied to mature Jonathan apple trees as tank-mixed dilute sprays according to the following application interval and rate combinations: 14 day - Dikar 2 1b plus Benlate 2 oz formulated material per 100 gal; 7 day - 1/2 of the above rates; 3-4 day - 1/4 of above rates. Percent leaves with powdery mildew infection following the season-long treatment schedules for the 14, 7, and 3-4 day application intervals was, respectively, 15, 5 and 3. Percent fruit infected for these application intervals was 19, 12, and 6. On untreated trees 99% of the leaves and 48% of the fruit were infected with mildew. Apart from its reduction of mildew infection, reducing the application interval and rate did not significantly affect the finish of the fruit.

EXPRESSION OF MULTIGENIC RESISTANCE TO PHYTOPHTHORA INFESTANS IN TUBERS OF SOLANUM TUBEROSUM. R. J. Young and S. K. Bhatia, 401 Brooks Hall, West Virginia University, Morgantown, WV 26506.

Multigenic resistance to <u>Phytophthora infestans</u> was studied in tubers of <u>Solanum tuberosum</u> with specific reference to selected stages of the disease cycle. Tuber slices were inoculated with race 1,2,3,4 of the pathogen and incubated for 10 days at 19 C. A virulent isolate of the pathogen was recovered from all inoculated tissues. The pathogen developed at a slower rate on resistant tubers, and tissue discoloration occurred more rapidly. Invasion of the tissue, mycelial growth and sporulation were also restricted in resistant tubers. The expression of multigenic resistance in potato tubers follows a similar pattern of resistance expressed in potato leaves.