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ABSTRACTS

A3

THE INTERRELATIONSHIPS OF HOST- AND PARASITE-GERMPLASM COLLECTIONS IN THE STUDY OF CEREAL RUSTS. L. E. Browder, USDA-ARS, Dept. of Pl. Path., KSU, Manhattan, KS 66506.

New knowledge comes only from experiments that test hypotheses and that use constants and variables. Specificity of host-(H) genotype, parasite-(P) genotype, and environment has been demonstrated to be a basis of resistance (R). A plausible premise for further study is that definitive factors in H, P, and environment interact to cause resistance. R genes cannot be detected without genes for avirulence (A); A genes cannot be detected without R genes; both are manifested as one phenotype. Many gene pairs are already known that relate to R. Progress in detecting new gene pairs depends on having "knowns" in both H and P to document known gene pairs. Using data from comparisons of these knowns with unknowns, testable hypotheses can be formulated concerning the unknowns, using logic. The hypotheses take the form that an unknown is alike or different from the knowns. Complexity arises due to the large numbers of gene pairs involved and gene combinations in both organisms. Thus, collections of both H and P germplasm are vital to any progress.

A4

THE USE OF BEAN RUST AND WHEAT STEM RUST COLLECTIONS IN CHARACTERIZING PATHOGEN DIVERSITY. J.V. Groth and A.P. Roelfs, University of Minnesota, St. Paul, MN 55108.

Large collections of rust fungi are maintained at Minnesota either under liquid nitrogen or, more commonly recently, in ultra-low temperature freezers. Uredospore accessions fall into three different categories: 1) heterogeneous field collections 2) single-uredial isolates and 3) progeny of crosses and selfs of isolates. Isolates, obtained from collections, have traditionally been used in race surveys, but mass collections are being used more and more with bean rust to characterize diversity for virulence. Isolates and progeny are both proving useful in explaining isozyme and virulence diversity. Collections of bean rust teliospores are also maintained for genetic studies. Field collections from 1975 have proved useful in understanding the history of important virulence genes. As genetic information about isolates becomes more extensive, their value and potential uses increase.

A5

USE OF DIFFERENTIAL CULTIVARS OF LACTUCA SATIVA TO DETECT, ISOLATE, CULTURE, AND STUDY DIVERSE PATHOGENIC GENOTYPES OF BREMIA LACTUCAE IN NEW YORK. J. W. Lorbeer, D. P. LoParco, and J. E. Yuen. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Growing cultivars of Lactuca sativa with differential resistance factors to Bremia lactucae in commercial lettuce fields enabled detection and isolation of collections of the pathogen having five virulence factors (V-5,6,7,8,10). When the collections were tested on 5-day-old cotyledons of 21 differential cultivars, five other factors (V-1,2,4,9,11) were detected. Only V-2 and V-7 were detected when collections from commercial cultivars (Ithaca and Minetto) were similarly tested. Specific genotypes were maintained on frozen seedlings or cotyledon discs of cultivar Ithaca (no resistance factors). Single-sporangium isolates with known virulence factors and opposite mating type were dually inoculated onto seedlings of cultivar Ithaca to produce abundant oospores for seedling inoculation experiments and future genetic studies.

A6

PATHOGEN AND HOST GERMPLASM COLLECTIONS FOR ASSESSING WEED AND CROP VULNERABILITY TO PATHOGENS. M. R. Bonde and W. M. Dowler. USDA-ARS Plant Disease Research Laboratory, Ft. Detrick, Bldg. 1301, Frederick, Maryland 21701.

An initial step in determining the potential threat of foreign pathogens to U.S. agriculture or usefulness of natural enemies in weed control is to determine the reaction of representative pathogens on selected hosts. Several pathogens have been evaluated for potential threat at PDRL, such as Phakopsora pachyrhizi (causal agent of soybean rust), and Peronosclerospora spp. (causal agents of downy mildews of corn). The necessity to have known plant germplasm collections for testing is obvious, but it is equally important to have reliable, documented pathogen collections. Evaluation of exotic pathogens as potential biocontrol agents for weeds requires pathogen collections as well as plant germplasm collections. Broad host range studies are required, including related endangered species. These are examples of the interdependence of pathogen and host germplasm collections.

A7

THE INTERDEPENDENCE OF SPHAEROTHECA PANNOSA AND ITS VARIOUS HOSTS. D. L. Coyier, USDA-ARS, Horticultural Crops Res. Lab., 3420 N. W. Orchard Ave., Corvallis, OR 97330.

The relationship between certain powdery mildews and their hosts has not been clearly defined. Sphaerotheca pannosa reportedly occurs on Lycium halimifolium, Photinia serrulata, Prunus persica and cultivated Rosa spp. Although the fungus has been separated into vars. persicae and rosae, respectively, on the latter two hosts, it has been shown recently that races of S. pannosa var. rosae exhibit a wide range of pathogenicity among various Rosa spp. and cultivars. Further, some races pathogenic on Rosa spp. infect Prunus persica during very limited periods of host development. Similar limitations in pathogenicity occur in the S. pannosa var. rosae/Photinia serrulata interaction. While young leaves in the spring are often highly susceptible to the fungus, new growth later in the season may be resistant. Recent unpublished work suggested that powdery mildews collected from Epilobium angustifolium, Lactuca, sp., and Taraxacum officinale (tentatively identified as S. macularis), were capable of infecting cultivated Rosa sp., but none of five races from Rosa sp. infected any of the three S. macularis hosts.

A8

INTERDEPENDENCE OF COLLECTIONS OF IPOMOEA BATATAS AND COLLECTIONS OF SOIL-BORNE PATHOGENS IN DEVELOPING RESISTANCE IN SWEET POTATO AGAINST ROOT, STEM AND VASCULAR DISEASES. P. D. Dukes and Alfred Jones, USDA, ARS, Vegetable Laboratory, Charleston, SC 29407

Developing multiple disease resistance is a major goal of our breeding program (BP). The BP depends upon a wide gene base collection of Ipomoea spp. along with a diversified collection of major pathogens (Fusarium oxysporum f. sp. batatas, Meloidogyne incognita, etc.). The hexaploid sweet potato (SP) is extremely heterozygous. Therefore, selections are vegetatively propagated. The maintenance of stable genotypes as standards for reference in disease evaluation is essential. The most virulent and dissimilar isolates of each pathogen are used in the BP. Inoculum from each isolate is produced separately and composite inoculum is prepared that represents equal no. of each isolate. Standardized inocula are mixed and seedlings inoculated. The survivors and/or the best seedlings are retained. These methods have enabled us to achieve higher levels of multiple disease resistance in SP.

A9

THREE RESISTANCE SYSTEMS TO BACTERIAL SPOT OF PEPPER IN A PLANT INTRODUCTION LINE. A. M. Hibberd, R. E. Stall, and M. J. Bassett. Vegetable Crops Dept. and Plant Pathology Dept., Univ. of Florida, Gainesville, FL 32611.

The development of stable resistance to bacterial spot of pepper may follow along the lines described for bacterial blight of cotton (L. A. Brinkerhoff et al., 1984. Plant Disease 68:168). The basis for stable resistance in pepper may be plants of PI 271322, which possess resistance to race 1, to race 2, and to races 1, 2, and 3 of Xanthomonas campestris pv. vesicatoria. Resistance to race 1 and to race 2 can be detected after injections of high concentrations of inocula (10^8 cells ml^{-1}) into leaves. Collapse of tissue occurs within 24 hours with race 1 and within 12 hours with race 2. The resistance to races 1, 2, and 3 can be identified after injections of low concentrations of inocula (10^3 cells ml^{-1}) into leaves. Lesions are usually fewer and always smaller than those that develop in susceptible plants. The three resistances segregate independently in crosses with susceptible pepper.

A10

CONTRIBUTION OF INERTIAL IMPACTION TO SPORE DISPERSAL GRADIENTS. Donald E. Aylor and Francis J. Ferrandino, The Connecticut Agric. Expt. Station, Box 1106, New Haven, CT 06504.

Dispersal gradients of airborne spores in crops depends to a large extent on the efficiency with which spores are deposited on plants, mainly by settling under gravity and inertial impaction. To calculate the net deposition of spores by inertial impaction on stems and leaves requires knowledge of spore sticking efficiency. Spore sticking efficiency was evaluated by releasing particles in a mature wheat crop. These particles were trapped with sticky cylinders distributed in the crop and were sampled directly on the stems and foliage. Efficiency of deposit on leaves was essentially that theoretically expected by gravitational settling. Deposit on stems, however, was only about 10% of that theoretically expected by inertial impaction and decreased with increasing wind speed, contrary to theory. This reduced trapping efficiency leads to dispersal gradients that are considerably less steep than predicted by current models.

A11

A GENERAL MODEL FOR DISEASE PROGRESS IN CHANGING HOST GROWTH WITH A SUB-MODEL FOR VARIABLE LATENCY. R. D. Berger and J. W. Jones, Depts. of Plant Pathology and Ag. Engineering, Univ. of Florida, Gainesville 32611.

A disease progress model was derived to aid interpretation of previously perplexing epidemiological responses. The model consisted of an infection equation combined with a host growth function. Variants of common growth models (logistic, Gompertz, exponential, linear, etc.) were used as the basic model equations. Variable latency was handled in a sub-model by a distributed delay function. The rapid increase in disease after epidemic interruption or from low initial disease could be explained solely by the amount and proportion of healthy tissue rather than by a change in any pathogenic variable. Because the model has a feedback function of disease intensity on host growth, it can be used to quantify the impact of disease stress on crop yield and to determine loss thresholds in disease management systems.

A12

Improvement in modeling disease progress. E. W. Park and S. M. Lim. Department of Plant Pathology, University of Illinois at Urbana-Champaign, Urbana, IL. 61801.

Growth functions that have been commonly used to depict plant disease progress are restricted by the asymptote value which is fixed at 1. Disease progress curves with asymptote values of less than 1 cannot be accurately described by growth models with the asymptote value of 1. Vanderplank's apparent infection rate, which is the rate parameter of the logistic function with the asymptote value of 1, can not only underestimate the rate of increase in disease severity but also can change the rank of the rates of epidemics to be compared. The weighted mean absolute and the weighted mean relative growth rates of the Richards function can be used as the absolute (Ra) and the relative (Rr) infection rates, respectively, for describing and comparing epidemics with different shapes of development and different asymptote values. The Ra and Rr values are biologically meaningful and universally comparable parameters for disease progress models.

A13

A MODIFIED VERSION OF GREGORY'S DISPERSAL MODEL FOR USE IN COMPUTER SIMULATION OF EPIDEMIC DEVELOPMENT. C. C. Mundt and K. J. Leonard, Dept. of Plant Pathology, North Carolina State Univ., Raleigh, NC 27695-7616.

Gregory (Annu. Rev. Phytopathol. 6:189-212) proposed a dispersal model $y = ax^{-b}$ in which y is the number of spores or lesions at x distance from the source, a is a constant that is proportional to the source strength, and b is a measure of the steepness of the gradient. This model does not provide for a finite number of spores or lesions at the source (i.e. $x = 0$) and, therefore, has not been used in computer simulations of spatial progression of plant disease. We modified Gregory's model to $y = a(x+c)^{-b}$ in which c is a constant derived by iterative fitting of dispersal data to the model. The modified model provides a finite y intercept and adequately describes disease gradients from known sources of rust spores. We have successfully incorporated the modified dispersal model into EPIMUL for simulation of the temporal and spatial increase of disease in genetically diverse host populations.

A14

A SIMPLE MODEL FOR PREDICTING INFECTION OF VEGETATIVE SOYBEAN TISSUE BY PHOMOPSIS SP.. John Rupe and Richard S. Ferriss, Dept. of Plant Path., University of Kentucky, Lexington, Ky 40546.

Epidemics of pod and stem blight of soybean, caused by Phomopsis sp., can be divided into two distinct phases: 1) initiation of infections of green tissue, and 2) growth of the fungus into the seeds from infected pods. A simple model was developed relating infection to accumulated hours of leaf wetness accompanying rain multiplied by a function of the mean hourly temperature for each leaf wetness period. Data for model development was obtained from greenhouse, growth chamber and laboratory experiments on infection by and germination and growth of the fungus. The model assumes a uniform supply of splash-dispersed spores which would be present in a field cropped to soybeans for several successive seasons. Infections of pods and seedlings under field conditions was significantly correlated to the model's predictions. The model provides a core on which to develop more complicated models and may be useful in predicting the necessity of fungicide sprays for controlling seed infection.

A15

DISEASE PROGRESSION AS A FUNCTION OF PLANT GROWTH. N. Lalancette, Jr., and K. D. Hickey. The Pennsylvania State University Fruit Research Lab, Biglerville, PA 17307-0309.

The growth of plants and of populations of plant pathogens can be described by various mathematical models. Usually these models describe the rate of change of plant growth or of the pathogen population relative to time. Since disease is dependent on the simultaneous occurrence of both pathogen and suscept, an alternative approach is to describe the change of the pathogen population relative to plant growth. Several models were developed from the logistic, Gompertz, monomolecular, and power functions for describing disease as a function of plant growth. 'Mixed' models were also derived for situations in which pathogen and plant dynamics differ, e.g. monomolecular plant growth and logistic pathogen growth. Furthermore, alterations can be made for non-synchronous dynamics, as when plant growth occurs prior of onset of an epidemic. The assumptions, applications, and fitting of the models via nonlinear regression to several powdery mildew epidemics will be discussed.

A16

AN APPLE POWDERY MILDEW MODEL BASED ON PLANT GROWTH, PRIMARY INOCULUM, AND FUNGICIDE CONCENTRATION. N. Lalancette, Jr., and K. D. Hickey, The Pennsylvania State University Fruit Research Lab, Biglerville, PA 17307-0309.

In 1981, 1982, and 1983 Rome Beauty apple trees with different amounts of primary mildew were sprayed with various concentrations of the fungicide bitertanol. The total number of leaves and the number of leaves infected with secondary mildew per shoot were determined periodically throughout each growing season. When the frequency of infected leaves was modeled as a function of leaf production, the initial amount of secondary mildew was related to the level of primary mildew whereas the rate of the epidemic was related to fungicide concentration. Given estimates of the amount of primary mildew and the amount of plant growth, the concentration of fungicide necessary to manage secondary mildew at a desired level can be determined.

A17

EFFECTS OF FUNGICIDE TREATMENTS ON THE YIELD OF VARIOUS CULTIVARS OF WHEAT (Triticum aestivum L.). F. E. Wright, Agricultural Research, P.O. Box 2340, State University, AR 72467.

Six fungicide treatments (Benomyl, Mancozeb, Benomyl + Mancozeb, Triadimefon, Triadimefon + Mancozeb, and Tilt (CGA 64250) were applied to eight cultivars (Rosen, Nelson, Pioneer 2550, Abe, Coker 747, Doublecrop, Caldwell and Hunter) of wheat. Treatments were applied twice, once at growth stage 10.0 with a second application at stage 10.5.2 (Feek's Scale). Disease control varied with the materials used. Yield was the parameter used to measure performance of the materials. Yields varied within cultivar and treatment. The highest yield produced was 5242 kg/ha with Tilt (CGA 64250) and the cultivar Pioneer 2550, 1727 kg/ha greater than the control. The lowest yield was found with the Triadimefon treatment and Abe cultivar which was 109 kg/ha better than the control. The mean yield by cultivar compared to the control was +934, +175, +1001, +120, -517, +1277, +260 kg/ha for Rosen, Nelson, Pioneer 2550, Abe, Coker 747, Doublecrop, Caldwell, and Hunter, respectively.

A18

MICROFLORA ASSOCIATED WITH DISCOLORED BARLEY KERNELS. M. R. Miles and R. D. Wilcoxson. Department of Plant Pathology, University of Minnesota, St. Paul, Minnesota 55108.

Nine cultivars differing for resistance to kernel discoloration were grown at Crookston, Morris, Rosemount, and St. Paul, MN in 1982. Kernels were evaluated for discoloration. Chevron and CI 9539 scored 1-2 at each location; Cebada Capa and Karl scored 4-5; Bumper, Glen, Morex, Robust and Larker were 2-4. A score of 1 indicated light and 5 severe discoloration. A random sample of 150 kernels/cultivar from each location were surface sterilized then incubated 5-7 days on potato dextrose agar under fluorescent lights at 25 C. Fungi present on each kernel were noted. Isolated from each cultivar at each location were Bipolaris sorokiniana (3-15% of the kernels), Alternaria alternata (87-97%), Fusarium culmorum and F. graminearum (12-24%), Cladosporium herbarium (14-35%), and bacteria and yeasts (2-4%). Kernel discoloration and fungi present varied significantly (P = .05) with cultivar.

A19

NEW HOSTS OF FUNGI FOUND ON SMALL GRAINS, CORN AND PERENNIAL FORAGE GRASSES. Martha K. Roane and Curtis W. Roane. Dept. of Plant Pathol., Physiol. and Weed Sci., VPI&SU, Blacksburg, VA 24061.

Several fungi previously reported on small grains, corn and perennial forage grasses were found on new hosts. They are Acremoniella verrucosa on Cynodon dactylon and Festuca elatior var. arundinacea; Ascochyta graminea on Arrhenatherum elatius; A. hordei on Triticum aestivum; A. sorghi on Agrostis perennans; Cladosporum herbarum on Agrostis hiemalis; Cochliobolus carbonum on Muhlenbergia schreberi; Dilophospora alopecuri on Dactylis glomerata; Dinemasporium graminum on Zea mays; Fusarium acuminatum on Dactylis glomerata; Phyllosticta sorghina on Agrostis hiemalis and Septoria nodorum on Muhlenbergia schreberi. These hosts are new for Virginia.

A20

PERONOSCLEROSPORA GLOBOSA, A NEW DOWNY MILDEW OF GRAMINEAE, ATTACKING CUPGRASS IN TEXAS. Q.B. Kubicek and R.G. Kenneth, Dept. of Plant Pathology & Microbiology, Texas A&M Univ., College Station, TX 77843.

A downy mildew was found near Orange Grove, Texas, in 1982 and 1983, on wild cupgrass, Eriochloa contracta. In 1979, others had found in New South Wales a downy mildew, designated as Peronosclerospora sp., on wild Eriochloa crebra. The Texas pathogen, with conidia rather than sporangia, is a Peronosclerospora and by inoculation infected E. contracta, E. crebra and E. nubica, but not sorghum and corn. The pathogen induced systemic infection, with mild chlorosis and profuse sporulation. Conidia were globose, occasionally subglobose, only 12-18 µm diam. Oospores, 28-36 µm diam., within adherent oogonia, 40-54 µm, were in lower leaves of greenhouse-inoculated plants; they presumably enable overwintering in Texas. This is the first Peronosclerospora proven to attack plants of the tribe Paniceae, and the second one (after P. sorghi) found in the New World. Because of morphological and hostal differences, the Texas (and perhaps the Australian) isolate is a new species, soon to be validly named Peronosclerospora globosa.

A21

ATYPICAL SYMPTOMS IN RYE CAUSED BY PSEUDOMONAS SYRINGAE PV. CORONAFACIENS. C. W. ROANE. Department of Plant Pathology, Physiology & Weed Science, VPI&SU, Blacksburg, VA 24061.

Typically, Pseudomonas syringae pv. coronafaciens (Elliott) Young et al. (Psc) causes halo blight of oats and rye. In southwestern Virginia, rye is used extensively as a winter cover crop and mulch for no-till corn production. Several rye stands died out during the winter of 1978-79. On two such farms in Montgomery County, Psc was associated with symptoms similar to those observed in oats in 1960 (Plant Dis. Rep. 44:696). Rye plants remaining in the fields were slightly to severely stunted. Blades on some plants had chlorotic and necrotic lesions although halo blight was not observed. On some other plants, entire blades were yellowed. Leaves of stunted plants were often characterized by chlorotic veins. Veinal chlorosis was probably caused by a toxin translocated from colonized crowns. Bacteria isolated from such plants produced halo blight when inoculated to oats. No additional occurrences of this problem have been observed since 1979.

A22

ERGOT ALKALOIDS IN TALL FESCUE INFECTED WITH SPHACELIA TYPHINA. P. C. Lyons and C. W. Bacon. Department of Plant Pathology, University of Georgia, Athens, GA 30603 and USDA-ARS, R. B. Russell Agricultural Research Center, Athens, GA 30613.

Ergot alkaloids were isolated from greenhouse and field-grown tall fescue grass infected with the fungal endophyte Sphacelia typhina. The field grown grass was toxic to cattle. The three major alkaloids were identified as ergovaline, ergovalinine and chanoclavine which also were produced by the fungus in vitro. Alkaloids were isolated from leaf sheath in which fungal growth was extensive and leaf blade into which hyphae did not penetrate. The presence of these compounds in the blade suggested that they were translocated from the sheath where they were presumably synthesized by the fungus. Ergot alkaloids were quantitated in greenhouse plants grown under low and high rates of NH_4^+ and NO_3 and preliminary data suggested that high levels of nitrogen resulted in increased alkaloid accumulation.

A23

HEAT AND DROUGHT STRESS AND AFLATOXIN PRODUCTION IN PREHARVEST MAIZE. J. R. Wallin, 312 Curtis Hall, Univ. of Missouri, Columbia, 65211.

Samples of commercial maize hybrids commonly grown at eight locations in Missouri 1982 and 1983 were analyzed for aflatoxin B_1. Weekly temperature and rainfall data were obtained to relate to toxin occurrence. Only a trace of toxin was found at three locations in 1982. Weekly average temperature maxima reached 35 C (95 F) one week at one location, the remainder were less than 32 C (90 F). Coincident weekly total rainfall for each location was much above normal from August 1 to 29. In 1983, aflatoxin was found in maize at all locations from 20-100 ppb. Hybrids differ in content. Weekly temperature maxima averages were generally above 35 C from July 18 to August 29. Coincident precipitation was much below normal. These data show that heat and drought stress produce high levels of aflatoxin.

A24

PREFERENTIAL FEEDING BY TWO APHID SPECIES ON ENDOPHYTE-FREE VERSUS ENDOPHYTE-INFECTED TALL FESCUE. M. C. Johnson, M. R. Siegel and L. P. Bush, Depts. of Plant Pathology and Agronomy, University of Kentucky, Lexington, KY 40546

Apterous forms of greenbug (Schizaphis graminum) and oat bird cherry aphid (Rhopalosiphum padi) preferred endophyte-free tall fescue leaf sheaths for feeding by up to a 7 to 1 margin over leaf sheaths infected with the endophytic fungus, Acremonium coenophialum, in choice situations. Both species of aphids were considerably less discriminatory between leaf blades of endophyte-free and endophyte-infected plants. No preference was evident in tests comparing endophyte-free and endophyte-infected leaf sheaths of perennial ryegrass. Vegetative tall fescue stems dipped in dilute preparations of methanol extracts of endophyte-free tall fescue seed were preferred by R. padi over stems dipped in similarly prepared extracts of endophyte-infected seed by a 7-10 fold margin.

A25

TOXICITY OF FUNGI ISOLATED FROM FOODSTUFFS AND SOIL: COMPARISON OF TOXICITY IN FIBROBLASTS AND RAT FEEDING TESTS. H.K. Abbas, C.J. Mirocha. Dept. of Plant Pathology and W.T. Shier. Dept. of

Medicinal Chemistry and Pharmacognosy, Univ. of Minnesota, St. Paul, MN 55108.

The toxicities of solid rice medium culture extracts (50% aqueous methanol) of 39 fungi from various parts of the world were compared in 3 systems: (1) cytotoxicity to cultured mouse and human skin fibroblasts; (2) topical application to the skin of rats; and (3) rat feeding tests examined for death, weight loss, food refusal, congestion and hemorrhage of tissues and uterine growth. In feeding tests 16 isolates were highly toxic by all criteria except uterine growth, 4 caused uterine growth as the major clinical sign, 10 were moderately toxic and 11 caused no major clinical signs. Cytotoxicity (log_{10} LC_{50}) values with both types of fibroblasts correlated well with weight loss and food refusal, but not with skin toxicity or uterine growth.

A26

Effects of IPM on Postharvest Quality of Texas Onion.
B. D. Bruton, USDA-ARS, P. O. Box 267, Weslaco, TX 78596.

An IPM system on Texas onion, Yellow Grano 502, was evaluated for impact on postharvest quality. Preharvest pesticide treatments included all combinations of the following: chlorothalonil fungicide, telone nematicide, bensulide herbicide, and parathion and malathion insecticides. Statistical analaysis demonstrated that the influence of pesticides were additive. Marketable yield decreased 23.2%, from 45,844 to 35,193 k/hectare when fungicide was included in the treatment regime. Marketable yields increased by 11.4% in nematicide treatments. The incidence of Aspergillus niger was significantly (P=.05) affected by preharvest application of pesticides. After 52 days postharvest storage, the incidence of A. niger was 17.4% in herbicide treatments, 19.4% in nematicide treatments, and 7.8% in fungicide treatments as compared to the control with 12.3%. The use of insecticides had no measurable effect on postharvest quality.

A27

PHYTOTOXICITY IN DECIDUOUS TREE FRUITS FUMIGATED WITH METHYL BROMIDE TO CONTROL THE MEDITERRANEAN FRUIT FLY. J. M. Harvey, C. M. Harris and J. S. Tebbets. USDA-ARS, Protection and Quarantine Research Unit, P. O. Box 8143, Fresno, CA 93747

Fumigation with methyl bromide (MB) at 30 C increased efficacy, reduced time, lowered the required dosage and reduced residues and phytotoxicity (vs. fumigation at 20 C) in host crops infested with Mediterranean fruit fly (Ceratitis capitata Wied.). Most peach and nectarine cvs. fumigated with 32 g MB/m^3 for 1.5, 2.0 or 2.5 hr were not injured and had residues below 20 ppm. Many plum cvs. and pears were injured by these treatments. Injured plums had black spots and injured pears (Bartlett) had internal breakdown. Injury increased with ripeness of pears. MB residues in pears were below 5 ppm.

A28

PHYTOTOXIC RESPONSES OF CITRUS FRUIT FUMIGATED WITH ETHYLENE DIBROMIDE. L. G. Houck, J. S. Tebbets, J. F. Jenner and P. L. Hartsell. USDA-ARS, Protection and Quarantine Research Unit, P. O. Box 8143, Fresno, CA 93747

Commercially-packed California citrus fruit fumigated with 24 or 32 g ethylene dibromide (EDB)/m^3 for 2 hr at 20 C (30% v/v load factor) developed a high incidence of unacceptable rind injury during post-fumigation storage (5 C for 3 wks then 20 C for 1 wk). Order of susceptibility to EDB injury at 32 g EDB/m^3 was navel oranges > Valencia oranges > lemons. At 12 and 16 g EDB/m^3 the susceptibility order was Valencias > lemons > navel oranges. Rind injury increased with temperature of fumigation (10, 20, or 30 C). Rind injury decreased with an increase in the load factor (v/v) in the fumigation chamber (15, 30, or 58%). Ripe yellow lemons were injured more by EDB fumigation than less-ripe, "silver" lemons.

A29

Effect of hydrostatic pressure and chlorine level in flume and dump tank water on control of postharvest decays in tomato fruit. J. A. Bartz and M. Sherman, Plant Pathology and Vegetable Crops Depts., respectively, University of Florida, Gainesville, 32611.

Chlorine (100-150 ppm free chlorine) is added to the water in flumes and dump tanks in tomato packinghouses in Florida to prevent the spread of postharvest decay pathogens among incoming fruit. In laboratory tests, free chlorine levels of 100-1000 ppm (from NaOCl) reduced but did not prevent decay when tomato fruit previously dipped in a suspension of Erwinia carotovora pv. carotovora were submerged in the water and subjected to a hydrostatic pressure that caused infiltration. Chlorine dioxide (from stabilized 2 or 5% solutions) at similar concentrations was not as effective in reducing disease from control levels as was the NaOCl. Thus, the presence of conventional chlorine compounds in flume and dump tank water does not prevent postharvest disease if fruit become infiltrated during postharvest handling.

A30

VOLATILES FROM WOUNDED CITRUS FRUITS STIMULATE GERMINATION OF PENICILLIUM DIGITATUM CONIDIA. J. W. Eckert, M. Ratnayake and Y. Gutter. Department of Plant Pathology, University of California, Riverside, CA 92521.

Conidia on water agar germinated about 50% when exposed to volatiles from superficially-wounded citrus fruits, compared to <5% germination in their absence. Limonene, α-pinene, sabinene, β-myrcene, acetaldehyde, CO_2, and ethylene were identified by gas chromatography as the major volatiles around wounded oranges. Conidial germination was not stimulated by exposure to any of the compounds alone at concentrations bracketing those measured in the atmosphere around injured fruits, but a mixture of limonene (4.8 µg/ml), acetaldehyde (0.03 µg/ml), ethanol (0.2 µg/ml), and CO_2 (1% v/v) in air stimulated germination on water agar and on silica gel to the same degree as the volatiles surrounding injured fruits. Ethylene (10 and 100 µl/L), alone or in combination with the stimulating mixture of compounds, did not increase germination.

A31

SUBSTRATE SPECIFICITY FOR RED PIGMENT PRODUCTION BY CANTALOUP ISOLATES OF MACROPHOMINA PHASEOLINA. J. R. Dunlap and B. D. Bruton, USDA-ARS, P. O. Box 267, Weslaco, Texas 78596.

Experiments were conducted to identify the substrates and biosynthetic pathway responsible for production of a red pigment by selected cantaloup isolates of Macrophomina phaseolina. Previous research had determined that glycine was required for the in vitro synthesis of the red pigment. Pigment producing isolates were grown in liquid culture supplemented with analogs and additional compounds structurally related to glycine. Of the many two- and three-carbon compounds examined, only L-alanine resulted in pigment synthesis. However, the spectral and chromatographic properties were not the same as the glycine stimulated pigment. The evidence obtained thus far indicates a strict structural requirement for the glycine molecule with no involvement of the glycine or purine biosynthetic pathway.

A32

SELECTION FOR VIRULENCE IN COCHLIOBOLUS HETEROSTROPHUS. J. A. Kolmer, and K. J. Leonard, Department of Plant Pathology, North Carolina State University, Raleigh 27695.

An initial population of 25 ascospore progeny was derived from crosses of 5 compatible pairs of Cochliobolus heterostrophus isolates. Virulence of parents and progeny was evluated from lesion lengths on inbred maize line 316 from the open pollinated variety Jarvis. The four most virulent isolates of each mating type were selected and crossed in all combinations. In the next generation 28 ascospore progeny isolates along with the original parent isolates were evaluated for lesion length on line 316 in two separate trials. In both trials progeny isolates produced significantly longer lesions than original parental isolates. A low correlation among isolates between trials indicates that lesion length has a low heritability and gains from selection will be gradual.

A33

VARIATION AND SELECTION FOR FERTILITY IN COCHLIOBOLUS HETEROSTROPHUS. J. A. Kolmer and K. J. Leonard, Department of Plant Pathology, North Carolina State University, Raleigh 27695.

Approximately 12 isolates of each mating type of C. heterostrophus were mated in all combinations and ascospore progeny were obtained from the 12-16 most fertile crosses for the next generation. Fertility was evaluated as the mean number of perithecia per 1.5 cm diam. leaf disk. Recurrent selection was continued for 3 generations. Genetic variance in the initial population was partitioned between polygenic and monogenic effects. The perithecial gene accounted for 58% of the genetic

variance. Selection based on general fertility resulted in populations with an average of 72, 90, 71 and 100 perithecia per leaf disk after 0, 1, 2 and 3 generations of selection, respectively. Random subpopulations from each generation were evaluated for fertility in a common environment. Subpopulations from 1, 2 and 3 generations of selection had means of 99, 133, and 125, respectively. Selection studies indicate that fertility is a heritable trait and can be used as a genetic marker.

A34

HERITABILITY OF DURABLE, ADULT-PLANT, TEMPERATURE-SENSITIVE RESISTANCE TO PUCCINIA STRIIFORMIS IN PACIFIC NORTHWEST WHEATS. Gene Milus and Roland F. Line, Dept. of Plant Pathology, Wash. State Univ. and USDA-ARS, respectively, Pullman, WA 99164.

Inheritance of resistance to stripe rust in Gaines, Nugaines, and Luke, based on parental, F1, F2, and backcross populations from crosses between these cultivars and with a susceptible line, was reported in 1983. This paper compares narrow heritability calculated by parent-offspring regression and by variance components. F3 rows were evaluated at heading for rust intensity and infection type. F2 and F3 data were transformed to standard units by dividing the data by their standard deviation. The relationship in standard units of F3 rows to parent F2 plants was determined by regression analysis. Narrow heritability estimated by regression was 0.08-0.51 for intensity and 0.13-0.60 for infection type. These estimates were 14-68% lower than estimates by variance components. The regression method may be more useful in plant breeding because it provides a lower limit for heritability and requires less data.

A35

TETRAD ANALYSIS USED TO EXAMINE POLYGENIC INHERITANCE OF VIRULENCE IN USTILAGO HORDEI. B. J. Christ and C. O. Person. Department of Botany, University of British Columbia, Vancouver, B. C. V6T 2B1

F_1 teliospores of Ustilago hordei from a cross of T_1 X T_4 were known to be heterozygous for a virulence gene matched to resistance in barley cultivar Trebi. Tetrad analysis was used for further examination of virulence in several F_1 and F_2 cultures. Sets of dikaryons produced from selfing were inoculated onto Trebi and Odessa, a resistant and susceptible cultivar respectively. Results showed that polygenes modified the effects of the virulence gene. The segregation patterns for many of the sets of dikaryons were similar regardless of cultivar. The only difference was that in general Odessa had a higher percent smutted plants than Trebi. This indicated that the polygenes were nonspecific in action.

A36

PATHOGENICITY CHANGES IN Uromyces appendiculatus AFTER FIVE ASEXUAL GENERATIONS ON A BEAN CULTIVAR. Helen Miller Alexander, Dept. of Biology, Univ. of Louisville, Louisville, KY 40292; J.V. Groth and A.P. Roelfs, Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

A field-collected population of Uromyces appendiculatus, the bean rust pathogen, was obtained from a Pinto 111 bean field in SW Minnesota, and maintained for five asexual generations on the partially resistant cv. Slimgreen. Latent period, pustule size, and urediniospore production on Slimgreen changed little. In contrast, there was an increase in the frequency of virulence on the line US#3, and decreases in the frequency of virulence to Early Gallatin, Roma, and B-1349 by the fifth generation. Therefore, changes in virulence frequencies can be independent of pathogen exposure to the corresponding host resistance. Minnesota populations of U. appendiculatus are polymorphic for virulence on all four bean cultivars, which are rarely grown in the Midwestern United States.

A37

WITHDRAWN

A38

CROSS INOCULATION OF RHIZOCTONIA SOLANI ANASTOMOSIS GROUP 2 TYPE 2 FROM CARROT, SUGARBEET, CORN AND ST. AUGUSTINEGRASS. M. P. Grisham, Department of Plant Pathology and Microbiology, Texas A&M University, College Station, Texas 77843.

Specific isolates of Rhizoctonia solani anastomosis group 2 type 2 (AG2-2) that cause Rhizoctonia canker of carrot, crown and root rot of sugarbeet, brown patch of St. Augustinegrass, and crown and lateral root rot of corn were tested for pathogenicity on carrot, sugarbeet, St. Augustinegrass and corn. Distinct differences in pathogenicity and aggressiveness were observed among the AG2-2 isolates tested. The sugarbeet isolate was pathogenic on all hosts except corn and was more aggressive to carrot and St. Augustinegrass than other pathogenic AG2-2 isolates on these hosts. The carrot isolate was pathogenic to carrot, sugarbeet, and corn. The corn isolate was pathogenic on all hosts. On St. Augustinegrass, the corn isolate caused distinct leaf lesions in addition to the nodal rotting caused by the St. Augustinegrass and sugarbeet isolates. The St. Augustinegrass isolate was not pathogenic on corn and was less aggressive on carrot and sugarbeet than other AG2-2 isolates.

A39

RESISTANCE IN MICHIGAN WINTER WHEAT TO WHEAT SPINDLE STREAK MOSAIC VIRUS (WSSMV). K. Zagula Haufler & D.W. Fulbright, Dept. of Bot. & Plant Path.; M. Van Koevering & E.H. Everson, Dept. of Crop & Soil Sci., Mich. State Univ., E. Lansing, MI 48824

WSSMV has been found in Michigan winter wheat fields during the last several growing seasons. Various degrees of symptom severity have been observed among hundreds of wheat lines screened for resistance. Five commercial varieties and ten experimental lines were selected and rated on their reactions to WSSMV under field and growth chamber conditions. Ratings were based on symptom severity and virus particle counts using immunospecific electron microscopy. Three of the commercial varieties were considered susceptible, while the remaining varieties and experimental lines exhibited varying degrees of resistance to WSSMV. Because of the phenotypic variation found among different genotypes, three varieties and four experimental lines were chosen as parents and crossed in a diallel mating design to study the inheritance of resistance to WSSMV. Initial results from analysis of F_1 progeny show that resistance to WSSMV is dominant and controlled by one or two major genes.

A40

ZUCCHINI YELLOW MOSAIC VIRUS ASSOCIATED WITH A SEVERE DISEASE OF CANTALOUPE AND SQUASH IN CALIFORNIA. S. T. Nameth, J. A. Dodds, and A. O. Paulus, Department of Plant Pathology, University of California, Riverside, CA 92521.

A severe mosaic disease affected both cantaloupe and squash production in southern and central California in 1982/1983. The disease caused severe mosaic symptoms on both foliage and fruit. The symptoms resembled those described for zucchini yellow mosaic virus (ZYMV). Antisera prepared against ZYMV reacted specifically with both purified virus and plant sap infected with the California agent (ZYMV-Ca). Fifty cultivars of cantaloupe, mixed melon, watermelon and squash showed no resistance or tolerance to ZYMV-Ca, nor has the virus been detected in any of 1000 seedlings of squash grown from seed from infected plants. Additional isolates from cucurbits grown in Oregon, Arizona and Mexico also reacted with antisera prepared against ZYMV-Ca. Results indicate that a serious disease of cucurbits caused by ZYMV previously reported in the eastern US and Europe is also widespread in the western US and Mexico.

A41

CORRELATION OF YIELD REDUCTIONS WITH SEVERITIES OF DISEASE SYMPTOMS IN GRAIN SORGHUM [SORGHUM BICOLOR (L.) MOENCH] INFECTED WIIH SUGARCANE MOSAIC OR MAIZE DWARF MOSAIC VIRUSES. J. D. Alexander, R. W. Toler, and L. M. Giorda. Dept. of Plant Pathology and Microbiology, Texas A&M Univ., College Station, Texas 77843

Twenty-seven grain sorghum accessions were inoculated with either maize dwarf mosaic virus strain B or one of two isolates of sugarcane mosaic virus strain H in three separate field tests.

Disease percentages and symptom severities were compared to percent yield reductions using linear regression. The resulting estimates of yield reductions in diseased grain sorghum are 10% with moderate mosaic symptoms, 25% with severe mosaic symptoms, 40% with slight redleaf symptoms, 70% with severe redleaf symptoms, and 100% with redleaf symptoms and generalized necrosis. Yield reductions in response to a particular disease severity vary across different accessions and, therefore, limit the reliability of these parameters for predicting yield reductions.

A42

REACTIONS OF SUDANGRASS ACCESSIONS TO STRAIN A OF MAIZE DWARF MOSAIC VIRUS. Stephen R. Vann, Robert W. Toler, Dept. of Plant Pathology and Microbiology, and Frederick R. Miller, Dept. of Soil and Crop Sciences, Texas A&M University, College Station, TX 77843

Seventy-seven foreign and domestic sudangrass (Sorghum sudanense Hitchc.) accessions were screened for resistance to maize dwarf mosaic virus--strain A. During the spring of 1983, half-row field inoculations were made of seedlings at the 4-6 leaf stage using a DeVilbiss spray gun at 6.33 kg/cm^2. Disease symptom severity and percent infection were rated after two and one-half weeks. Symptoms varied from mild to severe mosaic and stunting of plants. Of the accessions tested, 10 were susceptible, 49 intermediate, and 18 resistant. For further testing, examples of resistant candidates included Georgia 337 and P.I. 302223 USA; intermediate candidates included TE-1005 and Tift; and susceptible candidates included TE-1001 and Nebraska 7035. Symptomatic tissue was prepared for transmission electron microscopy and examined to confirm presence of the virus.

A43

FIRST REPORT OF A SUBTERRANEAN CLOVER RED LEAF-LIKE VIRUS IN THE WESTERN HEMISPHERE. G. R. Johnstone, Hsing-Yeh Liu and James E. Duffus, Tasmanian Department of Agriculture and USDA-ARS, U.S. Agricultural Research Station, Salinas, CA 93915.

A luteovirus closely related serologically to subterranean clover red leaf virus (SCRLV) has been found in California. SCRLV was previously reported only from Australia and New Zealand. The California virus isolates (SCRLV-C) were found from naturally infected legumes in central California and reacted in ELISA tests with antiserum to the Australian isolate of this virus but not to legume yellows virus antiserum or to beet western yellows virus antiserum. SCRLV-C virions, purified and concentrated from pea (Pisum sativum L. cv. Puget), appeared identical to those from Australia on the basis of particle morphology and serological tests. The Australian isolates of SCRLV are, however, transmitted only by Aulacorthum solani (Kalt.) whereas the California isolates are transmitted only by the pea aphid Acyrthosiphon pisum (Harris).

A44

TOMATO NECROTIC DWARF -- A NEW TYPE OF WHITEFLY-TRANSMITTED VIRUS. Richard C. Larsen, James E. Duffus and Hsing-Yeh Liu, USDA-ARS, U.S. Agricultural Research Station, Salinas, CA 93915.

A new whitefly-transmitted virus causing leaf necrosis and severe stunting of tomato plants has been recently isolated from the Imperial Valley, California. The infectious agent, Tomato Necrotic Dwarf Virus (T_{om}NDV), which affects tomato, pepper, tomatillo, eggplant, and several weed hosts is transmitted by Bemisia tabaci as well as being mechanically transmitted. Purified virus has been shown to contain three distinct isometric components ca. 30 nm in diameter. Preliminary investigations of the virions by electron microscopy and spectrophotometric analysis suggest that only the middle and bottom components contain nucleic acid. The $A_{260/280}$, uncorrected for light scattering, was typically 1.0, 1.78 and 1.91 for the top, middle, and bottom components, respectively. Sedimentation coefficients ($S_{20,w}$) of the virions are 57S(T), 117S(M) and 138S(B). Tests by mechanical inoculation indicate that both middle and bottom components are required for infection.

A45

PERFORMANCE OF PHOMOPSIS-INFECTED SOYBEAN SEEDLOTS: INFLUENCE OF SOIL WATER POTENTIAL. M. L. Gleason and R. S. Ferriss. Dept. of Plant Pathology, University of Kentucky, Lexington, KY 40546.

In growth chamber and greenhouse experiments, soybean seedlots were incubated for 3 days at 0,-0.02,-.1,-1,-5,-15 and -60 bars soil water potential, followed by 18 days at near-optimum soil moisture. In pasteurized soil, emergence and establishment (EM/ES) of lots heavily infected (20% of seeds) by Phomopsis sp. peaked at -.02 to -1 bar and declined in drier treatments. Low-Phomopsis seedlots (0.05% infection) had little decline in EM/ES for treatments <-.02 bar. Benomyl or Vitavax 200 seed treatment lessened EM/ES decline of high-Phomopsis lots in treatments <-.02 bar. In natural soil, EM/ES were lower than in pasteurized soil, especially for high-Phomopsis lots in moderate-moisture treatments. Vitavax 200, but not benomyl, effectively prevented this reduction. Saturated soil treatment gave lowest EM/ES; values were similar for all seedlot-by-soil treatment combinations. These findings support the idea that Phomopsis sp. infection of soybean seeds contributes to reduced EM/ES primarily in dry soils.

A46

HISTOPATHOLOGY OF MIXED INFECTIONS BY COLLETOTRICHUM TRUNCATUM AND PHOMOPSIS SPP. IN SOYBEAN SEEDS. I. Kunwar, T. Singh and J. B. Sinclair, Dept. of Plant Pathology, Univ. of Illinois, 1102 S. Goodwin, Urbana, IL 61801.

Hyphae of C. truncatum and Phomopsis spp. were most abundant in the hourglass cell layer of the seedcoat and were intercellular in all layers of the seedcoat and cotyledons, with C. truncatum hyphae being intracellular in the hourglass cells. Mycelial mats were formed by both fungi in the endosperm. Phomopsis spp. hyphae were most abundant on the surface of and within cotyledons. When both fungi were present, cotyledonary cell protoplasm was coagulated, ruptured and had prominant nuclei and large vacuoles. In shrivelled seeds, the palisade and hourglass cell layers remained intact, while adjacent tissues in the seedcoat and cotyledons disintegrated. Hyphae of both fungi were found in intercellular spaces.

A47

DISTRIBUTION OF CERCOSPORA KIKUCHII, CERCOSPORA SOJINA AND PHOMOPSIS SPP. IN SOYBEAN SEEDS. T. Singh and J. B. Sinclair, Dept. of Plant Pathology, Univ. of Illinois, 1102 S. Goodwin, Urbana, IL 61801.

Field-grown seeds were used in histopathology studies that showed C. kikuchii and Phomopsis spp. became established in the embryo, endosperm and seedcoat, and C. sojina only in the seedcoat. Each fungus penetrated the hillar groove, grew into the hillar tracheid and aggregated in stellate parenchyma. Phomopsis spp. were most aggressive, causing cotyledon cells to be thick-walled, hypertrophied and vacuolated. Cell lysis occurred. Phomopsis spp. were common in embryos and between the seedcoat and embryo. C. kikuchii grew in the cotyledons, rarely in the hypocotyl-radicle axis, and formed sclerotia in seedcoats and caused necrosis in cotyledons and vascular elements. C. sojina formed sclerotia in seedcoats, occasionally on the seed surface and between seedcoat and embryo, but seldom in the hypocotyl-radicle axis.

A48

EFFECT OF CROP HISTORY, TILLAGE, ROW-SPACING AND HERBICIDE ON SOYBEAN SEED QUALITY. J. E. Bowman, G. L. Hartman, R. D. McClary, J. B. Sinclair (Plant Pathology), J. W. Hummel and L. M. Wax (USDA, Agr. Engr. and Agronomy), Univ. of Illinois, 1102 S. Goodwin, Urbana, IL 61801.

The effect of continuous soybeans vs. a corn-soybean rotation, conventional vs. reduced tillage, 25 and 76 cm-row spacings and four levels of weed control [postemergence (POST), preplant incorporated (PPI), no weed control, and hand weeded] on Corsoy-79 seed quality was studied for 3 years (1981-83). Soybeans from rotation plots with corn had greater yield, weed control, 1000-seed weight, germination, vigor and lower recovery of seedborne Phomopsis spp. compared to continuous soybeans. Soybeans in 25 cm rows had better weed control and were infected with lower levels of Alternaria spp. than 76 cm rows. The POST treatment (sethoxydim + alachlor) had greater yield and seed quality than PPI (alachlor + metribuzin). Tillage did not affect yield or seed quality.

A49

INOCULUM THRESHOLDS FOR SEEDBORNE BACTERIA. N. W. Schaad. Department of Plant, Soil and Entomological Sciences, University of Idaho, Moscow, ID 83843.

The increased severity of diseases caused by seedborne bacteria has increased efforts in developing laboratory assays based upon serology and selective agar media. Such assays should be 1) rapid, 2) specific, 3) sensitive, and 4) correlated with field disease. It is the correlation which is perhaps most important. For example, epiphytotics of black rot result from seed lots with an infection level of 0.03% in the warmer production fields of Southeastern U.S. On the other hand, black rot fails to develop in the cooler seed

fields of western Washington. With halo blight of bean, one plant/16,000 can result in a crop failure. In France, lots with infection levels of 0.5, 0.05, and 0.005%, respectively, resulted in severe, variable and no halo blight. In Idaho, levels of X. translucens > 1.0 x 10^4 CFU/ml in seed washing, agar plating assays resulted in severe black chaff whereas levels < 1.0 x 10^4 did not.

A50

SEED-BORNE VERTICILLIUM ALBO-ATRUM IN ALFALFA. H. C. Huang and M. R. Hanna, Agriculture Canada Research Station, Lethbridge, Alberta, Canada T1J 4B1.

Infection of alfalfa seeds by Verticillium albo-atrum was investigated in root-inoculated plants of the cultivars Anchor, Vela, and Vernal, and hybrids of Beaver x Lutèce. The frequency of infected seeds ranged from 0 to 4.8%, following hand-pollination of plants showing disease symptoms. Although the pathogen was present throughout the stems, it remained sporadic in peduncles, pedicels, pods, and seeds. Inoculation of stigmas of healthy plants of Vernal at flower-tripping resulted in infection and discoloration of the stigma and upper style. Even though hyphae were present in lumina of the parenchyma of the style, the pathogen was restricted to the style during all stages of seed development. Furthermore, it was not detected in the seeds from pods with infected styles. Under humid conditions, the fungus in the remnant style tissue of a mature seed pod was able to colonize the pod and seed coat.

A51

DETECTION OF XANTHOMONAS CAMPESTRIS PV. CAROTAE IN CARROT SEED - T.-L. Kuan, G.V. Minsavage and R.L. Gabrielson Asgrow Seed Company, San Juan Bautista, CA 95045 and Western Washington Extension and Research Center, Puyallup, WA 98371

Field observations have suggested that soil and seed can be inoculum sources for carrot bacterial blight and use of clean seed is an important disease control method. Effective disease control in the seed fields and effective eradicant seed treatments are needed to produce clean seed. An accurate method of detecting the pathogen in seed is necessary to evaluate the effectiveness of control procedures.

A method has been developed for the isolation of Xanthomonas campestris pv. carotae from carrot seed consisting of a low temperature stationary soak for extraction of the bacteria from seeds, followed by a vigorous shake wash of the seeds, concentration of bacterial cells by centrifugation and dilution plating onto modified Kado and Heskett's D5 medium. Replicate testing of seed stocks has proven the method to yield reproducible and consistent results when testing seed lots with infection rates as low as one infected seed in 10,000.

A52

A VOLUMETRIC SPORE TRAP DESIGNED FOR MONITORING VENTURIA INAEQUALIS SPORE RELEASE IN APPLE SCAB MANAGEMNENT PROGRAMS. M.G. Zuck and F.L. Caruso, Dept. of Botany and Plant Path., University of Maine, Orono, 04469.

A simple volumetric spore trap has been developed for use by apple growers to enable them to conveniently monitor levels of V. inaequalis ascospores in their orchards. A moisture-activated switch operates the trap during rainy periods. Spores are trapped in a narrow band on a dry or coated microscope slide located inside the trap. A wind-oriented orifice approximately 5 cm in diameter is the entry point for the spores. Vacuum (15-30 l/min) is supplied with a 12V DC blower powered by a car battery. Trapping efficiency of the device was determined for spores of varied sizes. The relationship between spore catch and disease incidence was studied in the greenhouse. The spore trap reliably detected V. inaequalis ascospores at levels below that required to cause infection on McIntosh seedling leaves.

A53

ECONOMIC THRESHOLDS FOR CHEMICAL CONTROL OF BEAN RUST. Richard A. Meronuck and P.S. Teng. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul 55108.

Bean growth stage-dependent economic thresholds for rust (Uromyces phaseoli) were developed from field experiments in which epidemics on Pinto beans cv. UI114 were produced by two inoculation dates and different spray schedules with chlorothalonil. Weekly rust ratings and yield data were used in regression analysis to determine equations for estimating 1) yield loss expected with existing rust severity but no spray program and 2) yield loss recovered by spraying on assessment date and one or two additional sprays. Equations were programmed as a bean rust spray decision aid, "BEANRUS", and implemented on hand-held microcomputers. With inputs of potential yield (2000 lb), bean price (24¢/lb), growth stage (R4), rust rating (150) and spray cost ($7/ac), BEANRUS estimated net return of $34.7/ac with 3 sprays and yield increase of 648 lb/ac.

A54

INTEGRATED CONTROL OF PHYTOPHTHORA ROOT ROT OF SOYBEANS. A. F. Schmitthenner and D. M. Van Doren, Department of Plant Pathology and Department of Agronomy, respectively, The Ohio State University/OARDC, Wooster, OH 44691.

Combinations of tile and no tile drainage; six tillage levels, ranging from zero to complete; continuous soybeans and corn-soybean rotation; multirace resistance; high and low-tolerance to Phytophthora root rot and metalaxyl seed treatment were evaluated as integrated control components. In the fifth year of the experiment the greatest Phytophthora damage occurred using the low-tolerant cultivar-surface drainage only-reduced tillage-continuous soybean cropping combination. Overall (main effects), high tolerance improved yield 25%; complete tillage, 19%; tile drainage, 10%; rotation, 9%; and metalaxyl seed treatment, 6%. All factors combined increased yield only 48%, indicating that the main effects were not additive. Multirace resistance (main effect) improved yield only 24%. Under optimum conditions for Phytophthora metalaxyl was more effective as a soil treatment than as a seed treatment and seed treatment was more effective on high than on low-tolerant cultivars.

A55

SOIL SOLARIZATION FOR THE CONTROL OF PHYTOPHTHORA CINNAMOMI: THERMAL AND BIOLOGICAL EFFECTS. Y. Pinkas, Arna Kariv and J. Katan*, Dept. of Plant Pathology, ARO, Volcani Center, Bet-Dagan, and *Dept. of Plant Pathology & Microbiology, The Hebrew University, Rehovot, Israel.

P.cinnamomi was first detected in Israel in Sept.1982. The efficacy of solarization in controlling this pathogen was studied in an avocado grove where infected trees had been removed. The soil was covered with transparent polyethylene for 42 days during July-August 1983. Soil samples,up to 70 cm depth,were collected and the existence of P. cinnamomi in the soil was assessed by Persea indica seedlings as trap plants and by culturing the roots of avocado seedlings planted in the sampled soil. Only 3% of the solarized samples were infested with the pathogen as compared to 69% in the control. In naturally infested soil only 10% of the pathogen propagules survived 4 h heating at 36°C. In addition to thermal effects other factors apparently contributed to pathogen control. Pathogen reinfestation of solarized soils was suppressed as expressed by slower hyphal growth accompanied by fewer chlamydospores. Sporangia formation was also suppressed.

A56

MANAGEMENT OF TWO PYTHIUM SPP. IN HYDROPONIC LETTUCE PRODUCTION. Andrew C. Schuerger and Kristin G. Pategas, The Land, EPCOT Center, P. O. Box 40, Lake Buena Vista, Florida 32830

A root death of lettuce cultivars 'Summer Bibb', 'Ostinata', and 'Salad Bowl', caused by Pythium myriotylum or P. irregulare frequently occurred in greenhouse hydroponic systems. Routine sanitizing of the hydroponic systems were effective in reducing but not eliminating the disease outbreaks. Thirty chemical compounds representing surfactants, organic acids, antibiotics, and food additives were screened in vitro, at rates up to 2000 ppm, using a modified, petri dish, grass-bait technique. Sodium laural sulphate (800 ppm), Ivory Liquid (800 ppm), and potassium sorbate (2000 ppm), chlorotetracycline (100 ppm), and polymyxin B (1000 ppm) were effective in preventing grass-bait colonization. NaOCL (50 ppm) was also effective, but in vivo tests established it to be phytotoxic at levels effective in reducing the incidence of the disease.

A57

INTEGRATED PEST MANAGEMENT IN ILLINOIS: SOYBEAN DISEASEPOPULATIONS AS AFFECTED BY TILLAGE, CROP ROTATION, AND PEST MANAGEMENT LEVEL. T.D. Rogers, S.M. Lim, and L.M. Wax, 1102 S.Goodwin Ave., AE106 Turner Hall, University of Illinois, Urbana, Illinois, 61801

This study was undertaken in 1979 to determine the effects of cropping system, tillage, and level of pest management on crop yields, and populations of weeds, insects, and diseases at three locations in Illinois. Pest management, applied at three levels, consisted of several pesticide applications of varying number and rate. Disease surveys were taken every two weeks beginning three weeks after planting until crop harvest maturity. Both the

incidence and severity of soybean diseases differed by treatment, location, and year. The major disease at all three locations, septoria brown spot, was significantly affected by the level of pest management(2%-78% at R6). Downy mildew severity(19%) was significant in the double cropped soybean plots following wheat at Dixon Springs in southern Illinois. Incidences of brown stem rot(1-28%), and charcoal rot(1-78%) greatly differed by location and year.

A58

TRANSMISSION OF VERTICILLIUM ALBO-ATRUM TO ALFALFA VIA FECES OF LEAF-CHEWING INSECTS. H. C. Huang and A. M. Harper, Agriculture Canada Research Station, Lethbridge, Alberta, Canada T1J 4B1.

Alfalfa leaves infected with Verticillium albo-atrum were fed to the leaf-chewing insects; grasshoppers (Melanoplus sanguinipes and M. bivittatus), alfalfa weevils (Hypera postica), and wooly bears (Apantesis blakei), to determine survival of the pathogen after passage through the digestive tracts. V. albo-atrum survived in the digestive tracts of all tested species, usually appearing in the feces one day after feeding. The percentage of V. albo-atrum-contaminated feces varied greatly among individuals within species but it was positively correlated with the duration of feeding on diseased tissues. When the V. albo-atrum-contaminated feces from grasshoppers were buried near roots of alfalfa seedlings, 20.8% of the plants were infected and showed wilt symptoms after six weeks. The role of leaf-chewing insects on the dissemination of V. albo-atrum in alfalfa and other crops is discussed.

A59

FUTURE DIRECTIONS FOR ALFALFA IPM. Larry E. Gholson and J. Keith Waldron, Plant Science Division, University of Wyoming, P.O. Box 3354, University Sta., Laramie, WY 82071

A survey was developed soliciting feedback concerning future needs and directions for Alfalfa Integrated Pest Management. Questionnaires were distributed to 85 university alfalfa IPM research and extension contacts in 32 states. Fifty-five persons (28 states) responded. Results indicated a dedication to improving current alfalfa IPM programs. The consensus of opinion revealed a desire to shift emphasis from predominantly insect pest management efforts to multidisciplinary efforts with greater input from plant pathology, weed science, agronomy, agricultural engineering, and agricultural economics. Suggestions offered reflecting this desired shift of emphasis included: retitling IPM to Integrated Crop Production Management, improved responses to clientele needs and other measures designed to reflect the total needs of alfalfa production.

A60

THE ROLE OF PLANT DISEASE IN THE DEVELOPMENT OF CONTROLLED ECOLOGICAL LIFE SUPPORT SYSTEMS. B. D. Nelson. Dept. of Plant Pathology, North Dakota State University, Fargo, 58105.

Controlled ecological life support systems (CELSS) for human habitation are proposed for long duration exploration of extraterrestrial habitats. The prime candidates for the photoautotrophic components of CELSS are edible higher plants. Plants on earth, however, are often seriously damaged by infectious and non-infectious diseases. There is reason to assume the same would occur in CELSS. Diseases would adversly disturb food and oxygen production, carbon dioxide utilization and waste recycling. Disease prevention should be a major design factor in CELSS development to reduce potential damage to plants. Examples of prevention aspects for consideration are: mechanisms for excluding pathogens; confinement schemes to prevent spread; erradication procedures; plant selection for resistance; introduction of protective phylloplane and rhizosphere microbial communities. A model for the prevention of plant disease in CELSS is proposed.

A61

TARALAN'S MULTIFACTOR SYSTEM AND PLANT PATHOLOGY: A WORKING INTEGRATED CROP MANAGEMENT PROGRAM. Bill Becker, Taralan Corporation, 1229 W. Edwards, Springfield, Illinois 62704.

The Taralan Multifactor System formulated by John L. Strauss has been used profitably by farmers since 1972. By working with 50-plus production factors, Taralan has helped farmers reduce their cost per unit by increasing yields and decreasing waste. The precision of the system and the management of the farmer are very important. Any decision whether it is tillage, fertility, crop selection, varietal choice, crop protection or timing, needs to be both economically and ecologically sound. By eliminating the man-controlled stresses such as the wrong fertilizer and plant population, disease diagnosis becomes easier and more accurate. Of particular interest is the stalk rot, nematode, and soil compaction complex in corn.

A62

DELINEATION OF XANTHOMONADS WITH MONOCLONAL ANTIBODIES. A.M. Alvarez, Dept. Plant Pathology, A.A. Benedict, and C.Y. Mizumoto, Dept. Microbiology, University of Hawaii, Honolulu, HI 96822.

Monoclonal antibodies (MCA) were produced to identify Xanthomonas campestris pv. campestris (Xcc) strains for epidemiological studies. MCA were characterized by reactions with 325 Xanthomonas strains including different species (X.fragariae, X.albilineans), pathovars of X. campestris (pv. begoniae, campestris, dieffenbachiae, euphorbiae, malvacearum, manihotis, oryzae, phaseoli, poinsetticola, vesicatoria, vitians), and xanthomnads isolated from local hosts (Allium, Cordyline, Cynodon, Polyscias, Zingiber). One MCA (X-1) reacted with all xanthomonads but not with 70 bacteria of diverse animal, plant, and soil origins representing 44 species in 18 Gram + and Gram - genera. Three MCA (X-9, -13,-17) were specific for Xcc. Crucifer strains were divided into subgroups based on reactions with 6 MCA (X-2,-3,-4,-10,-15, -16). Based on this analysis strains having distinct serotypes were used in field studies to trace the inoculum source of the black rot disease.

A63

ERWINIA CAROTOVORA SUBSP. CAROTOVORA DNA ENCODING PECTATE LYASES CLONED INTO PLASMID PRB322. D. P. Roberts, P. M. Berman*, G. H. Lacy, M. S. Mount*, C. Allen, Dept. of Plant Pathology, Physiology, and Weed Science, Virginia Polytechnic Institute and State University, Blacksburg, VA, 24061 and *Department of Plant Pathology, University of Massachusetts, Amherst, MA, 01003.

Erwinia carotovora subsp. carotovora strain EC14 DNA was cloned into plasmid pBR322 and transformed into Escherichia coli strain HB101. These hybrid plasmids encode at least two pectate lyases (PL) that are produced extracellularly by the E. coli host. The PL's were purified by DEAE cellulose chromotography and isoelectric focusing. PL activities were divalent cation dependent at pH 8.5 and degraded sodium polypectate polymers in a random fashion. No lyitic or hydrolytic activities were detected from the E. coli control strain.

A64

CONJUGATIONAL TRANSFER OF COPPER RESISTANCE AND AVIRULENCE TO PEPPER WITHIN STRAINS OF XANTHOMONAS CAMPESTRIS PV. VESICATORIA. R. E. Stall, D. C. Loschke, and R. W. Rice. Plant Pathology Dept., Univ. of Florida, Gainesville, FL 32611.

Resistance to copper occurs frequently among strains of Xanthomonas campestris pv. vesicatoria isolated in Florida. Copper resistance (Cu^+) was transferred to copper sensitive (Cu^-) strains by conjugational procedures. Copper resistant strains were donors and the frequencies of transfer varied from 0 to 1.6×10^{-3} per donor cell, depending upon strains that were mated. Transfer of a plasmid of about 125 megadaltons to recipients was confirmed by agarose gel electrophoresis. If copper resistant strains also caused a hypersensitive reaction (Cu^+HR^+) in incompatible lines of Capsicum annuum, and were mated with Cu^-HR^- strains, the conjugants selected for Cu^+ also were HR^+. When a donor strain, Cu^+HR^- was mated with Cu^-HR^- strains, conjugants were HR^-. In some strains of X. campestris pv. vesicatoria, resistance to copper and avirulence to certain pepper plants are associated with a conjugative plasmid.

A65

ISOLATION AND MAPPING OF Tn5 MUTATIONS IN PATHOGENICITY GENES OF ERWINIA AMYLOVORA. E. M. Steinberger and S. V. Beer, Department of Plant Pathology, Cornell University, Ithaca, New York 14853

To study the molecular basis of pathogenicity in Erwinia amylovora non-pathogenic mutants were induced by mutagenesis with Tn5. Total genomic DNA of each mutant was isolated and

digested with the restriction endonuclease Eco R1, which does not cut Tn5 sequences. Southern blot hybridization was performed using Tn5 as the radioactive probe. The results indicate that several genes are involved in pathogenicity. To further characterize the mutants, genomic DNA was size-fractionated on sucrose gradients. DNA fractions of interest were purified, ligated into the plasmid vector pBR322 and cloned in Escherichia coli HB101. Since Tn5 codes for resistance to kanamycin, Tn5 containing clones of E. coli were identified by their resistance to kanamycin. A map of the Tn5 insertions with respect to the adjacent E. amylovora sequences is being constructed.

A66

STRUCTURAL AND FUNCTIONAL ANALYSIS OF THE PSEUDOMONAS SYRINGAE PV. SYRINGAE ice REGION AND CONSTRUCTION OF ice⁻ DELETION MUTANTS. C. S. Orser, R. Lotstein, E. Lahue, D. K. Willis, N. J. Panopoulos, and S. E. Lindow, Dept. of Plant Pathology, Univ. of California, Berkeley, CA 94720.

The ice nucleation region of P. s. syringae Cit 7 has been physically mapped and analyzed through subcloning, deletion and insertion mutagenesis. It spans an apparently contiguous segment, ca 4 kb, which imparts upon E. coli ice nucleating ativity (INA). Deletions removing specific portions of this segment and Tn5 insertions at various points within it abolish INA. No signficant differences in INA were observed when the fragment was inserted downstream from the lacZ operator-promotor in opposite orientations. A pBR325-ice recombinant plasmid carrying a ca 1.4 kb deletion internal to the ice region was introduced into the DNA-source strain selecting for vector integration (ca. 10^{-3}/cell). Subsequent loss of the vector (ca. 3×10^{-4}/cell) with simultaneous loss of the wild-type ice region was observed. Southern blot analysis confirmed the replacement of the functional ice region for the deletion, presumably through reciprocal exchange. The ice deletion mutants did not incite frost injury to plants.

A67

MOLECULAR CHARACTERIZATION OF TISSUE-SPECIFIC AND GENERAL VIRULENCE GENES IN PSEUDOMONAS SYRINGAE PV. SYRINGAE. D. K. Willis and N.J. Panopoulos. Department of Plant Pathology, University of California, Berkeley, CA 94720.

We have isolated two Tn5 generated avirulent mutants of a Pseudomonas syringae pv. syringae isolate that is pathogenic on bean (Phaseolus vulgaris). Mutant NPS3139 is avirulent on both pods and leaves while strain NPS3136 is avirulent on pods but causes typical water-soaked lesions on leaves. Southern blot analysis using λ::Tn5 revealed that Tn5 is located in different EcoRI restriction fragments in each mutant. By selecting for the kanamycin resistance marker of Tn5, we have cloned the Tn5 and flanking chromosomal DNA from both mutants. Using the cloned mutant fragment from NPS3136 as a probe, we have found that the corresponding wild-type fragment is conserved with respect to homology and size among several isolates of P. s. syringae and in two isolates of P. s. phaseolicola. The cloned fragments are currently being used to isolate the wild-type virulence genes from a cosmid genomic bank of the parental strain.

A68

THIRTEEN MONCLONAL ANTIBODIES TO SPECIFIC EPITOPES ON SPIROPLASMA CITRI. C. P. Lin and T. A. Chen, Department of Plant Pathology, Rutgers University, New Brunswick, NJ 08903.

Thirteen hybridoma cell lines secreting specific monoclonal antibodies against distinct epitopes on S. citri were selected from hybridomas produced by fusing NS-1/1-Ag4-1 mouse myeloma cells with splenic cells from mice immunized with S. citri culture. Using indirect ELISA with biotinylated anti-mouse IgG and IgM, the antibodies secreted by these 13 clones reacted only with S. citri, and distinguished S. citri from the corn stunt spiroplasma (I-747), honey bee spiroplasma (AS-576) of the same serogroup (group I) as well as from the spiroplasmas in the different serogroups: SR-3, group IV; S. floricola, group III; and AES, group X. Isotyping of these monoclonal antibodies was by the Ouchterlony method using class and subclass specific antibodies for mouse IgG1, IgG2a, IgG2b, IgG3, and IgM. Three clones were of the IgM isotype, four of the IgG1 isotype, and six of the IgG3 isotype. Isotypic switch might have occurred in the 3 clones of the IgM class during subcultures.

A69

Chemotaxis by Pseudomonas syringae pv. tomato. D.A. Cuppels and W. Smith. Agriculture Canada, Research Centre, London, Ontario, N6A 5B7, Canada.

Optimal conditions for chemotaxis by Pseudomonas syringae pv. tomato DCT6D1 and DC838F were defined using the Adler capillary assay technique with tomato intercellular fluid (TIF) as an attractant. Ethylenediaminetetraacetic acid (10^{-4} M), $MgCl_2$ (10^{-3} M), and mannitol (10^{-4} M) were required for a maximum response to TIF in 0.01 M potassium phosphate at pH 7.2. Reproducibility was optimum when a low concentration of bacteria (4×10^6 cfu/ml) and a long incubation time (60 min) were used. Both strains also gave a positive chemotactic response toward glucose; the response was dependent upon prior growth of the bacteria in a glucose-containing medium. Mutants nonchemotactic toward glucose had normal flagella, were actively motile, and had growth rates comparable to those of the wild type; no differences in pathogenicity were seen.

A70

CHEMOTAXIS OF ERWINIA HERBICOLA. M. J. Klopmeyer and S. M. Ries. Department of Plant Pathology, University of Illinois at Urbana-Champaign, 1102 S. Goodwin Ave., Urbana, IL 61801.

Erwinia herbicola (112Y), a common leaf saprophyte, has been reported as inhibiting apple blossom infection by Erwinia amylovora, the causal agent of fire blight of apples and pears. The chemoattractants of E. herbicola were determined in order to compare them to the attractants of E. amylovora. E. herbicola is attracted chemotactically to Jonathan apple nectar extract, specifically to the neutral and basic fraction of the extract. The neutral and basic fraction is a significantly better attractant than either the amino acid or organic acid fractions but not as good as unfractionated nectar extract. E. herbicola is attracted to a variety of compounds in contrast to E. amylovora, which is attracted to only a specific class of organic acids. The amino acids asparagine, serine, and tryptophan are the best attractants of E. herbicola with glucose and fructose being good sugar attractants and malate and tartarate being the best organic acid attractants.

A71

ADHESION OF EPIPHYTIC PHYTOPATHOGENIC BACTERIA TO LEAF SURFACES. C. A. Jasalavich and L. Sequeira, University of Wisconsin, Madison WI 53706, U. S. A.

Pseudomonas syringae lives epiphytically on plant surfaces, yet only certain pathovars cause disease in certain hosts. Degree of adhesion to external plant surfaces may be one factor that differentiates pathovars. In order to measure adhesion of P. syringae pv. lachrymans (Psl) or P. syringae pv. glycinea (Psg) to cucumber leaves, a binding assay was developed. Leaf discs were incubated separately in a buffer suspension of ^{14}C-labelled bacteria at 28°C for short periods (0 to 2 hr). After incubation leaf discs were transferred to buffer and washed for 10 min. at 28°C with shaking. Leaf discs, washes, and cell suspensions were digested separately, and counted on a Packard scintillation counter. In this assay the homologous pathovar Psl consistently bound to cucumber leaf discs to a greater extent than the heterologous pathovar Psg. These results demonstrate differential adhesion of pathovars to a plant surface.

A72

DIFFERENTIAL ADSORPTION OF AGROBACTERIUM TUMEFACIENS TO GRAPE TISSUE CULTURE CELLS. Greg L. Cleveland and Robert N. Goodman, Department of Plant Pathology, University of Missouri-Columbia, Columbia, MO 65211.

Adsorption of a virulent strain of Agrobacterium tumefaciens to grape suspension culture cells has been measured using a kinetic assay. When a biotype 3 strain of A. tumefaciens was incubated with callus suspension culture cells derived from a grape cultivar (Chancellor) susceptible to crown gall disease, 88% of the inoculum adsorbed to the callus cells. The same strain of biotype 3 showed no detectable binding to callus cells of a resistant cultivar (Seyval). The ratio of callus cells to bacteria was 12:1 for each assay. These preliminary results suggest that resistance of certain grape cultivars to crown gall disease may be due to the decreased ability of A. tumefaciens to adsorb to host cells. The adsorption of additional strains of A. tumefaciens, and A. radiobacter and Rhizobium spp to grape tissue culture cells will be discussed.

A73

IN SITU DETECTION OF THE MYCOPLASMALIKE ORGANISM CAUSING X-DISEASE IN RELATION TO SYMPTOM EXPRESSION. S. M. Douglas, The Connecticut Agric. Expt. Station, Box 1106, New Haven, CT 06504.

Levels of the mycoplasmalike organism causing X-disease (XMLO) were surveyed throughout the growing season in relation to symptom expression. Leaf mid-veins and petioles were sampled biweekly from peach trees (Prunus persica (L) Batsch) and chokecherry bushes (P. virginiana L.) of varying symptom severity. Samples were fixed with glutaraldehyde, sectioned, and stained with the fluorochrome DAPI (4'-6-diamidino-2-phenylindole). XMLOs were observed in all peach and chokecherry samples from trees which became symptomatic but were never detected in samples from trees which remained apparently healthy. XMLO-invasion of sieve tubes was greater in severely than mildly symptomatic peach, and was often evident up to 8 weeks before symptoms. In comparison, XMLO-invasion of symptomatic chokecherry was always earlier and more extensive than of peach. XMLOs were consistently detected before evidence of phloem necrosis. XMLO staining was verified by SEM and TEM.

A74

AMINO ACID UTILIZATION BY SPIROPLASMAS IN A CHEMICALLY DEFINED MEDIUM. C. Stevens and A. Patterson, Dept. of Agric. Sciences, Tuskegee Institute, AL 36088; and R. M. Cody and R. T. Gudauskas, Dept. Botany, Plant Pathology, and Microbiology, Auburn University, AL 36849.

Spiroplasma citri, S. floricola, SR-3, and AS576 metabolized arginine in a chemically defined medium. Deletion of amino acids from the complete medium (contained 19 amino acids) indicated that asparagine, glutamic acid, glycine, leucine, isoleucine, lysine, phenylalanine, proline, threonine, and valine also were important for growth of these spiroplasmas. In medium deficient in these amino acids but supplemented with arginine, glucose, or arginine + glucose, growth of spiroplasmas was greatest in the presence of arginine + glucose. For example, in lysine-deficient medium, numbers of S. citri cells/ml reached 0.1, 0.4, and 1.2 x 10^8 in medium containing arginine, glucose, and arginine + glucose, respectively.

A75

CELL WALL HYDROLYSIS OF HOST AND NON-HOST FUNGI DURING INTERACTION WITH THE MYCOPARASITE PYTHIUM NUNN. Y. Elad, R. Lifshitz and R. Baker. Department of Botany and Plant Pathology, Colorado State University, Fort Collins, CO 80523.

Localized degradation of hyphal cell wall of several hosts, by the mycoparasite,-Pythium nunn,-was observed with fluorescent microscopy following staining with Calcofluor White M2R New. P. nunn produced extracellular β-1,3-glucanase (glu), cellulase (cel) and chitinase (chi) in liquid culture containing laminarin, cellulose or chitin, respectively. Hyphae of Pythium ultimum, P. vexans, P. oligandrum, P. aphanidermatum and Pythium sp., induced high glu and cel activity by P. nunn. Hyphae of Mucor sp. and Rhizopus sp. induced chi production. P. nunn excreted chi and glu when grown on Rhizoctonia solani or Sclerotium rolfsii cell walls or with them in dual culture. Hydrolytic enzymes of P. nunn were not induced by Fusarium oxysporum f. sp. cucumerinum or by ten other nonhost deuteromycetes. Crude enzyme solutions of P. nunn released N-acetylglucosamine and glucose from trypsin- or KOH treated Fusarium hyphae.

A76

ROOT TIP COLONIZATION BY A PSEUDOMONAD SUPPRESSIVE TO THE TAKE-ALL DISEASE OF WHEAT. J.L. Parke*, R. Moen, A.D. Rovira and G.D. Bowen. Dept. of Plant Pathology, University of Wisconsin, Madison WI 53706* and CSIRO Div. of Soils, Glen Osmond, S.A. 5064 Australia.

Root tips of four day old aseptic wheat plants were inoculated with 10^5 CFU *Pseudomonas fluorescens* strain 2-79 resistant to rifampin and naladixic acid. Seedlings were transplanted into root chambers containing nonsterile soil premoistened to 17% gravimetric water (-0.6 bars). Following incubation at 15°C for periods up to one week, the rate of root elongation and vertical spread of 2-79 were measured. 2-79 populations means greater than the limit of detection (5 CFU per 2 mm root length) were present 0.2, 2.0, 6.0, and 1.8 cm below the point of inoculation at 0.2, 2, 4, and 7 days, respectively. Corresponding mean root elongation for these time intervals were 0.3, 2.4, 7.8 and 14.3 cm. We suggest that transport of 2-79 by the apex of the elongating root is inadequate to explain colonization of wheat roots observed in the field.

A77

CHARACTERISTICS OF A PSEUDOMONAS FLUORESCENS STRAIN INVOLVED IN SUPPRESSION OF BLACK ROOT ROT OF TOBACCO. Défago,G., Ahl, P., Berling,C.H., Stutz,E., Voisard,C., *Haas,D. and *Rella,M. Institut für Phytomedizin and *Institut für Mikrobiologie, Eidgenössische Technische Hochschule, ETH-Zentrum, CH-8092 Zürich.

Pseudomonas fluorescens strain Ao was isolated from tobacco roots growing in soil suppressive to Black Root Rot Thielaviopsis basicola. Strain Ao showed the following properties: (1)it caused suppressiveness when added to conducive soil; (2)on agar plates it inhibited the growth of T. basicola, Gaeumannomyces graminis, Fusarium oxysporum, F. solani, Rhizoctonia solani, Helminthosporium sativum, Alternaria alternata, and Pythium paroecandrum, probably by antibiotic production; (3)there was no evidence that inhibition of fungal growth was related to the Fe^{2+}/Fe^{3+} supply; (4)under sterile conditions strain Ao multiplied on the roots and could be isolated also from the inside of the stem and from the leaves which do not contain b-proteins. Spontaneous mutants from strain Ao resistant to streptomycin, nalidixic acid, rifampicin conserved the properties 1-3 listed above. Strain Ao could be mutagenized by a Tn 5 derivative.

A78

GERMINATION OF ASPERGILLUS FLAVUS SCLEROTIA IN SOIL. J. P. Stack and R. E. Pettit, Dept. of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843.

Studies on the survival and activity of A. flavus in natural soil involved a determination of the conditions which influence the germination of sclerotia. Sclerotia were placed at the surface or buried 2 cm deep in a sandy-loam soil (pH 6.7). Soil moisture levels were adjusted gravimetrically to 0, -0.1, -0.33, -1.0, -10, and -15 bars and the soil was held at 20, 30, or 35 C. With the exception of sclerotia buried 2 cm at -15 bars, there was abundant germination in all treatments. Sclerotia germinated in 4 days by the production of hyphae and/or several conidiophores with conidia. At -15 bars and 20 C, it took 13 days for the sclerotia to germinate. Hyphae from sclerotia grew up to 2 cm in 4 days to contact native soil organic matter. There was no apparent tropism with respect to conidiophore orientation; conidiophores grew from sclerotia down into the soil as well as upwards.

A79

A HAWAIIAN SOIL SUPPRESSIVE TO PHYTOPHTHORA CAPSICI. W. H. Ko and K. A. Nishijima, Department of Plant Pathology, University of Hawaii, Beaumont Agricultural Research Center, Hilo, HI 96720.

The method of Ko and Ho (Ann. Phytopathol. Soc. Japan 49:1-9, 1983) was used to detect Phytophthora capsici-suppressive soils. Sporangia of P. capsici were incubated in 50% V-8 juice for 1 h before being added to the smoothed surface of soil blocks to ensure uniform direct germination. A forest soil collected on the island of Hawaii was found to be suppressive to sporangial germination of P. capsici and damping-off of tomato seedlings caused by the pathogen. The soil remained suppressive to sporangial germination of P. capsici after autoclaving, or ignition at 500 C for 16 h. The inhibitory factor existed in the fractions of clay, silt and sand, but not in the aqueous extract of suppressive soil. Results suggest that inorganic compound might be responsible for the inhibitory effect of suppressive soil against P. capsici.

A80

FACTORS RESPONSIBLE FOR INHIBITION OF PYTHIUM SPLENDENS IN A SUPPRESSIVE SOIL IN HAWAII. C. W. Kao and W. H. Ko, Department of Plant Pathology, University of Hawaii, Beaumont Agricultural Research Center, Hilo, HI 96720

The Ca content in suppressive soil was about 60 times of that in conducive soil. Conducive soil amended with $CaCO_3$ or mixed microorganisms slightly decreased sporangial germination of P. splendens. However, when both $CaCO_3$ and mixed microorganisms were added to conducive soil, germination was decreased from 94% to 44%. Addition of $CaCO_3$ and alfalfa meal to gamma-irradiated conducive soil before inoculation with soil suspension rendered it strongly suppressive to germination of P. splendens after 3-day incubation. Stored suppressive soil contained about the same concentration of Ca as fresh suppressive soil, but was not inhibitory to sporangial germination of P. splendens. The soil became inhibitory to P. splendens when its microbial population was increased by addition of fungi, bacteria or actinomycetes. Results suggest that a combination of high Ca content and high total microbial population is responsible for inhibition of P. splendens in this suppressive soil.

A81

ABSENCE OF TAKE-ALL DECLINE UNDER WHEAT-SOYBEAN DOUBLECROPPING. C. S. Rothrock and B. M. Cunfer, Department of Plant Pathology, Georgia Station, University of Georgia, Experiment, GA 30212.

In the Southeast most small grain production is in a double-

cropping sequence with soybeans. In recent years take-all, caused by Gaeumannomyces graminis var. tritici, has been increasingly observed in Georgia. Soil suppressiveness was assessed in fields with long term wheat-soybean doublecropping. These fields had enough consecutive wheat crops for suppressiveness to have developed. Neighboring fields having short wheat cropping histories and a field with 15 years of wheat or barley and summer fallow were also included. Soil suppressiveness was assessed using a wheat seedling bioassay in which 1 or 10% of the test soil was transferred to a fumigated soil. No differences in percent plant infection were found between short and long term wheat doublecropping. Soil suppressiveness was found under the small grain-fallow cropping system. Results suggest that soil suppressiveness does not develop under doublecropping situations.

A82

ALLELOPATHIC PROPERTIES OF ASPARAGUS: INTERACTION WITH FUSARIUM SPP. AND BIOASSAY TECHNIQUES. A.C. Hartung and C.T. Stephens*, Departments of Horticulture and *Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824

Asparagus has been reported to release autotoxic substances from root tissue. These substances increase plant susceptibility to Fusarium oxysporum f. sp. asparagi and F. moniliforme, the causal agents of asparagus root rot. Greenhouse studies were conducted using measured amounts of dried root tissue incorporated into soil with either no fungus or in combination with millet infested with F. moniliforme or F. oxysporum f. sp. asparagi. Root rot was increased in the presence of increasing amounts of dried asparagus tissue. The chemical components which are believed to increase asparagus root rot are currently being isolated from dried asparagus root tissue. Different bioassay and chemical assay methods have been evaluated to identify a sensitive and rapid assay for quantifying the allelopathic properties of these substances.

A83

SURVIVAL POTENTIAL OF VERTICILLIUM ALBO-ATRUM IN SOIL. A. P. Keinath and R. L. Millar, Department of Plant Pathology, Cornell University, Ithaca, NY 14853-0331.

Persistence of V. albo-atrum (Vaa) was assessed after infected alfalfa root and stem segments had been buried 4 or 8 wks in nonsterile field soil (NSF) in petri plates. The soil was at -10 or -300 mb ψm and 5 or 21 C. Competitive saprophytic ability of Vaa in NFS was determined by assaying for colonization of dried healthy tissue (baits) located 5 mm from infected tissue (sources). Use of a benomyl-tolerant isolate (BT) and benomyl-amended selective medium precluded difficulties from indigenous benomyl-sensitive Verticillium spp. Recovery of BT from sources, regardless of temperature, was 81-100% for stem segments buried 8 wk at each ψm, but was < 30% for root segments buried 4 wk at -300 mb. Formation of dark resting mycelium and conidiophores was maximum for stems at 21 C and -300 mb. Colonization of stem baits was greater at -300 mb than at -10 mb, regardless of temperature. Root segments were rarely colonized.

A84

EFFECT OF DEPTH OF BURIAL AND SCLEROTIAL TREATMENT ON SURVIVAL OF SCLEROTIA OF SCLEROTIUM ROLFSII IN FIELD SOIL. V. L. Smith, Z. K. Punja, and S. F. Jenkins, Dept. of Plant Pathology, North Carolina State University, Raleigh 27695-7616.

Sclerotia from laboratory oat cultures were 1) dried 24 hr; 2) dried, washed 5 hr and redried; 3) not dried; 4) or produced in soil and dried. Four replicates of 50 sclerotia per treatment were placed in nylon mesh bags and buried in Sept 1983 in soil at six depths from 0-15 cm at two locations (GA and NC). Samples were retrieved monthly and percent viability determined. For treatments 1, 2, and 4, survival at depths > 7.5 cm after 1, 3, and 5 months was reduced to 15, 8, and 7 %, respectively, in GA and 26, 16, and 2 % in NC. At the soil surface, survival was 66, 50, and 51 % in GA and 86, 50, and 19 % in NC, respectively. Depth of burial and sclerotial treatment significantly affected survival (P=0.01) at all sampling times and at both locations. Survival was greatest in treatment 3. Fungi, primarily Trichoderma spp., were frequently isolated from nonviable sclerotia. Enhanced exudation of nutrients following drying or deep burial may contribute to reduced survival of sclerotia.

A85

CHARACTERIZATION OF HCN-PRODUCING SOYBEAN RHIZOSPHERE BACTERIAL ISOLATES. B. Hemming, D. Drahos, J. Brackin, C. Jonsson. Monsanto Co., 800 N. Lindbergh Blvd., St. Louis, MO 63167.

Bacteria obtained from roots of soybean plants from several locations were subjected to a battery of tests including an agar plate method for the detection of low levels of HCN (Castric K.F., 1983 Appl. & Environ. Micro. 45:701-2). The assay for HCN production was modified for rapid testing of large numbers of isolates. Results demonstrate the number of rhizosphere pseudomonads possessing the trait is large. By an R-matrix technique, clusters of associated traits have permitted identification of the most prevalent HCN-producing organisms as members of the *Pseudomonas fluorescens-putida* group. Examination of >400 fluorescent pseudomonad isolates from the rhizosphere has also revealed the constant absence of the o-nitrophenylgalactoside (ONPG) hydrolysis positive phenotypes, and the inability to grow on lactose as a sole carbon source. The lack of these traits has permitted the establishment of a unique marker system.

A86

β-GALACTOSIDASE, A SELECTABLE NON-ANTIBIOTIC CHROMOGENIC MARKER FOR FLUORESCENT PSEUDOMONADS. D. Drahos, B.Hemming, S.McPherson, J.Brackin. Monsanto Co., 800 N. Lindbergh Blvd., St. Louis, MO 63167.

Plasmids bearing *E. coli* lacZ and lacY genes have been constructed from derivatives of a broad host-range vector, RSF1010. These plasmids were maintained in fluorescent pseudomonad cells under rhizosphere conditions of an environmental chamber, and express β-galactosidase and lactose permease activities shown lacking in wild-type strains. In the presence of a chromogenic substrate (X-gal) colonies of these strains exhibit both their natural fluorescent pigment(s) and the blue coloration of the hydrolyzed substrate. In addition, lacZ and lacY gene products confer the metabolic capacity necessary for fluorescent pseudomonad growth on lactose minimal medium, atypical of wild-type isolates. This provides a selectable non-antibiotic marker for tracking the organism or plasmids in the environment. Further, expression has been regulated in these pseudomonads by linking the lacZY coding sequence to several controllable promoters.

A87

A GEMINIVIRUS-LIKE PARTICLE FROM OATS. Steve Haber & C.C. Gill, Agriculture Canada, 195 Dafoe Rd., Winnipeg, Manitoba R3T 2M9

In addition to the expected particles of barley yellow dwarf virus (BYDV), particles resembling geminiviruses were isolated from Coast Black oats and Avena fatua inoculated with a BYDV strain (RMV-type) vectored specifically by the aphid Rophalosiphon maidis. The geminivirus-like particles sedimented in sucrose gradients as a broad peak at 72 S. Electron microscopy of the 72 S peak fractions by negative staining with phosphotungstic acid pH 7.0 revealed predominantly geminate and some 'triplet' particles;the proportion of triplet particles was lowest in the slower-, highest in the faster-sedimenting fractions of the peak. The geminate particles measured 13 x 22 nm, the triplets 13 x 33 nm. The major protein species isolated from the 72 S peak fractions was 27.7 kd. Aphids of five differential vector species of BYDV strains induced chlorotic striping in Coast Black oats after membrane feeding on sucrose solutions from 'geminivirus'peak fractions and 'geminivirus'-conbined-with-BYDV fractions, but not after feeding on control solutions or solutions from BYDV fractions alone.

A88

GEMINIVIRUS-LIKE NUCLEAR INCLUSIONS ASSOCIATED WITH PSEUDO-CURLY TOP DISEASE IN FLORIDA. R. G. Christie, B. W. Falk, N.-J. Ko, and F. W. Zettler. University of Florida, Depts. Agronomy and Plant Pathology, Gainesville 32611 and AREC-Belle Glade 33430.

Pseudo-curly top (PCT) infects tomatoes and other plants in FL (Simons and Coe. 1958. Virology 6:43), is unique in having a treehopper (Membracidae) vector, and induces symptoms in tomatoes like those of beet curly top, a leafhopper-borne geminivirus. PCT was maintained in Nicotiana x edwardsonii and Solanum nigrum and routinely transferred to healthy plants by Micrutalis malleifera treehoppers. PCT-infected leaf tissues of both species stained in azure A for light microscopy revealed conspicuous nuclear inclusions like those induced by 4 whitefly-borne geminiviruses (Christie and Bird. 1984. Phytopathology 74:in press). Likewise, virus particles and "fibrillar bodies" as described for whitefly-borne geminiviruses by Kim and Fulton (1984. Phytopathology 74:236) were seen in the nuclei of thin-sectioned tissues examined by electron microscopy. This is the first report of 1) the viral etiology of PCT and 2) a possible treehopper-transmitted geminivirus.

A89

HEAT-STRESSED TOMATO PLANTS CONTAIN AN RNA BAND THAT COMIGRATES WITH POTATO SPINDLE TUBER VIROID IN NON-DENATURING POLYACRYLAMIDE GELS. M. E. Grasmick[1], A. Branch[2], S. A. Slack[1], and H. Robertson[2]. Depts. of Plant Pathology, University of Wisconsin-Madison 53706[1], and Genetics, Rockefeller University, New York, NY 10021[2].

Nucleic acid extracts of Rutgers tomato plants grown 8 wks $\geq$34 C and with $\geq$650 $\mu Em^{-2}s^{-1}$ supplemental light contained an RNA (s-RNA) that comigrated with PSTV upon electrophoresis in non-denaturing polyacrylamide gels (PAGE). Stressed plants appeared normal, but non-stressed tomato plants developed mild PSTV-like symptoms after 4 sequential subinoculations. The s-RNA band in PAGE was faint in stressed tomato plants, but was distinct in subinoculated plants. No circular RNAs were detected in samples from stressed plants. Fingerprint analyses of s-RNA from stressed plants revealed a mixture of RNAs, but PSTV was not detected. Data indicate a unique response of Rutgers tomato to heat stress and suggest caution in the interpretation of PAGE assays.

A90

MOLARITY AND pH EFFECTS ON FIVE BARLEY YELLOW DWARF VIRUS ISOLATES. S. A. Slack, University of Wisconsin-Madison 53706; W. F. Rochow, USDA/ARS, Cornell University, Ithaca, NY 14853; and H. T. Hsu, ATCC, Rockville, MD 20852.

Partially purified preparations of the MAV, RPV, RMV, PAV, and SGV isolates of BYDV were tested at ionic strengths of 0.001 M-1.0 M in KPO_4 (pH 7) and at pH 2.5-8.0 in 0.1 M buffers. Treatments were evaluated by enzyme-linked immunosorbent assays within 4 h and after 8 days. Selected treatments were also tested by aphid transmission following virus acquisition through Parafilm membranes. The MAV, RPV, and SGV isolates were most stable at 0.01 and 0.1 M. The RMV isolate was favored by 0.1 and 1.0 M and PAV was most stable at 0.01 M. All isolates were most stable at pH 6 or 7 in KPO_4 or $NaPO_4$. All isolates were completely dissociated within 15 min at pH 2.5. Immediate detrimental effects that increased with time were noted at pH 3-5 and 7.5-8.0. Triton X-100 (0.5%) enhanced stability, but 0.1 M TRIS caused complete dissociation at pH 7-9. This information was utilized to develop strategies for elution of BYDV from monoclonal antibody-labelled Sepharose affinity columns.

A91

CHARACTERIZATION OF THE COAT PROTEINS OF CITRUS TRISTEZA VIRUS. R. F. Lee, L. A. Calvert, and J. D. Hubbard*, Univ. of Florida, Citrus Research and Education Center, Lake Alfred, FL 33850 and *U.S. Grain Marketing Research Lab., USDA, SEA-AR, Manhattan, KS 66502.

Several aphid-transmitted isolates of citrus tristeza virus (CTV) which vary widely in their biological properties were purified and the coat proteins examined. By sodium dodecyl sulfate polyacrylamide gel electrophoresis, a major coat protein (CP1) and a minor coat protein (CP2) with estimated molecular weights of 23,000 and 21,000 daltons, respectively, were found in all Florida isolates. Both CP1 and CP2 react with CTV specific antisera. The amino acid composition of purified coat proteins was determined and both were low in methionine and high in lysine, aspartic acid, and glycine. CP1 and CP2 were digested with trypsin, V8 protease, or thermolysin and the resultant polypeptide maps were similar.

A92

THE EFFECT OF COAT PROTEIN OF CHALLENGE TMV ON SUPERINFECTION OF BEAN INFECTED BY TMV-LEGUME. Thomas M. Zinnen and R. W. Fulton, Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Cross protection by TMV-legume (TMV-L) in systemically invaded Phaseolus vulgaris cv. Pinto was weak against TMV-common (TMV-C). TMV-C RNA encapsidated in TMV-C coat protein caused 0.2 - 0.9 times as many lesions in TMV-L infected Pinto as in healthy Pinto. TMV-C RNA encapsidated in TMV-L coat protein, however, caused only 0.01 - 0.07 times as many lesions in TMV-L infected Pinto as in healthy Pinto. The addition of TMV-C coat protein (1 mg/ml), but not TMV-L coat protein, to TMV-C inoculum decreased its infectivity for healthy Pinto and for TMV-L infected Pinto. The two viruses differ serologically. Apparently one of the factors determining specificity of cross protection in this system is the coat protein of the challenge virus.

A93

THE PRESENCE OF VIRAL ANTIGEN IN THE APOPLAST OF SYSTEMICALLY INFECTED PLANTS. **G. A. de Zoeten and G. Gaard. Dept. of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706.**

Cell walls were isolated from healthy and TMV-U_1-infected Nicotiana sylvestris plants. Antigen in enzyme digested cell walls from light green (LG) and dark green (DG) areas of infected leaves was quantified with ^{125}I-labeled antibody. Bound ^{125}I-TMV-U_1-IgG/mg cell wall ranged between 1 and 1.5 µg for LG tissues (unfixed, acetone washed) and from 0.2 to 0.5 µg for DG tissues. These results were confirmed by autoradiography with ^{125}I-anti-TMV-U_1-Fab fragments of IgG in electron and light microscope examinations. Quantitative and qualitative differences in uncoating of virus in healthy and virus-infected tissues were established.

A94

REACTION OF PROTOCLONES OF POTATO TO POTATO VIRUS Y. H.H. Murakishi, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

Protoclones regenerated from leaf mesophyll protoplasts of potato cv. Russet Burbank, considered to be a systemic host of potato virus Y (PVY), were transplanted to soil and inoculated with a common strain (Y^O) of PVY. The inoculated plants were screened for the presence or absence of PVY by enzyme-linked immunosorbent assay (ELISA). Clones which appeared free of virus were selected for reinoculation and virus assay. Of 300 protoclones tested, five showed little or no virus after 2-3 inoculations over a 2-month period. Cuttings were taken from each of the clones which showed apparent resistance and were rooted in soil. Assay of upper leaves one month after inoculation of lower leaves showed that four of the five subclones had very little or no virus. A susceptible subclone developed high virus titer within two weeks.

A95

DOUBLE-STRANDED RNA (dsRNA) BANDING PATTERN CHANGES IN HYPOVIRULENT ENDOTHIA PARASITICA. D. W. Fulbright and S. W. Garrod, Department of Botany and Plant Pathology, Michigan State University, E. Lansing, MI 48824-1312.

Strains of E. parasitica from American chestnut trees in Michigan recovering from chestnut blight contain cytoplasmic dsRNA. Wide variations in the dsRNA banding patterns observed after gel electrophoresis can exist from strain to strain. These patterns are reproducible and are correlated to specific culture morphology and virulence phenotype of the pathogen. However, deviations from the expected dsRNA banding pattern occurred in one strain (GHU4) after single spore isolation or growth on media with cycloheximide (10 µg/ml). Concomitant changes were observed in morphology and virulence. Banding pattern changes were also noted in another strain, CL1(GH2) after it was released into the environment and recovered 12 months later. Changes also occurred when CL1(GH2) was infected with dsRNA from GHU4. Mixtures of dsRNA segments from both strains were isolated from the cytoplasm of the infected strain and its conidia, but some segments were lost.

A96

CROSS PROTECTION BETWEEN STRAINS OF CUCUMBER MOSAIC VIRUS. J. A. Dodds and S. Q. Lee, Department of Plant Pathology, University of California, Riverside 92521.

Two strains of cucumber mosaic virus (CMV) differed in three characteristics of value for cross protection experiments. Their virions and also their dsRNAs could be separated and distinguished by electrophoresis on polyacrylamide gels, and the symptoms of one strain were milder than the other in tobacco, tomato and squash. The mild strain, CMV-S, protected plants of these three hosts from the effects of the second strain, CMV-P, and also prevented the accumulation of virions and dsRNAs of the challenge strain. Protection was detected in leaves inoculated with the challenge strain and also in later formed leaves. The only exception to this result was the accumulation of dsRNAs and to a lesser extent virions of the challenge strain when infectious viral RNA was used as the challenge inoculum instead of virus particles. This breakdown of cross protection only occurred in those leaves inoculated with the challenge strain RNA. No accumulation of challenge RNA or virions occurred in later formed leaves.

A97

HONEYBEE-MEDIATED TRANSMISSION OF BLUEBERRY LEAF MOTTLE VIRUS VIA INFECTED POLLEN TO HIGHBUSH BLUEBERRY. A.M. Childress and D.C. Ramsdell, Dept. of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

The blueberry leaf mottle virus (BBLMV) is classified as a

putative member of the nepovirus group. This group of viruses are primarily spread by nematodes and secondarily by pollen. An association between BBLMV and a nematode vector has not been found. Spread occurs in a random manner in the field, uncharacteristic of a nematode vector. Using ELISA and RIA, BBLMV particles were found associated with pollen collected from infected plants and pollen baskets of honeybees, Apis mellifera. Serological blocking techniques revealed virus both on the inside and outside of the pollen grains. Transmission of BBLMV was obtained by rub-inoculating infected pollen to Chenopodium quinoa. To determine whether the virus is transmitted via pollen, infected caged and noncaged source plants were encircled by 2-yr-old healthy blueberry trap plants. During bloom, honeybee hives were located within some cages and near to uncaged source and trap plants.

A98

PROTEOLYSIS OF TURNIP YELLOW MOSAIC VIRUS TOP COMPONENT. Rose N. Skopp and Leslie C. Lane, Dept. of Pl. Path., University of Nebraska, Lincoln, NE 68583-0722.

The capsid of TYMV is extremely stable. We do not yet understand how it assembles or how the RNA escapes during infection. Trypsin specifically cleaves a portion of the coat protein of both artificial and natural top components of TYMV. The cleavage appears to be at arginine 12. Cleavage produces a class of particles which electrophorese as a discrete band, with lower net charge than cleaved particles. The thermal stability of cleaved particles is similar to that of uncleaved particles. Roughly 50 percent of the protein resists cleavage. A similar process could be involved in releasing the RNA during infection.

A99

BIOLOGICAL EVALUATION OF BARLEY STRIPE MOSAIC VIRUS HYBRIDS. M. C. Edwards and R. G. Timian, Dept. of Plant Pathology, North Dakota State University, Fargo, North Dakota 58105.

Four BSMV strains which differ in pathogenicity, symptomatology, and seed transmission were used in pseudorecombination experiments. CV35(ND131) and CV52(ND18) are both pathogenic to barley cultivars Silver King (CI 905), Modjo 1 (CI 14048), and Moreval (CI 5724), but not to oats. CV40(ND161) and CV42(ND159) are pathogenic to oats, but not to the above mentioned barley cultivars. CV40 and CV52 are also known to differ in seed transmissibility. To determine the RNA species responsible for the characters tested, all possible combinations of purified RNA were first inoculated onto Black Hulless barley (CI 666) and later transferred to differential hosts. Experiments with pseudorecombinants constructed from CV40 and CV52 indicate that RNA 1 is responsible for pathogenicity to oats and Modjo 1 barley. Experiments with CV35 and CV42 support these conclusions and further indicate that RNA 1 is responsible for pathogenicity to Moreval and Silver King barleys. Preliminary evidence also indicates that at least RNA 1 is involved in seed transmission.

A100

MONITORING OF MAIZE DWARF MOSAIC VIRUS INFECTION AND SPREAD IN CORN LEAVES VIA IMMUNOFLUORESCENCE. J. D. Lei and G. N. Agrios, University of Massachusetts, Amherst, 01003.

The number of infection loci and the spread of maize dwarf mosaic virus in symptomless inoculated leaves of resistant (PA405 and BSQ) and susceptible (MA5125) corn lines was monitored by a new immunofluorescent procedure. Inoculated leaves were brushed with carborundum, treated with cellulolytic enzymes, fixed with acetone, treated with virus-specific and then with an FITC-conjugated antibody, and observed under an epi-fluorescent microscope. Infected cells were observed in all three lines 24 hrs after inoculation. More infection sites developed per leaf area unit and reached larger sizes in BSQ than in the other lines. Secondary infection loci appeared along veins 6 days after inoculation in MA5125 and BSQ and after 10-11 days in PA405. MA 5125 had significantly more secondary infection loci than BSQ or PA405. In inoculated leaves of MA5125 the infection spread faster and towards the stem while in BSQ and PA405 it spread slowly and equally towards the stem and the leaf tip.

A101

HISTOPATHOLOGY OF LOBLOLLY PINE NEEDLES INFECTED BY SCIRRHIA ACICOLA (Dearn.) Siggers. F.F. Jewell, Sr., School of Forestry, Louisiana Tech University. Ruston, LA 71272.

Field-grown loblolly pine (Pinus taeda L.) needles with and without typical bar-spot or yellow/brown spot symptoms of Scirrhia acicola, the brown-spot needle blight pathogen were collected. Tissue samples were fixed, embedded, sectioned serially, stained, and examined by light microscopy. Tissues from non-symptom samples were normal for P. taeda. Symptom-bearing samples, regardless of type, exhibited a similar major host reaction, confined to the symptom area, and consisting of a collapse, not dissolution, of the mesophyll cells. Longitudinally, affected tissue exhibited a lattice-like appearance devoid of cell contents. Reaction in bar-spot symptoms was limited to the mesophyll. S. acicola hyphae were infrequent. Other symptom-types exhibited a lesion-forming type host reaction in the endodermis but seldom into the vascular system, with frequent and prominent hyphae. Fumago sp. was consistently observed, particularly in stomata, on all non- and symptomatic tissue, while Lophodermium sp. was a frequent associate on symptomatic tissue.

A102

Chemical control of Diplodia shoot blight in forest tree nurseries. M. A. Palmer, T. H. Nicholls, and C. F. Croghan; USDA Forest Service, 1992 Folwell Ave., St. Paul, MN 55108.

Diplodia shoot blight, caused by Sphaeropsis sapinea (Fr.) Dyko & Sutton, is a periodic problem of Pinus resinosa Ait. seedlings in forest tree nurseries in the north central United States. Fungicide evaluations performed 1981-1983 demonstrated that benomyl controlled this disease. Two applications at 14-day intervals during August reduced infection from 34% to 4% in 1981 in rising 1-year-old (1-0) seedlings, although somewhat better control was achieved with 5 or 7 applications from July through August or June through August, respectively. Severe infection occurred in both the first and second years of growth when 1-0 seedlings were not protected. Four applications at 14-day intervals during bud break and shoot elongation (late April-early June) reduced second-year infection from 33% to 4% in previously infected seedbeds. Infection of 3-year-old seedlings was reduced using a similar application schedule.

A103

SUSCEPTIBILITY OF DIFFERENT AGE SLASH PINE SEEDLINGS TO FUSIFORM RUST IN THE GREENHOUSE. C. H. Walkinshaw, Southern Forest Experiment Station, Gulfport, MS 39505; and F. F. Jewell, Sr., School of Forestry, Louisiana Tech. Univ., Ruston, LA 71272

One- and two-year-old resistant and susceptible seedlings of slash pine (Pinus elliottii var. elliottii) exhibited as high an infection with fusiform rust as 6-week-old seedlings. Spring and summer shoots, as well as terminal and lateral shoots, on 2-year-old seedlings appeared equally susceptible. Typical purple spots appeared on needle bases regardless of age or type of shoot. Galls formed at the same time on small and large seedlings but grew at a faster rate on larger ones. Pycnia and aecia production occurred at identical times for all ages. It is concluded that disease development correlates well between seedlings used in our screening tests (6 to 8 weeks old) and larger ones which were thought to be similar to susceptible seedlings in the field. Moreover, histology of galls on larger seedlings showed disease development to parallel the course previously described for 6-week-old seedlings.

A104

TEMPORAL AND SPATIAL PATTERNS OF FUSIFORM RUST INCIDENCE IN FIVE-YEAR-OLD PINE PLANTATIONS. R. C. Holley, M. C. Klapproth, and R. A. Schmidt. School of Forest Resources and Conservation, University of Florida, Gainesville, FL 32611.

Temporal and spatial patterns of fusiform rust incidence (% trees with rust) were analyzed for ca. 1500 five-year-old slash and loblolly pine plantations established from 1963-79 by Container Corp. of America in eight management areas in the N. Fla. and Ga. Coastal Plain. Five management areas had low rust incidence and three had high incidence. Areas with low incidence showed no disease increase over the 17-year period. In areas with high incidence, disease increased prior to 1970 (with considerable year-to-year variation); rust was significantly less in later years following utilization of rust-resistant seed sources. Within the natural range of slash pine, loblolly generally had more rust than slash; within the range of natural loblolly, slash had more rust than loblolly. Analysis of site factors will seek to delineate rust hazard.

A105

EFFECTS OF TWO SYSTEMIC FUNGICIDES AS SEED DRESSINGS ON FUSIFORM RUST OF LOBLOLLY PINE SEEDLINGS. W. D. Kelley and J. C. Williams, Dept. of Botany, Plant Pathology, and Microbiology, Ala. Agric. Exp. Stn., Auburn University, AL 36849.

Both triadimefon and triadimenol decreased the incidence of fusiform rust on emerging loblolly pine seedlings when applied

as seed dressings at rates of 0.31, 0.62, and 1.25 g ai/kg of seed. Triadimefon was superior to triadimenol in providing protection at all rates tested, and the degree of protection for each compound decreased as seedling age increased. The most drastic decreases in protection with increasing ages were observed among triadimenol treatments. Each of the compounds was compatible at all rates tested with the animal repellent and fungicide, thiram. Results indicate that triadimefon applied as a seed dressing at a rate of 1.25 g ai/kg of seed provided protection equal to that of the triadimefon seed soak (800 mg triadimefon/l, 24 h seed soak) currently being used by forest tree nursery personnel; either method protected seedlings for at least 36 days after sowing.

A106

ARTIFICIAL INOCULATION OF JACK PINE WITH PINE OAK RUST. R.A. Dietrich, W.K. Stewart, and R.A. Blanchette. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

Six-week-old seedlings of jack pine (Pinus banksiana Lamb.) were inoculated with Cronartium quercuum (Berk.) Miyabe ex Shirai f. sp. banksianae using a controlled basidiospore spray technique. Purple lesions were visible on the hypocotyls of 74% of the seedlings 2.5 weeks after inoculation. Fourteen weeks after inoculation, 65% of the stems had swellings and incipient galls. Cortical cells of seedlings collected 3 weeks after inoculation were distorted and occluded. Hyphae were observed from the cortex to the xylem. Seedlings collected 11 weeks after inoculation were colonized by the fungus from the cortex to the pith. Infected xylem had an increased number of ray parenchyma cells and also had trachieds that were shorter and wider than uninfected xylem. Artificial inoculation of jack pine has potential use for the rapid screening of jack pine for rust resistance.

A107

INOCULATION OF PINUS MONTICOLA TISSUE CULTURE PLANTLETS WITH VEGETATIVE AXENIC CRONARTIUM RIBICOLA. Alex M. Diner, Biosource Institute and Dept. of Forestry, Michigan State University, Houghton, MI 49931

Pinus monticola plantlets generated by cotyledon culture were inoculated in vitro using vegetative hyphae of Cronartium ribicola grown from basidiospores in axenic culture. Host tissue invasion reliably followed application of young, actively growing fungal colonies to stem tip wounds. Lower stem wounds to which colonies were similarly applied, became quickly flooded with wound exudates and did not become colonized. Colonization of intact needles was infrequent, and occurred by chance penetration of stomates. All instances of successful inoculation showed intracellular haustoria. P. monticola plantlets cloned in culture and inoculated with an axenically grown, vegetative pathogen offer opportunities for rapid, in vitro appraisals of tissue or organ-specific mechanisms of disease resistance.

A108

ULTRASTRUCTURAL HISTOPATHOLOGY OF EASTERN COTTONWOOD CLONES RESISTANT OR SUSCEPTIBLE TO LEAF RUST. L. Shain and U. Järlfors. Dept. of Plant Pathology, Univ. of Kentucky, Lexington, KY 40546.

Leaf disks from field-grown eastern cottonwood (Populus deltoides) clones, resistant or susceptible to Melampsora medusae, were inoculated artificially with urediospores. The leaf tissue was prepared for electron microscopy one through six days after inoculation. At six days uredia were being produced on the susceptible clone. Haustoria were observed in both clones and only minor differences were observed in host cells on day one. By day two, however, almost all haustoria-containing cells in the resistant interaction were necrotic and finely granular, the vacuole had disappeared, and most organelles had disintegrated. Haustoria were small and their contents appeared disorganized. Haustoria were larger and survived up to six days in the susceptible clone. Granular cytoplasm similar to that of infected cells of the resistant clone was seen rarely and much later in the susceptible clone.

A109

A NEW STEM RUST EPIDEMIC OF PINUS OOCARPA IN GUATEMALA. R.S. Webb. School of Forest Resources and Conservation, University of Florida, Gainesville, FL 32611

A new form species of Cronartium quercuum has been associated with an epidemic of stem infections of young Pinus oocarpa seedlings. Symptoms in P. oocarpa seedlings occur as swellings at the lower stem at the ground line and continuing below ground as a greatly thickened taproot. Adventitious shoots just above the basal swellings are present usually. Branch galls occurred infrequently on young branches of older pines (>10 yrs) yet no stem galls on these trees were observed. Disease incidence in Instituto Nacional Forestal (INAFOR) nurseries exceeded 80% in central and western Guatemala. Infection of young naturally-regenerated seedlings (<3 yrs) in the mountainous sections of the country also frequently exceeded 80% incidence. Since pine reforestation depends primarily upon natural regeneration, this epidemic on P. oocarpa seedlings is particularly threatening and may remove this species in time from its natural range in Guatemala.

A110

RUSTS OF THE SINO-HIMALAYAS. Mo-Mei Chen, Plant Pathology, University of Wisconsin, Madison, WI 53706

I have been a member of the Chinese scientists Qinghai-Xizhang investigation team and have made pathological and fungal investigations in Tibetan forests at altitudes between 1000-5000 meters among the highest peaks of the Himalayas (8812-8848 m). Over 800 specimens of rusts and polyporus were collected and I described 8 new species of rusts between 1979-1981. The region comprises a comprehensive spectrum of vertically-distributed forest. In this rare natural laboratory, very good conditions exist for the ecological study of rust fungi. Named the Sino-Himalayas rust flora, rust fungi in this group have been assigned to 20 genera and classified into three types according to the warmth index: 1) the plateau type; 2) the temperate zone type; and 3) the sub-tropical zone type. Each contains species which are uniquely characteristic. These regions have a place of origin for a new rust type and the rust flora has affinities to rust in other regions. Thus, it is possible for this rust flora to constitute a potential source of inoculum for new forest diseases.

A111

ASSOCIATION OF BACTERIA WITH TARNISHED PLANT BUG STEM LESIONS ON HYBRID POPLARS IN ONTARIO. J. Juzwik and M. Hubbes, Faculty of Forestry, University of Toronto, Toronto, Ontario, Canada M5S 1A1.

The association of bacteria with different stages of tarnished plant bug (TPB), Lygus lineolaris (Hemiptera:Miridae), lesions on young hybrid poplar shoots was investigated. In isolation attempts from 140 stem sections, bacteria were always recovered from degraded tissue of large and mid-sized TPB lesions, less frequently from small lesions and bumps, and rarely from healthy tissues. All 209 bacterial isolates obtained were gram-negative rods; 61 were members of the Enterobacteriaceae. By light and scanning electron microscopy, large numbers of bacteria were found within pockets of thick-walled fibers in stylet-disrupted tissues of bumps and large lesions. Evidence suggests that bacteria contribute to degradation of tissues through breakdown of the fibers. However, the role of bacteria in TPB lesion formation is still not clear.

A112

CULTURE OF FASTIDIOUS, XYLEM-LIMITED BACTERIA FROM DECLINING OAKS IN THE NORTHEASTERN STATES S.J. Kostka, J.L. Sherald*, and T.A. Tattar, Shade Tree Labs, Dept. of Plant Pathology, University of Massachusetts, Amherst, MA 01003 and *Ecological Services Laboratory, Natl. Park Serv., Washington, DC 20242

During the summers of 1982 and 1983, surveys were conducted to determine the range of bacterial-associated leaf scorch and decline of oaks from northern Virginia to Massachusetts. Affected oak species included red scarlet, and pin oaks. Symptomatic trees were observed from northern Virginia north to New York City. In one mixed planting of red and scarlet oaks in Wilmington, DE, 58 of 116 trees were affected. Wood chips were aseptically excised from stems of 28 symptomatic trees from VA, DC, MD, DE, NJ, PA, NY and incubated in a modified PW broth medium. Turbidity developed in all cultures after 2-6 weeks incubation at 28 C. Subcultures were maintained on modified PW agar medium. Bacteria, isolated from affected trees throughout the observed range, were immunofluorescent positive against antisera to the elm leaf scorch bacterium and the Pierce's disease bacterium.

A113

SUPPRESSION OF ELM LEAF SCORCH SYMPTOMS WITH OXYTETRACYCLINE S.J. Kostka, T.A. Tattar, and J.L. Sherald, Shade Tree Labs., Dept. of Plant Pathol., Univ. of Massachusetts, Amherst 01003 and Ecol. Serv. Lab., Natl. Park Serv., Washington, DC 20242

Elm leaf scorch (ELS) affected trees were injected with oxytetracycline (OTC) in Aug., 1982 and June 1983. Presence of the ELS-bacterium in affected trees was confirmed in 1982. OTC was injected via Mauget capsules into 10 trees at 40 mg and 50 mg a.i./cm dbh in 1982 and 1983 respectively. Thirteen trees were injected at the same rates using pipette injection in 1982 and Mauget capsules in 1983. Six trees were injected at 80 mg a.i./cm dbh by pipette injection (1982) and Mauget capsules (1983). Ten untreated, symptomatic trees served as controls. In July, 1983, 7 of 10 control trees and 3 OTC-treated trees developed ELS symptoms. By late Aug., symptoms were absent or reduced in 22 of 23 trees treated at the low OTC levels and 3 of 6 treated at the high OTC level, while controls were at or above 1982 severity. Suppression of ELS symptoms by OTC supports the hypothesis that the associated fastidious, xylem-limited bacterium is the primary causal factor of ELS.

A114

PARTIAL PURIFICATION OF ASH YELLOWS AND ELM YELLOWS MYCOPLASMA-LIKE ORGANISMS FROM INFECTED SYMPTOMATIC PERIWINKLE TISSUE. J. D. Castello, P. Shiel, J. A. Austin, F. Jones, C. Craft, and G. Delgado. State University of New York, College of Environmental Science and Forestry, Syracuse, NY 13210.

Leaf tissue from periwinkle [Catharanthus roseus (L.) G. Don] displaying characteristic yellows symptoms and infected either with the elm yellows or ash yellows mycoplasma-like organism (MLO) was purified through the first high speed centrifugation according to the protocol of Sinha (Phytopathology 64:1156-1158). Electron microscopy of resuspended pellets revealed numerous pleomorphic bodies up to approximately 1 micron in length. Pellets that were thin sectioned and examined by electron microscopy also revealed numerous pleomorphic bodies containing ribosome-like granules and surrounded by a characteristic trilaminar unit membrane. Both the ash and elm yellows MLO's were partially purified from at least six batches consisting of 100-150g of infected, symptomatic periwinkle leaf tissue. MLO's were not detected in tissue collected from asymptomatic plants and purified in the same manner.

A115

ASSOCIATION OF HYPOVIRULENT ENDOTHIA PARASITICA WITH AMERICAN CHESTNUT IN FOREST CLEARCUTS AND WITH MITES. G. J. Griffin*, R. A. Wendt*, and J. R. Elkins**. Dept. Plant Pathology, Physiology & Weed Science, VPI&SU*, Blacksburg, VA 24061 and Div. of Natural Sciences, Concord College **, Athens, WV 24712.

In preliminary studies, 3 of 27 (11.1%) strains of E. parasitica recovered from American chestnut sprouts in forest clearcuts were hypovirulent in pathogenicity trials, while 5 of 171 (2.9%) strains recovered from sprouts in understory, pole-timber sites were hypovirulent. Hypovirulent strains containing dsRNA were found in clearcuts. We hypothesize that maintaining American chestnut in forest clearcuts with minimal competition from other trees (by cutting or selective thinning of competing trees) may favor the natural development of hypovirulent E. parasitica on American chestnut sprouts from inoculum initially present or produced in nature. Three of 31 E. parasitica strains recovered from mites that were isolated from cankers on six large, surviving American chestnut trees were hypovirulent in pathogenicity trials.

A116

INTERACTIONS BETWEEN POWDERY MILDEW, TILLETIOPSIS WASHINGTONESIS AND NON-TARGET ORGANISMS ON CUCUMBER. Hartmann, H., W.A. Riggs and J.W. Hall. Saanichton Research & Plant Quarantine Stn., Sidney, B.C., M.B. Research & Development Ltd., P.O. Box 263, Saanichton, B.C. Research Station, Agric. Canada, Vancouver, B.C.

Four tests were carried out to identify volunteer organisms which interact with powdery mildew (PM) and Tilletiopsis washingtonesis (Tw) on cucumbers. Sampling was carried out on 4 occasions per experiment, spaced 72 hr apart. Organisms were sampled by shaking leaves in broth and plating. Numbers of colony forming units /cm^2 of leaf were determined for each organism isolated. Fifteen fungi, 7 yeasts and 21 bacteria were isolated. Organisms were indifferent to or increased in the presence of PM alone; only Rhizobium decreased with PM and medium combination; Azotobacter increased while the rest were either unaffected or decreased in the presence of Tw and medium; Alcaligenes and Azotobacter increased with PM-Tw combination while the rest declined or remained unaffected.

A117

ESTABLISHMENT OF DICYMA PULVINATA IN CERCOSPORIDIUM LEAFSPOT OF PEANUTS: INTERACTIONS OF SPRAY FORMULATION, INOCULATION TIME, AND TEMPERATURE. James K. Mitchell and Ruth A. Taber, Department of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843.

In growth chamber experiments at 25 C, visible signs of Dicyma pulvinata(Dp) colonization of Cercosporidium personatum(Cp) lesions appeared within 58-65 hrs(21-32 hrs leaf wetness) after application. Spores of Dp suspended in either H_2O, 0.2-0.3% carboxymethyl cellulose(CMC), 0.2-0.4% citrus pectin, or 0.25% ghatti gum colonized more Cp lesions and at a faster rate when Cp lesions on peanut leaves were inoculated with Dp at the beginning of the day cycle. CMC suspended spores exhibited the least amount of variability between day vs night inoculation. In broth culture, maximal growth(dry weight) of Dp occurred at 23-25 C. On detached peanut leaves infected with Cp, maximum colonization of Cp by Dp occurred at 23-28 C. No colonization was apparent at constant temperatures above 30 C.

A118

THE EFFECT OF STEM INJECTIONS WITH PERONOSPORA TABACINA AND METALAXYL TREATMENT ON GROWTH OF TOBACCO AND PROTECTION AGAINST BLUE MOLD IN THE FIELD. S. Tuzun, W. Nesmith and J. Kuć, Department of Plant Pathology, University of Kentucky, Lexington KY 40546.

Sporangial suspensions of P. tabacina (Ky isolate 82) were injected into stem tissue, external to the cambium, of plants growing at three field locations during 1983-1984: Kentucky 14 burley tobacco in Lawrence Co., Ky, MS 14XL8 burley hybrid in Owen Co., Ky and PR 5-65 cigar tobacco planted in Gurabo, Puerto Rico. Metalaxyl was added to the foliage and soil of comparable plants at the time of stem injection and untreated plants served as controls. Treatments were arranged into a randomized complete block design with three or more replications. Plants receiving the stem injection had significantly greater height and fresh weight than plants treated with metalaxyl or control plants. Marketable yield was increased by up to 25% over the controls at the two Ky locations. Stem-injected plants were protected against natural blue mold infections equal to or better than metalaxyl-treated plants in Puerto-Rico.

A119

ANTAGONISTIC ACTIVITY OF THREE IMPERFECT FUNGI TOWARDS THE MUMMY BERRY FUNGUS, MONILINA VACCINII CORYMBOSI Caruso, F.L. and M.G. Zuck, Department of Botany and Plant Pathology, University of Maine, Orono, ME 04469

Species of Gliocladium, Trichoderma, and Penicillum sporulating on lowbush blueberry mummies in the field or in moist chambers were evaluated for possible antagonism to M. vaccinii-corymbosi. Mummies were collected in September from commercial blueberry fields and stored dry at 20 C for 4-6 wk before being rehydrated and inoculated with conidial suspensions (3 x 10^4-6/ml) of each fungus. Suspensions were sprayed singly or in combination on 30 mummies per treatment. After 2 wk incubation in moist chambers at 25 C mummies were assessed for colonization by the fungi; percent colonization ranged from 20 (Penicillium) to 100 (Trichoderma). The three fungi were also tested for direct hyphal antagonism to M. vaccinii-corymbosi on potato dextrose agar. Potential antagonism was noted for all three imperfect fungi.

A120

A METHOD FOR SCREENING PHYLLOPLANE ANTAGONISTS TO SCLEROTINIA SCLEROTIORUM ON LETTUCE. J. Mercier and R. Reeleder, Department of Plant Science, Macdonald College of McGill University, Ste-Anne-de-Bellevue, Québec, Canada H9X 1C0.

Lettuce drop in Québec was found to be caused by Sclerotinia sclerotiorum (large sclerotial type). No infections by S. minor have been observed. Lettuce plants were readily infected by ascospores when discharging apothecia were placed with the plants in a growth chamber. Phylloplane antagonism thus appears to be a possible approach to control of the disease. Fungal and bacterial isolates from the phylloplane of lettuce are screened for antagonism to ascospores with the following in situ technique: leaf discs are inoculated with a spore/cell suspension of the candidate antagonist, with or without a nutrient source (0.1% malt extract). After an incubation period of 24 h to permit establishment of the antagonist, an ascospore suspension is added. The leaf discs are later fixed for observation with the SEM. Inhibition of germ tube growth and percentage of germination of ascospores are then assessed. Antagonistic organisms can then be tested on the plant for their effectiveness in controlling the disease.

A121

MICROORGANISMS ANTAGONISTIC TO OR PRODUCING ANTIBIOTIC INHIBITORY TO CERATOCYSTIS ULMI. Gregory, G. F., Schreiber, L.

R. and Ichada, J. USDA-FS and USDA-ARS, 359 Main Road, Delaware, OH 43015.

Studies were conducted to identify organisms antagonistic to the Dutch elm disease fungus Ceratocystis ulmi. The colonizing ability of candidates was determined by introducing them into the vascular system of American elm (Ulmus americana) seedlings and then isolating periodically from leaf petioles. Trichoderma and Bacillus spp. are the most promising. B. subtilis and B. coagulans were isolated from the xylem of elms inoculated with the pathogen. When mixtures of B. subtilis and C. ulmi spores, impregnated into blank bioassay discs, were placed on PDA, bacterial growth was dominant even when the bacterium was a very low proportion of the mixture. The bacteria produced an antibiotic in liquid or on agar media inhibitory to C. ulmi. The antibiotic is soluble in methanol and ethanol but not in nonpolar solvents. Purification by column chromatography is in progress.

A122

ON THE BIOLOGICAL CONTROL OF CERATOCYSTIS ULMI WITH PSEUDOMONAS FLUORESCENS. C.W. Murdoch, R.J. Campana and J. Hoch. N.E. Plant, Soil and Water Laboratory and Department of Botany & Plant Pathology, University of Maine, Orono, ME 04469.

Five isolates of P. fluorescens that exhibited in vitro antagonistic activity towards C. ulmi were used as combination liquid treatments to control development of Dutch elm disease. Bacteria-water suspensions (10^{10}cfu/ml) were introduced on the same day as C. ulmi or one week earlier into a total of 40 American elms (4-10cm dia at 1.3m) using either gravity-flow stem or inner bark injection techniques. Twenty more trees were inoculated with C. ulmi and 30 uninoculated trees were injected with bacteria or water. Preventative were more effective than therapeutic treatments. Trees treated with bacteria showed significantly (P=0.05) less mortality (25 vs 85%), and symptom development after 1 yr than non-treated trees. Treatments with P. fluorescens inhibited expression of Dutch elm disease symptoms in treated, inoculated trees.

A123

INABILITY OF THREE APPLIED FLUORESCENT PSEUDOMONADS TO SUCCESSFULLY COLONIZE ELMS. **K. J. Harrison-Lavoie, M. A. Hoffman, S. S. Selfridge, and B. L. McFarland, Chevron Chemical Company, Richmond, CA 94804.**

Fluorescent pseudomonads have been studied for biocontrol of Dutch Elm Disease (Ceratocystis ulmi). Recovery of 3 applied fluorescent pseudomonad strains from elm tissues, both before and 3 months after vascular application of 10^{10} cells/tree. A total of 388 pre-and 1871 post-treatment fluorescent pseudomonads were recovered. Using previous workers criteria (fluorescence on King's B agar and antibiosis), strains with characteristics of those applied (M27, 33, 254) were identified at the following respective levels relative to total fluorescent bacterial populations recovered per treatment: 8%, 21%, 21% of untreated, 16%, 16%, 26% of pretreated, and 12%, 40%, 41% of post-treated samples. Characterization including LOPAT, of 16 carbohydrates, 24 enzymes, resistance to 6/15 antibiotics, and immunoassay, indicated that no treatment strain was recovered at a frequency greater than untreated control or pretreatment levels. No correlation was observed between the occurrence of C. ulmi antagonists and disease control.

A124

BIOLOGICAL AND CHEMICAL CONTROL OF VERTICILLIUM WILT OF ALFALFA. R. L. Millar, D. W. Kalb, and A. P. Keinath, Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

During harvesting of alfalfa, freshly cut stems serve as penetration courts for Verticillium albo-atrum (Vaa). In growth chamber experiments, 8- to 10-wk-old plants (cv. Iroquois) were cut 3.0 cm above the crown. The cut stems were inoculated with Vaa (1×10^4 conidia/ml), then treated immediately with Gliocladium roseum, Penicillium sp., Trichoderma sp., or a fungicide. Wilt incidence was reduced by G. roseum (8×10^6 conidia/ml) up to 4- to 5-fold, which persisted through two harvests. Lowering G. roseum concentrations to 10^4 conidia/ml reduced wilt control down to 1.5- to 2.0-fold. Control was not achieved if treatment with G. roseum was delayed 2 min or longer or if Vaa was 10^5 conidia/ml or higher. Isolates of G. roseum varied in their effectiveness; isolates of Penicillium and Trichoderma were less effective or ineffective, respectively. G. roseum (8×10^6 conidia/ml) was as effective as thiram - 75% WP (0.52 g, a.i./250 ml) and 2 to 3 times more effective than benomyl, chlorothalonil, and maneb.

A125

EFFECT OF BACILLUS SUBTILIS ON VERTICILLIUM WILT IN SILVER MAPLE. T. J. Hall, L. R. Schreiber, and Curt Leben. USDA-ARS Nursery Crops Research Laboratory, Delaware, OH 43015 and the Dept. of Plant Pathology, The Ohio State University/OARDC, Wooster, OH 44691.

Bacteria isolated from healthy silver maple stems were identified as Bacillus subtilis. Two isolates were antagonistic to Verticillium dahliae in vitro. Suspensions of rifampicin (rf) resistant mutants B-26rf and B-5rf and a mixture of the two were introduced into silver maples (40-60 cm high) through stem wounds. After 3 days, stems were wound-inoculated with a conidial suspension of V. dahliae. After 16 wk, stem pieces were cultured for the fungus and rf bacilli. There was a highly significant reduction in the frequency of isolation of V. dahliae by B-26rf but not by B-5rf or the mixture. Rifampicin resistant bacilli were recovered from treated trees only. These results suggest that bacteria found naturally in stems of silver maples may reduce Verticillium wilt disease.

A126

SELECTION OF FLUORESCENT PSEUDOMONAS STRAINS ANTAGONISTIC TO ERWINIA CAROTOVORA. G. W. Xu and D. C. Gross, Dept. of Plant Pathology, Washington State University, Pullman, WA 99164.

A procedure was developed for screening fluorescent pseudomonads, isolated from potato roots and tubers, for inhibition of Erwinia carotovora and suppression of potato seedpiece decay. Pseudomonads were first screened on an iron deficient medium for antagonism of E. carotovora subsp. atroseptica growth, resulting from production of inhibitory siderophores with high affinities for iron. A diversity of siderophores were produced; some were highly effective in reversing iron deprivation of the pseudomonad producer strain at EDDA concentrations ranging up to 5,000 µg/ml. Antibiotic production was assayed under high iron conditions conducive to secondary metabolism. Potato seedpieces were treated with antagonistic pseudomonads (10^8 CFU/seedpiece), inoculated 24 hr later with subsp. atroseptica strain W3C37 (10^6 CFU/seedpiece), planted into fumigated soil, and grown in the greenhouse for two weeks. Several pseudomonad strains significantly improved plant emergence (up to 63%) and increased plant weights six-fold.

A127

EFFECT OF ANTAGONISTIC FLUORESCENT PSEUDOMONADS ON COLONIZATION OF POTATO ROOTS BY ERWINIA CAROTOVORA AND ON YIELD. G. W. Xu and D. C. Gross. Department of Plant Pathology, Washington State University, Pullman, WA 99164.

Fluorescent pseudomonad strains, that were antagonistic to Erwinia carotovora in vitro and reduced potato seedpiece decay in the greenhouse, were tested in the field for ability to colonize potato roots and increase yield. Seedpieces were treated (10^{10} CFU/seedpiece) with fluorescent pseudomonads just prior to planting. The more successful strains colonized roots to populations exceeding 10^5/g and were detected in high populations throughout the growing season. Appreciable populations of E. carotovora developed on the roots in late June on non-treated potatoes and approached 10^6 CFU/g by mid-July. Some fluorescent pseudomonads appeared to suppress the population of Erwinia ten-fold. Moreover, Pseudomonas putida strain W4P63 increased the yield of Russet Burbank by 12 and 10% in 1982 and 1983, respectively, and reduced the soft rot potential of the tubers. Most strains, however, did not significantly improve yield or reduce soft rot disease incidence in the field.

A128

POPULATION DYNAMICS OF SOIL PSEUDOMONADS IN THE RHIZOSPHERE. J.E. Loper, C. Haack, and M.N. Schroth. Dept. Plant Pathology, University of California, Berkeley, CA. 94720.

Rhizosphere populations of seven Pseudomonas strains originally isolated from rhizospheres of agricultural plants were studied on potato in field soil. Rhizosphere populations of two strains (B10 and B4) were quantitatively related to initial seed piece inoculum but were consistently higher for B4 than for B10 at a given inoculum level. In vivo growth curves on root segments indicated that both strains grew at similar rates in the potato rhizosphere. However, high populations of strain B10 were not maintained at 24 C after 7 hr, while those of strain B4 were maintained for at least 40 hours. Both strains grew more rapidly in the rhizosphere at 24C than at 12C, although their rhizosphere populations following seed piece inoculation were higher at 12C or 18C, indicating that rhizosphere populations were not solely determined by in vivo growth rates. In vitro osmotolerance of seven Pseudomonas strains (including strains B4 and B10) was correlated with their populations and may be involved with bacterial survival in the rhizosphere of potato.

A129

COLONIZATION DYNAMICS OF A RHIZOBACTERIUM ON POTATO. J. B. Bahme and M. N. Schroth, Dept. of Plant Pathology, Univ. of California, Berkeley, CA 94720.

The ability of seedpiece-introduced Pseudomonas fluorescens strain A1 to colonize roots, underground parts of shoots, stolons, and daughter tubers of field-grown potato plants was determined in peat-loam and loamy-sand soils at Tulelake and Bakersfield, CA, respectively. Mean seedpiece populations of A1 were ~10^5 colony-forming units (cfu) per cm^2 periderm for 2 months after planting. Strain A1 formed a bacterial gradient in the soil extending 6 cm below the seedpieces with the first irrigation. Primary roots were most heavily colonized for a distance of 8 cm beyond the seedpiece at mean populations of 1.2×10^3 cfu cm^{-1} (Tulelake) and 1.7×10^3 cfu cm^{-1} (Bakersfield), but only lightly colonized from 8 cm to 32 cm. Stolons and tubers attached near the seedpiece were more extensively colonized (1.5×10^3 cfu cm^{-1} and 2.1×10^4 cfu per tuber, respectively) than stolons and tubers arising >4 cm above the seedpiece. It appeared that colonization sufficient to influence soilborne plant pathogens or plant growth occurred only on plant parts from 8 cm below to 4 cm above the seedpiece.

A130

DOWNWARD MOVEMENT OF BIOLOGICAL CONTROL AGENTS IN THE RHIZOSPHERE W. L. Chao, H. C. Hoch, and G. E. Harman, Depts. of Horticultural Sciences and Plant Pathology, Cornell University, NYSAES, Geneva, NY 14456.

The vertical movement of fungi and bacteria in the spermosphere and rhizosphere, was studied by introducing the organisms as seed treatment. Plant roots by themselves could not serve as dispersal agents for Trichoderma spp. Conversely, they appear to be able to transport Enterobacter cloacae with them. The biotic status of the soil markedly affects the colonization of microorganisms on roots. In sterile soil, both T. harzianum and E. cloacae have been detected further down in the soil column, however, in natural soil, none of the organisms studied could be detected in the rhizosphere 3 cm below the seed. By removing part of the microbial population we could manipulate their activities (i.e. movement and colonization) to a certain extent. Percolating water enhanced the downward movement of both bacterial and fungal propagules and the depth of penetration was influenced by the amount of water added.

A131

APPLICATION OF FLUORESCENT PSEUDOMONADS TO IMPROVE THE GROWTH OF WHEAT. D. M. Weller and M. C. Graham, USDA-ARS, Washington State University, Pullman, WA 99164-6430.

Fluorescent pseudomonads from roots of wheat were screened for ability to improve the growth of wheat when applied as seed treatments. Bacterial strains were grown on King's medium B and then suspended (10^9 CFU/ml) in water containing 0.1 M $MgSO_4$. Wheat seeds were soaked in the bacterial suspension for 30 min., planted in natural Puget silt loam and incubated at 15 C. Of 64 strains tested, 17 increased the shoot height of 4-wk-old seedlings as compared to nontreated wheat, probably by suppressing Pythium spp. Addition of the fungicide metalaxyl either as a seed treatment or soil drench gave equivalent increases in seedling height, and plants from untreated seed had symptoms of Pythium root rot. Four strains tested in the field as seed treatments for winter wheat produced significant increases either in stand, plant height, number of heads, or all of these parameters. These four strains increased the grain yields 26, 20, 10, and 2%, respectively, as compared to the nontreated wheat, but only the 26% yield increase was statistically significant.

A132

PYTHIUM CONTROL BY SIDEROPHORE-PRODUCING BACTERIA ON ROOTS OF WHEAT. J. O. Becker, Plant Genetic Systems, Gent, Belgium, and R. J. Cook, USDA-ARS, Pullman, WA 99164.

About 350 of 5,000 isolates of fluorescent pseudomonads from roots of "healthy" wheat grown 3-4 weeks in a Pythium-infested Palouse silt loam (PSL) exhibited in vitro inhibition of P. ultimum on KMB, PDA, or both. About 100 of the 350 increased seedling height by 10-30% when applied to seeds of cv Daws (10^7-10^9/seed) and the seeds then sown in Pythium-infested PSL (pH 5.3; available Fe^{+3} 60 ppm). Some isolates produced growth responses equivalent to either soil fumigation or metalaxyl seed treatment. P. ultimum was inhibited by EDHHA added to the KMB agar. Of 12 growth-promoting isolates tested, 5 remained inhibitory when $FeCl_3$ (50 µg/ml) was added to the medium but 7 others became noninhibitory when $FeCl_3$ was added at 10 µg/ml. Five siderophore-negative mutants from one isolate produced no growth response in wheat, nor did the original isolate produce a growth response if FeEDTA was added to the soil. Pythium control by this isolate apparently results from iron competition, but others may suppress Pythium by other mechanisms.

A133

SUPPRESSION OF TAKE-ALL OF WHEAT IN SOUTH AUSTRALIAN SOILS BY FLUORESCENT PSEUDOMONADS. D. M. Weller & A. D. Rovira, USDA-ARS, Pullman, WA 99164 & CSIRO, Div. of Soils, Glen Osmond, SA 5064.

Strains of Pseudomonas fluorescens were tested for their ability to suppress take-all of wheat, caused by Gaeumannomyces graminis var. tritici (Ggt), in two fields in So. Australia. The bacteria were originally isolated from roots of wheat grown in soil from fields in Washington State where take-all had declined. Seeds of cv. Condor were treated with bacteria and then sown in soil naturally infested with Ggt or where 4000 or 8000 Ggt-colonized rye grass seeds per m^2 were incorporated. Of six strains tested alone or in combinations, strain 2-79 increased plant height by 4 cm at 126 days after planting (significant at $P = 0.05$) and the number of heads by 29%, compared to nontreated wheat, at one of two sites where take-all developed only from the natural inoculum. Strain 2-79 had no significant effect on the severity of take-all on wheat at either site in soil with added Ggt inoculum. During the first month after planting, antibiotic-resistant strain 2-79RN$_{10}$ was detected on seminal roots (root and rhizosphere soil), 0.5-3.0 cm along the root, at 10^5-10^6 CFU/0.1 g of root.

A134

SOIL MATRIC POTENTIAL AFFECTS COLONIZATION OF WHEAT ROOTS BY A PSEUDOMONAD SUPPRESSIVE TO TAKE-ALL. J.L. Parke*, A.D. Rovira and G.D. Bowen, Dept. of Plant Pathology, Univ. of Wisconsin, Madison, WI 53706* and CSIRO Div. Soils, Glen Osmond, S.A. 5064 Australia.

Wheat seed coated with *Pseudomonas fluorescens* strain 2-79 was planted into γ-irradiated or nonsterile South Australian wheatfield soil adjusted to -0.14, -0.21, -0.53, or -1.40 bars matric potential. Harvests 1-3 cm below the seed at 7, 14, and 28 days indicated that in nonsterile soil, populations of 2-79 were highest at -1.40 bars (2.2×10^7 CFU/g soil) and lowest at -0.14 bars (1.0×10^5 CFU/g soil). The ratio of 2-79 to indigenous soil bacteria increased with a decrease in soil moisture. In irradiated soil, -0.53 bars was the optimal matric potential but high populations of 2-79 occurred even at -0.14 bars (1.5×10^8 CFU/g soil). The different response of 2-79 in irradiated and nonsterile soil suggests that soil matric potential influences root colonization by 2-79 through an effect on competition between rhizosphere microorganisms.

A135

INFLUENCE OF BIOMASS, MICROELEMENTS AND NUTRIENT LEVELS ON ACTIVITY OF SIDEROPHORE-PRODUCING PSEUDOMONADS IN SOIL. Y.Elad and R. Baker, Dept. of Botany and Plant Pathology, Colorado State Univ., Fort Collins, CO 80523.

Direct correlations were obtained between siderophore (sid) production by pseudomonads (psd) in culture and their ability to induce suppressiveness in soil to Fusarium wilts of radish ($r = 0.83$) and cucumber ($r = 0.75$). Inhibition of chalamydospore (chl) germination in soil (to 85%) of F. oxysporum f. sp. cucumerinum was greater in the presence of sid-producing pds, at population densities of >10^7/g, than with psd producing low sid even when the latter had a higher biomass (<10^8/g). Inhibition of chl germination by pds was counteracted partially by cations due to competition on the Fe^{3+} binding site of the sid and/or inhibition of multiplication of psd in soil. Psd reduced germination of chl 10-15% in soil at pH 5.0-5.5, whereas addition of purified sid reduced germination by 41-43%.

A136

IRON COMPETITION BY ALCALIGENES SP. REDUCES FUSARIUM CHLAMYDOSPORE GERMINATION. G. Y. Yuen and M. N. Schroth. Dept. of Plant Pathology, Univ. of California, Berkeley, CA 94720.

Alcaligenes sp. strain MFA1 reduced the germination of Fusarium oxysporum f. sp. dianthi (Fod) chlamydospores by 65% when applied to Fod-infested soil at 10^3 and 10^9 colony-forming units (cfu) g^{-1} along with 0.1% glucose and 0.1% asparagine. MFA1 at 10^3 cfu g^{-1} along with 1% glucose and 1% asparagine (1% GA) also reduced germination by 65%. No inhibition of germination occurred when 10^9 cfu g^{-1} MFA1 were added with 1% GA. Soil populations of MFA1 increased 10-fold and decreased 100-fold over 24 h when added to soil at 10^3 and 10^9 cfu g^{-1}, respectively. Reduction of germination did not occur when UV-killed cells of MFA1 or two strains of Pseudomonas sp. were added to soil. Reduction of germination appeared to be caused by iron competition with MFA1, as addition of $FeCl_3$ to soil reversed the inhibitory effect of MFA1 and mutants of MFA1 deficient in siderophore production had no effect on germination.

A137

LECTIN-MEDIATED ATTACHMENT OF THE BIOCONTROL AGENT ENTEROBACTER

CLOACAE TO FUNGAL HYPHAE: I. ROLE IN FUNGAL GROWTH INHIBITION. E.B. Nelson, W.L. Chao, J.M. Norton, and G.E. Harman, Dept. of Horticultural Sciences, Cornell Univ., NYSAES, Geneva, NY 14456

In vitro interactions between E. cloacae and seed-rotting pathogens were evaluated to determine mechanisms by which E. cloacae protects against Pythium seed rot. All bacteria isolates tested induced growth reductions of P. ultimum. However, antibiotics, toxic substances, or cell wall degrading enzymes were not detected in culture filtrates. E. cloacae was able to move from a point source along fungal hyphae and throughout the entire colony. This interaction could be modified by changing the sugar composition of the medium or the fungus with which E. cloacae was paired. In the presence of sugars promoting movement, no growth inhibition of P. ultimum occurred. In the absence of sugar or in the presence of sugars preventing movement, growth inhibition ranged from 22-45%. Growth inhibition was associated with the binding of E. cloacae to fungal hyphae as determined by scanning electron microscopy and fungal cell was agglutionation. Sugars which eliminated binding also eliminated fungal growth inhibition.

A138

LECTIN-MEDIATED ATTACHMENT OF THE BIOCONTROL AGENT ENTEROBACTER CLOACAE TO FUNGAL HYPHAE: II. ROLE IN THE BIOLOGICAL CONTROL OF PYTHIUM SEED ROT. E.B. Nelson, G.T. Nash, and G.E. Harman, Dept. of Horticultural Sciences, Cornell Univ., NYSAES, Geneva, NY 14456

E. cloacae was evaluated as a biological seed treatment on a variety of plant species susceptible to Pythium seed rot. The degree of protection provided by E. cloacae was dependent upon the levels of carbohydrates exuded from seeds during germination. Seeds exuding low levels of carbohydrates were effectively protected by E. cloacae from Pythium seed rot whereas seeds with large amounts of exudates were severely damaged. Artificially increasing the sugar concentration around low exudation seeds reduced the level of protection by E. cloacae. Adherence of E. cloacae and the plant lectin, Concanavalin A to P. ultimum hyphae on seed coats in soil was examined using fluorescence microscopy to determine the relationship between seed exudates and attachment of E. cloacae to P. ultimum hyphae in vivo. The relationship between hyphal attachment in the spermosphere and biological control of Pythium seed rot is discussed.

A139

BIOCONTROL OF PHYTOPHTHORA CINNAMOMI ON PERSEA INDICA AND P. AMERICANA BY PRIOR INOCULATION WITH PHYTOPHTHORA PARASITICA. T. E. Dolan and M. D. Coffey, Department of Plant Pathology, University of California, Riverside 92521

Root dipping 45-day-old seedlings of Persea indica (PI), a host highly susceptible to P. cinnamomi (PC), for 48 h in minced mycelium of P. parasitica (PP) reduced disease development after challenge with PC zoospore suspensions. PC-challenged plants exposed to PP mycelium showed no wilting and few root lesions. Control plants had severe root rot symptoms and 83% wilting 7 days after challenge. Plating PP treated roots before challenge yielded 80% recovery. Dipping PI roots in killed PP mycelium did not significantly reduce disease. Exposing transplanted PI seedlings to PP infested soil for two weeks reduced disease after challenge with an overlay of PC infested soil. All control plants were dead in 4 weeks while only 18% of PP treated plants had wilted 8 weeks after challenge. With "Topa Topa", an avocado (Persea americana) rootstock, exposure to PP infested soil prior to PC challenge reduced root infection relative to the control by 71%.

A140

PROTECTING PERSEA INDICA SEEDLINGS FROM PHYTOPHTHORA CITRICOLA BY A PRIOR INOCULATION WITH THREE OTHER PHYTOPHTHORA SPECIES. Yigal Cohen and Michael D. Coffey, Department of Plant Pathology, University of California, Riverside, CA 92521.

Roots of 35- 45-day-old P. indica plants were dipped in zoospore suspensions (about 3 x 10^4/ml) of P. capsici (PC) P. parasitica (PP), or P. parasitica nicotiana race 0 (PPN) and challenged on the stem 3 days later with P. citricola (PCT) zoospores (about 1000 zoospores/stem). PC, PP, and PPN produced slight root infection but reduced the amount of disease caused by PCT on stems by 65, 48, and 57% respectively. PCT produced moderate root infection but induced no protection of the stem. Dipping the roots and stems in mycelial fragments of either PP or PPN, but not of 3 Pythium species, or inoculating the stems with zoospores of PP or PPN resulted in >90% protection against PCT. Preliminary experiments revealed that plants grown in PP-infested soil were protected against PCT.

A141

INHIBITION OF PHYTOPHTHORA MEGASPERMA F. SP. GLYCINEA ZOOSPOROGENESIS BY GLIOCLADIUM ROSEUM, TRICHODERMA HARZIANUM AND TRICHOTHECIUM ROSEUM CULTURE FILTRATES. M. B. Al-Heeti and J. B. Sinclair, Dept. of Plant Pathology, Univ. of Illinois, 1102 S. Goodwin, Urbana, IL 61801.

Culture filtrates (CF) of G. roseum, T. harzianum and T. roseum grown separately on potato-dextrose broth for 14 days at room temperature were fitler sterilized and diluted (1:99, 5:95, 10:90 v/v) with double distilled water (DDW). Pmg was cultured on lima-bean agar (6 days, 25 C). Cultures were washed every 30 min for a total of five times using 10 ml DDW per wash and finally flooded with 15 ml of one of each CF treatment. Four replicates were used in each of three experiments. A control of 15 ml DDW yielded 1.8×10^6 zoospores/ml. CF of T. roseum completely inhibited zoosporogenesis at all conc. G. roseum and T. harzianum CF at 5 and 10% yielded 4.9×10^5, 4.3×10^5 and 1.8×10^5, 1.5×10^5, respectively. These CF at 1% did not inhibit zoosporogenesis.

A142

CLONING AND NUCLEOTIDE SEQUENCING OF THE 5' AND 3' ENDS OF PAPAYA MOSAIC VIRUS RNA. M.G. AbouHaidar and V. Ramassar. Dept. of Botany, University of Toronto, Toronto, Ont. M5S 1A1. Papaya mosaic virus (PMV) is a potexvirus with an RNA of 2.10^6 daltons. The RNA binds to poly (U) sepharose column indicating the presence of poly (A). Complementary DNA copies of the 3' end of PMV-RNA were synthesized using oligo dT (12-18) as primer and reverse transcriptase. After dC tailing, the cDNA copies were cloned in the PST_1 site of PBR322 tailed with dG. A library of clones covering the 3' end region of PMV-RNA has been established. Similarly the 5' end region of PMV-RNA has also been cloned in the PST_1 site of PBR322. In this case, PMV-RNA fragments representing the 5' end of the RNA were isolated by partial and polar in vitro assembly followed by the addition of poly (A) with poly(A) polymerase and then cloning. DNA fragments representing both termini of PMV-RNA are being identified and sequenced in the M_{13} cloning-sequencing system. Several repeated sequences (GCAAA) were found at the 5' end. The primary and secondary structures of the two termini of PMV-RNA and their roles in terms of replication and packaging of the viral RNA will be discussed.

A143

A CARNATION MOTTLE VIRUS GROUP: A COMPARISION OF SOME TENTATIVE MEMBERS. T. J. Morris and J. C. Carrington, Department of Plant Pathology, University of California, Berkeley, CA 94720.

A new group of small RNA viruses with similarity to carnation mottle virus (CarMV) but distinct from tomato bushy stunt virus (TBSV) has been proposed (Hull, 1977, and Koenig et al., 1983). Several tentative members of this group including galinsoga mosaic (GMV), saguaro cactus (SCV), turnip crinkle (TCV) and cucumber soilborne (CSBV) viruses have been compared to both CarMV and TBSV. Physicochemical properties, genome size ($1.3\text{-}1.4 \times 10^6$ d) and the pattern of viral related RNA species all indicate that these viruses are most similar to CarMV and distinct from TBSV. An evaluation of CarMV related RNAs in infected tissue extracts by northern blot hybridization using complementary DNA probes and nick-translated probes from cDNA clones of CarMV RNA has established the production of two subgenomic RNAs (0.53 and 0.58 x 10^6 d) from the 3' domain of the virus genome. The genome organizations and possible relationships among these viruses will be discussed in relation to comparative northern blot hybridizations.

A144

CHARACTERIZATION OF LOW MOLECULAR WEIGHT RNA SPECIES OF MEMBERS OF THE TOMBUSVIRUS GROUP. B. I. Hillman and T. J. Morris, Department of Plant Pathology, University of California, Berkeley, CA 94720.

Low molecular weight RNA species have been isolated from tomato bushy stunt virus (TBSV), artichoke mottled crinkle virus (AMCV) and cymbidium ringspot virus (CyRSV), each a member of the tombusvirus group. The low molecular weight RNA species are single stranded and linear, with sizes ranging from 1.5×10^5 for TBSV to 2.5×10^5 for CyRSV. With TBSV, the low molecular weight RNA is encapsidated in protein encoded by genomic RNA, but probably in particles containing only low molecular weight RNA. In TBSV infection of Nicotiana clevelandii, symptom modulation was associated with presence of low molecular weight RNA. Gel purified TBSV-BS-3 strain low molecular weight RNA did not replicate when inoculated alone, but its replication was supported by the Prunus strain of TBSV, and the symptom modulating effect was again observed. Northern hybridization analysis suggests that the TBSV low molecular weight species is related to TBSV genomic RNA.

A145

THE 3' TERMINAL SEQUENCES OF BARLEY STRIPE MOSAIC VIRUS RNAS.

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Purified preparations of barley stripe mosaic virus contain both genomic (g) and subgenomic (sg) RNAs. The gRNAs are most abundant and consist of three sequences designated α, β, and γ that vary in size and number depending on the strain of the virus. All strains contain at least three sgRNAs derived from the gRNA. The most abundant of these is an 800 nucleotide (NT) sgRNA with a polyadenylated terminus varying in length from 10 to 150 NT. In contrast, the 3' ends of the α, β and γ gRNAs are highly conserved and have an unusual organization. The 240 terminal NT of all three gRNAs are nearly identical and appear to have considerable secondary structure as anticipated from their ability to aminoacylate tyrosine. This tRNA-like structure is flanked by an internal polyadenylated sequence of $\sim$40 NT.

A146

TERMINAL SEQUENCES OF THE DOUBLE-STRANDED RNAs OF CUCUMBER MOSAIC VIRUS AND ITS SATELLITE RNA.
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The double-stranded (ds) form of CARNA 5, the satellite RNA of CMV, contains an unpaired guanosine at the 3' end of the (-) strand. The (+) strand of ds CARNA 5 is uncapped at its 5' end, but the 3' end is identical to that of the encapsidated, single-stranded form of the satellite RNA. The terminal sequences of the ds forms (RFs) of RNAs 3 and 4 of CMV are being determined. The 3' end of the (-) strand of ds RNA 3 also contains an unpaired guanosine, whereas that of ds RNA 4 is heterogeneous, with most molecules lacking the extra guanosine. Furthermore, the 3' ends of the (+) strands of ds RNAs 3 and 4 are different from those of the encapsidated, single-stranded RNAs 3 and 4. These findings show intriguing analogies with certain animal and bacterial viral RNAs and have possible implications for the replication of both the virus and its satellite.

A147

EPITOPE SPECIFICITY OF SEVEN MONOCLONAL ANTIBODIES TO APPLE MOSAIC VIRUS (ApMV) AND PRUNUS NECTROTIC RINGSPOT VIRUS (PNRV). Ramon L. Jordan, J. Aebig and H. T. Hsu. USDA, Beltsville, MD 20705 and American Type Culture Collection, Rockville, MD 20852.

The immunoreactivities and epitope specificities of 7 monoclonal antibodies (McAbs) against ApMV and PNRV were analyzed for McAb isotype, virus serogroup specificity, affinity and binding activity in several different ELISA procedures, and competitive inhibition assays with biotin-labelled antibodies. McAbs were purified by sephacryl and DEAE Affi-gel blue column chromatography before labelling with biotin for use in an avidin-biotin ELISA system. The McAbs can differentiate at least 5 different serotypes of ApMV and 3 serotypes of PNRV. At least 6 different antigenic sites (epitopes) can be delineated by the 7 McAbs. The specific binding characteristics of these McAbs to whole, detergent-, or enzyme-treated virions were evaluated in ELISA, dot-blot and Western-blot analyzes.

A148

THE EFFECT OF pH ON THE DISSOCIATION OF PRECIPITATES OF SOUTHERN BEAN MOSAIC VIRUS WITH THREE MONOCLONAL ANTIBODIES.
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Nine antibodies from hybridomas derived from mice injected with virions of the bean strain of southern bean mosaic virus (SBMV-B) reacted with SBMV-B virions in enzyme-linked immunosorbent assays. SBMV-B virions were precipitated in gel diffusion or decorated in immunoelectron microscopy tests by only three of these antibodies (B1, B2, and B3). Only B3 precipitated virions of the cowpea strain (SBMV-C). Virion-antibody precipitates were washed, resuspended overnight at pH 2.25,2.50, 2.75, 3.00 and 4.00 and centrifuged on sucrose density gradients at the same pH. Reactive antibodies and SBMV-B were recovered from the gradients with B2 at pH 2.25, with B1 at pH 2.75 or lower and with B3 at pH 3.00 or lower. Heterologous precipitates of B3 with SBMV-C were completely dissociated at all pH values.

A149

PROPERTIES OF GRAPE AND SOYBEAN ISOLATES OF TOMATO RINGSPOT VIRUS. Robert G. Tuskan and Sue A. Tolin. Dept. Plant Pathol., Physiol. & Weed Sci., VPI&SU, Blacksburg, VA 24061.

Tomato ringspot virus was isolated from soybeans grown in a test for seed-borne virus (TmRSV-S) and from Vidal 256 grapes (TmRSV-G) in a declining ten-year-old vineyard in northern Virginia. The isolates were serologically indistinguishable from each other, from TmRSV isolated from weed species in vineyards and from other TmRSV strains. Virus was purified from soybean (cv. York) or cucumber (cv. National Pickling) by extraction in 0.05M sodium phosphate buffer containing 0.5-1% ascorbic acid, pH 7.0, and clarification with amyl alcohol: butanol or ammonium sulfate. Following differential centrifugation, sedimentation in sucrose gradients revealed components typical of Nepoviruses. The ratio of components varied depending on virus isolate and host. Analysis of sedimentation and other properties suggest these isolates differ from previously described TmRSV strains.

A150

POLYMERIZATION OF POTEXVIRUS PROTEINS. K. M. Ready and J. B. Bancroft, Dept. of Plant Sciences, Biological and Geological Sciences Building, the University of Western Ontario, London, Ontario, Canada N6A 5B7.

Coat proteins isolated from various members of the potexvirus family polymerize to form different structures. In terms of small aggregates, barrel cactus virus (BCV), potato virus X (PVX), foxtail mosaic virus (FTV), viola mottle virus (VMV) and tulip virus X (TVX) form species which sediment at about 4 and 7S near neutrality, at 20°C, whereas papaya mosaic virus (PMV) and clover yellow mosaic virus (CYMV) proteins form 14S aggregates. These differences suggest that although the potexviruses are structurally very similar, the aggregates from which they form are not. There is also considerable diversity in the structure of tubular protein polymers. The particles may have the same surface structure as their respective viruses (PMV, CYMV, BCV, FTV) but form under quite different conditions from one another, indicating different control mechanisms, or be composed of stacked rings (PVX) or a variety of multiple helices (VMV, TVX, narcissus mosaic virus). Some of these structurally different particles of protein from different viruses form under the same conditions. It appears that the proteins from PVX-related viruses can form more diverse structures than those from the TMV-related viruses.

A151

A SATELLITE-LIKE VIRUS PARTICLE ASSOCIATED WITH MAIZE WHITE LINE MOSAIC VIRUS. R. E. Gingery and Raymond Louie, USDA-ARS, Department of Plant Pathology, The Ohio State University, OARDC, Wooster, OH 44691.

A 17-nm diam satellite-like virus (SLV) was discovered in maize white line mosaic virus (MWLMV)-infected maize. SLV was serologically unrelated to MWLMV and the satellite viruses of tabacco necrosis and panicum mosaic viruses, but was related to a satellite-like virus associated with maize dwarf ringspot virus (MDRSV) from France; MWLMV and MDRSV were serologically related also. Both SLV and MWLMV were detected in the roots of all MWLMV-infected plants examined; the leaves of these plants contained either no particles, both particles, or only MWLMV, but never SLV alone. Yields of purified SLV from maize roots and leaves were usually 300-500 ug/g tissue. SLV had a sedimentation coefficient of 47S, a buoyant density in CsCl of 1.355 g/ml, and a 24.7 kd coat protein. SLV, like MWLMV, was soil transmitted.

A152

PURIFICATION OF TWO STRAINS OF SOYBEAN DWARF VIRUS. Adrianna D. Hewings[1], V.D. Damsteegt[1], S.A. Tolin[2], and P.L. Hunst[1,2]. [1]USDA-ARS, Plant Disease Research Laboratory, Ft. Detrick, Bldg. 1301, Frederick, MD 21701, [2]Dept. of Plant Pathology, Physiology & Weed Science, VPI & SU, Blacksburg, VA 24061.

Two strains of soybean dwarf virus (SDV) have been purified from 'Wayne' soybeans. Solutions (2%) of Driselase, pectinase, and cellulase, singly or in combination were equally effective in releasing virus from phloem tissue. More virus was recovered when .5 or .2M rather than .1 or .05M phosphate buffers were used for enzyme extraction. Yields were greatest when tissue was incubated with enzymes for 6 hr when compared to 3, 12, 18 or 24 hr. Triton X-100 and PEG reduced yield and purity of final preparations. The viruses were stable in chloroform, amyl alcohol-butanol and chloroform-butanol. After inoculation with Acyrthosiphon solani, virus concentration in greenhouse-grown soybeans increased rapidly, peaked in 10-12 days and remained at that level for at least 7 days. Yields were 10X greater from plants infected for 14 days than from 28 or more days.

A153

DOUBLE-STRANDED RNA ISOLATED FROM SOYBEAN PLANTS INFECTED WITH STRAINS OF SOYBEAN DWARF VIRUS. Penny L. Hunst[1,2], Adrianna D. Hewings[1], V.D. Damsteegt[1], S.A. Tolin[2]. [1]USDA-ARS Plant Disease Research Laboratory, Ft. Detrick, Bldg. 1301, Frederick, MD 21701, [2]Dept. of Plant Pathology, Physiology & Weed Science, VPI & SU, Blacksburg, VA 24061.

The dwarfing and yellowing strains of soybean dwarf virus (SDV) differ in symptomatology on soybean but do not cross-protect. Little is known about the physiochemical properties of the viruses and no information is available on SDV nucleic acids. The present study was initiated to isolate and identify the RNA replicative forms from tissue infected with either or both strains. Double-stranded RNAs of the two SDV strains were isolated from 20g of infected 'Wayne' soybean tissue by chromatography on Cellex N-1 according to a modified procedure of Dodds and Bar-Joseph (Phytopath. 73:419). Differences in the electrophoretic mobility were observed in the dsRNAs isolated from tissue infected with different strains. The dsRNAs were insensitive to digestion by RNase I and are believed to be the replicative forms of the viral genomes.

A154

EPIDEMIOLOGY AND YIELD LOSSES ASSOCIATED WITH FUNGAL BROWN SPOT OF WILD RICE. Clint L. Kohls and James A. Percich. Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Wild rice (Zizania pulustrus) yield reductions associated with disease incited by Bipolaris oryzae were 0, 0, 32, 56 and 67 percent in control, milk, 1/4 grain elongation, heading, and boot plant growth stage disease onset treatments; 39, 74, 69, and 74 percent in boot to: 1/4 grain elongation, milk, ripe (light disease), ripe (normal disease) stages in disease progression treatments; 71, 76, 90 and 90 percent for M3, Netum, K2 and Johnson cultivars; and 32, 26, 22, and 15 percent in 0, 30, 60, and 90 pounds spring applied nitrogen fertilizer treatments, respectively. The effect of plant density on disease and yield, and the influence of length of wet period and temperature on infection were also investigated. Experiments, field surveys and new growth stage and disease assessment keys form the basis for a new fungicide scheduling program for fungal brown spot of wild rice.

A155

EFFECT OF PHYTOPHTHORA MEGASPERMA VAR. SOJAE ON YIELD OF ASPARAGUS. Peter G. Falloon, L. M. Falloon*, B. L. Benson*, and R. G. Grogan. Departments of Plant Pathology and *Vegetable Crops, University of California, Davis, CA 95616.

Field plots of asparagus (Asparagus officinalis L.) established in Yolo loam at Davis in 1978 were inoculated with field soil containing Phytophthora megasperma var. sojae and regularly (3 to 5 month intervals) sprayed with metalaxyl (Ridomil 2E) at 1.12 kg a.i./ha to control Phytophthora rot. Sprayed plots yielded 43% and 118% more marketable spears during 1982 and 1983 respectively. Spear counts indicated that a significant amount of spear and/or crown rot occurred below the soil surface in unsprayed plots. Control of Phytophthora with metalaxyl resulted in earlier production and higher yields of larger diameter spears in 1983. Data for 1984 also will be presented.

A156

EFFECTS OF STEM RUST EPIDEMICS ON YIELD OF MIXTURES OF WHEAT CULTIVARS. I.S. Hoang, P.S. Teng. Dept. of Plant Pathology; and A.P. Roelfs, Cereal Rust Laboratory, Univ. of Minnesota, St. Paul, MN 55108.

Mixtures of a susceptible (Lee) and a resistant (Chris) cultivar in ratios of 100:0, 75:25, 50:50, 33:67, 17:83, and 0:100, were planted in 5 m x 10 m plots and replicated three times. Half of each plot was sprayed with triadimefon; the other half was left unsprayed. Plots were artificially inoculated with Puccinia graminis f. sp. tritici, race 15-TNM, at boot stage. Disease severity was rated twice per week with the modified Cobb scale. Plots of 75:25, 33:67, 17:83, and 50:50 had disease severities of 39, 29, 21, and 20% respectively, while pure stands of a susceptible and a resistant cultivar had 41 and 2% respectively. Plots of 100:0, 75:25, 50:50, 33:67, 17:83, and 0:100 produced yields of 120, 134, 146, 161, 194, and 215 g per 3 m row respectively.

A157

EFFECTS OF EARLY BLIGHT ON POTATO YIELD. P.S. Teng and H.L. Bissonnette. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

The effects of early blight (Alternaria solani) on potato cvs. Norland (N) and Russet Burbank (RB) were studied in three experiments by modifying epidemics using different timing and number of applications of captafol, maneb and triphenyltin hydroxide. Maximum yield losses were 58.4% with 60.0% terminal blight severity for N and 34.4% with 58.3% terminal blight severity for RB, both compared with control. Linear regression models for yield loss using 10-day blight severity, blight severity change per 10-day period or area under curve did not explain more than 50% of yield loss variation in both cvs. Linear multiple point models using 10-day blight severities as independent variables explained at most 81% (N) and 71% (RB) of yield loss while similar models using blight severity change per 10-day period explained at most 79% (N) and 70% (RB) of yield loss due to blight.

A158

THE EFFECT OF BUNCH DISEASE ON YIELD AND QUALITY OF PECANS. Paul F. Bertrand, Department of Extension Plant Pathology, The University of Georgia, Tifton, GA 31794 and T. R. Gottwald, USDA Southeastern Fruit and Tree Nut Research Lab., Byron, GA 31008

The effect of bunch disease on yield and quality of pecans was measured on 44 c.v. 'Schley' trees in 1981 and 107 c.v. 'Mahan' trees in 1983. The trees in each orchard were visually rated and placed in one of five categories defined by the percent of the bearing surface affected by bunch disease. Yields were recorded for each individual tree. Subsamples were collected from each tree for quality analysis. Increasing levels of bunch disease were associated with decreasing yields in both 1981 ($R^2 = 0.44$; $p = 0.0004$) and 1983 ($R^2 = 0.10$; $p = 0.0008$). Reduced nut size was related to increasing severity of bunch disease in 1981 ($R^2 = 0.54$; $p = 0.0028$) but not 1983 ($R^2 = 0.0007$; $p = 0.79$). The percent kernel and the moisture, oil and protein content of the kernels was not affected by bunch disease either year.

A159

EXPERIMENTAL DESIGN FOR DETERMINATION OF YIELD LOSSES DUE TO MAIZE DWARF MOSAIC VIRUS. L. V. Madden, J. K. Knoke, and R. Louie, Departments of Plant Pathology and Entomology, The Ohio State University/OARDC, Wooster, OH 44691.

The optimal number of rows and replications (blocks) for evaluating resistance and tolerance of maize to maize dwarf mosaic virus (MDMV) in a split-plot experimental design was determined. The "whole-plot" was maize genotype and the "split-plot" was inoculation treatment, i.e., a noninoculated control, and inoculation with MDMV strain A or B. Seven replications and four rows per treatment were used. Disease incidence and yield were determined for each row. The error variance and least significant difference (LSD) between two means were calculated for all combinations of two through seven replications and for one through four rows. The LSD for both disease incidence and yield declined exponentially with number of replications. Three or four, but not two, replications with two rows provided precise estimates of the LSD. Additional replications or rows did not substantially improve precision.

A160

THE STRUCTURE OF THE HYPODERMIS AND ITS INFLUENCE ON MYCORRHIZAL AND PLANT PATHOGENIC FUNGI. Nina Shishkoff Dept. Plant Pathology, Cornell U., Ithaca, N.Y. 14853

Penetration of roots by some fungi is affected by the presence and type of hypodermis. The hypodermis may be uniform or consist of two kinds of cells. In the latter, morphologically distinct, deep-staining (e.g. with trypan blue) cells are regularly interspersed with larger nonstaining cells. Endomycorrhizal fungi appear to penetrate only deep-staining specialized cells in plants with a dimorphic hypodermis. Pyrenochaeta terrestris (Hansen) Gorenz, Walker & Larson on onion only enters such specialized cells. Staining the roots of various plants with aniline blue showed that specialized cells fluoresced. When roots were stained with chlortetracycline, specific for membrane-bound calcium, the specialized cells fluoresced. These preliminary results indicate a difference in wall composition & structure from the unspecialized cells.

A161

INCIDENCE OF VESICULAR-ARBUSCULAR (VA) MYCORRHIZAL FUNGI IN

FLORIDA NATIVE PLANTS. N. C. Schenck, C. R. Johnson, and M. Niederhofer. University of Florida, Gainesville, FL 32611

Seventy-five root-soil samples from 7 locations and 54 native plant species were assayed for VA mycorrhizal fungi. Locations varied greatly in soil characteristics and predominant flora. Only a few plant species occurred in more than one site. Roots were cleared and stained to assay for mycorrhizal root colonization and soil samples were wet-sieved for spores. Thirty-two species from 5 genera of VA mycorrhizal fungi were identified. The average number of spores per liter (L) of soil was: Acaulospora spp., 1385; Glomus spp., 630; Gigaspora spp., 300. Spores of Entrophospora and Sclerocystis spp. were recovered only in low numbers from two locations. The highest incidence of spores occurred at a marine site for Acaulospora spp. (3860/L), at a phosphate mine area for Gigaspora spp. (1105/L), and at a marl soil site for Glomus spp. (2905/L). The predominant species for each genus was: A. mellea, Gl. geosporum, Gi. pelucida, E. colombiana, and S. rubiformis.

A162

INTERACTION OF GLOMUS INTRARADICES, MELOIDOGYNE INCOGNITA AND PHOSPHORUS ON COTTON. G. S. Smith, R. W. Roncadori and R. S. Hussey, Dept. Plant Path., Univ. of Georgia, Athens, GA 30602.

Root-knot susceptible cotton (Stoneville 213) was grown with or without Glomus intraradices (GI), Meloidogyne incognita (MI), or superphosphate (low P=75 mg/kg; high P=130 mg/kg) in all treatment combinations in field microplots (1982 + 1983) and the greenhouse. Yields in microplots were unaffected by P rate except that MI-infected plants in high P soil were lower than MI-infected plants in low P soil. Average yields were increased 33% by GI and suppressed 31% and 63% by GI+MI and MI, respectively, compared with noninoculated controls. MI reproduction was unaffected by GI. In the greenhouse shoot and boll weights were similar for GI and GI+MI treated plants in low P soil and greater than control or MI-infected plants at either P rate. GI suppressed MI egg production and juveniles an average 62% and 85% irrespective of P. In microplots GI apparently increased plant tolerance to MI whereas high P enhanced plant susceptibility. Conversely, in the greenhouse GI rendered cotton less suitable for MI and P enhanced plant tolerance.

A163

INFLUENCE OF MYCORRHIZAL FUNGI ON CYTOKININ PRODUCTION IN SOUR ORANGE. R. M. Davis, M. H. Edriss, and D. W. Burger, Texas A&I University Citrus Center, P.O. Box 1150, Weslaco, TX 78596

Nonmycorrhizal or mycorrhizal (Glomus fasciculatum, G. etunicatum, or Gigaspora heterogama) sour orange (Citrus aurantium) seedlings were fertilized with three phosphorus levels. At 0 mg P/L, dry weight and cytokinin production of mycorrhizal plants were more than twice those of the nonmycorrhizal plants. Concentrations of P in leaves of the mycorrhizal plants were also greater than the concentrations of P in the nonmycorrhizal plants. At 50 mg P/L, concentrations of P in leaves were similar whether or not plants were mycorrhizal. However, dry weight and cytokinin production of the mycorrhizal plants were still twice those of the nonmycorrhizal plants. At 100 mg P/L, dry weight and cytokinin production did not differ between nonmycorrhizal plants and plants infected with two of the three mycorrhizal fungi. In G. heterogama-infected plants cytokinin production was greater than that of nonmycorrhizal plants despite similar dry weights and P concentrations in the leaves. Enhancement of cytokinin production appeared to be associated with mycorrhizal infection rather than with increased P uptake.

A164

IPRODIONE-RESISTANT BOTRYTIS CINEREA FOUND IN ONTARIO VINEYARDS AND GREENHOUSES. J. Northover and J.A. Matteoni, Agriculture Canada, Vineland Station, Ontario, Canada, L0R 2E0.

In 1983, isolates of Botrytis cinerea with low level resistance (LLR) to iprodione were found in 5 of 11 vineyards where 10 or more applications of Rovral (50% iprodione) had been made during the last 3 to 5 years. Resistant isolates were found also in 13 of 21 greenhouses on various vegetable and ornamental crops. The linear mycelial growth of these isolates was inhibited by 10 to 50% on PDA amended with iprodione at 2 $ug.mL^{-1}$ and it was inhibited completely at 10 $ug.mL^{-1}$. The LLR isolates showed a 10-fold resistance in contrast to a 1000-fold resistance for most laboratory-selected isolates. The LLR isolates produced sclerotia, while mycelial growth and sporulation were slightly less than for the iprodione-sensitive isolates. Rovral or Captan alone provided 47-51% control of bunch rot in a vineyard with 78% bunches infected, while a combination of Rovral and Captan gave 72% control and reduced infection by low level iprodione-resistant isolates of B. cinerea.

A165

TOLERANCE OF RHIZOCTONIA SOLANI TO PENTACHLORONITROBENZENE AND SENSITIVITY OF PYTHIUM MYRIOTYLUM TO METALAXYL AND THEIR PATHOGENICITY TO PEANUT. A. B. Filonow, Department of Plant Pathology Oklahoma State University, Stillwater, OK 74078.

Isolates of Rhizoctonia solani AG4 (RS) and Pythium myriotylum (PM) were obtained from peanut pods in Oklahoma. Radial growth of RS on potato-dextrose agar containing pentachloronitrobenzene (PCNB) and of PM on corn meal agar with metalaxyl was assessed. Incubation was for 44-48h and at 25C for RS and 18-22h and at 37C for PM. Growth inhibition was based on a comparison to growth on fungicide free medium. Some RS isolates were inhibited 80-90% at 75-100 µg PCNB/ml, whereas others were inhibited 40-50 % at 1000 µg/ml. Sclerotia formed at 1000 µg/ml. For PM isolates growth inhibition was 0-45%, 27-88%, 44-83%, 76-86% and 79-92% at 1, 5, 10, 20 and 25 µg metalaxyl/ml, respectively. Continued mycelial growth and formation of sporangia and oospores were evident at 25 µg/ml. All isolates of RS and PM were pathogenic to cv. 'Tamnut 74' seedlings, causing damping-off, root rot and/or stunted growth. PCNB-tolerant isolates of RS produced pod rot on cv. 'Early Bunch'.

A166

EFFECT OF SINGLE, ALTERNATING, AND COMBINATION FUNGICIDE TREATMENTS ON PROPORTION OF METALAXYL RESISTANT:METALAXYL SENSITIVE INDIVIDUALS IN POPULATIONS OF PYTHIUM APHANIDERMATUM. P. L. Sanders, W. J. Houser, and P. J. Parish, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Perennial ryegrass was inoculated with populations of Pythium aphanidermatum with known proportions of metalaxyl resistant (R): metalaxyl sensitive (S) individuals. Metalaxyl was applied singly, in alternation, or in combination with mancozeb or propamocarb. The fungus populations were cycled through inoculation, fungicide application, incubation, harvest/assay, and reinoculation. Completely sensitive populations were cycled 16 times over 2 years in field and greenhouse without emergence of resistant individuals under any fungicide regimen. In populations with R:S ratios of 1:2, 1:10, and 1:1000, the R:S ratios were affected differentially by the single, alternating, and combination fungicide treatments.

A167

The Effects of Metalaxyl Exposure on the Sensitivity of Phytophthora parasitica var. nicotianae Isolates to Metalaxyl. H. D. Shew. Department of Plant Pathology, N. C. State University, Raleigh 27695-7616.

A total of 896 isolates of P. parasitica var. nicotianae was screened in vitro for sensitivity to metalaxyl. Isolates were collected prior to initial use of metalaxyl and after 1, 2, or 3 years of consecutive fungicide use at recommended field rates. Mean mycelial growth rate decreased linearly with increasing metalaxyl conc of 0.1 to 10 ug/ml. A wide range in fungicide sensitivity was observed; mycelial growth ranged from 3-140% of the control at 1.0 ug/ml metalaxyl. Isolates varied greatly in fungicide sensitivity within and among fields. Mean ED_{50} for growth was 0.4, 0.3, 0.7 and 0.9 ug/ml for isolates collected after 0, 1, 2, and 3 years of fungicide exposure, respectively. An in vivo screening procedure was developed which gave results similar to the in vitro procedure. Continuous use of metalaxyl may decrease the sensitivity of the pathogen population to metalaxyl.

A168

RESISTANCE IN RHIZOCTONIA SOLANI TO TOLCLOFOS-METHYL. Ariena H. C. Van Bruggen and P. A. Arneson, Cornell University, Ithaca, NY 14853

To obtain an isolate of Rhizoctonia solani with resistance to a fungicide for use in ecological studies, nine isolates (anastomosis groups 1, 2 and 4) were exposed to increasing concentrations of tolclofos-methyl, ranging from 100 to 1600 µg/ml. Some isolates were adapted to grow at the highest concentration used. Acquired resistance was retained after five transfers on a fungicide-free medium. Pathogenicity of resistant isolates were similar to that of the original isolates, but their growth rates on PDA was significantly reduced to about half of those of the original isolates. Recovery of the resistant isolates from soil was not improved on a selective medium amended with tolclofos-methyl. Because of their reduced growth rates on unamended PDA, and the lack of improvement in recovery on a selective medium with tolclofos-methyl, the resistant isolates are not suitable for their intended use.

A169

DEVELOPMENT OF RESISTANCE TO PHOSPHOROUS ACID IN PHYTOPHTHORA CAPSICI. L. A. Bower and M. D. Coffey, Department of Plant Pathology, University of California, Riverside 92521.

Isolates of P. capsici were recovered on agar containing high levels of phosphorous acid (PA), following exposure to N'-methyl-N'-nitro-N-nitrosoquanidine. Ten X greater ED_{50} values of PA were found in vitro for PA-resistant (PAR) isolates than for PA-sensitive (PAS) parental isolates. PAR isolates were also resistant to fosetyl-Na with ED_{50} values >10 X that of PAS isolates. PAR isolates were pathogenic to bare root peppers (Capsicum annuum cv Yolo Wonder) treated with of 900 µg/ml PA, whereas PAS isolates were not pathogenic at 150 µg/ml PA. PAR isolates were pathogenic to greenhouse grown peppers in soil at 4500 µg/ml PA and 4317 µg/ml fosetyl-Al (Aliette®) and PAS isolates were not pathogenic at 500 µg/ml PA or 719 µg/ml fosetyl-Al. In competition experiments, when peppers were inoculated with zoospore ratios of 1:3, 1:1, and 3:1 PAR to PAS spores, isolates were recovered from stem lesions in high proportions which were resistant to PA. This is the first report of Phytophthora resistance to PA, fosetyl-Al or fosetyl-Na.

A170

SENSITIVITY MONITORING OF ERYSIPHE GRAMINIS F. SP. TRITICI TO PROPICONAZOLE. G. R. Watson, C. C. Abbott, T. R. Young. CIBA-GEIGY Corporation, P. O. Box 1090, Vero Beach, FL 32960. Present address of senior author is Department of Plant Pathology, University of Florida, Gainesville, FL.

In order to determine the current level of sensitivity of Erysiphe graminis f. sp. tritici to propiconazole, disease samples were obtained from wheat fields in eight states. Sensitivity tests were performed on wheat seedlings grown in vermiculite in 25 mm x 200 mm glass tubes. The various treatment levels were achieved by the addition of propiconazole solutions of concentrations between 0 and 30 ppm to the vermiculite. ED_{50} values were calculated by determining the concentration of the fungicide corresponding to a 50% reduction in the percentage of leaf area supporting colonies of E. g. fsp t. The ED_{50} values for individual isolates ranged from 0.2 to 3.8 ppm, with a mean ED_{50} for all isolates of 1.5 ppm. This information will be utilized to detect shifts in the sensitivity of the pathogen population to this fungicide after commercial introduction.

A171

SENSITIVITY MONITORING OF PERONOSPORA TABACINA TO METALAXYL. C. C. Abbott, G. R. Watson, and T. R. Young. CIBA-GEIGY Corporation, P. O. Box 1090, Vero Beach, FL 32960

During 1983, 41 isolates of Peronospora tabacina were collected from naturally infected burley and flue-cured tobacco fields in the southeastern United States. Isolates were tested for sensitivity to metalaxyl at 0.01, 0.1, 1.0, and 2.5 ppm using a detached leaf method. The detached leaf method consisted of placing a metalaxyl treated leaf in a petri dish, inoculating it, and rating it 10 days later based on percent of the leaf surface on which the fungus was sporulating. All isolates of P. tabacina were sensitive to metalaxyl with ED_{50} values ranging from <0.01 to 2.0 ppm with a mean of 0.5 ppm. Monitoring of P. tabacina sensitivity to metalaxyl will continue in the future.

A172

PREVENTIVE AND CURATIVE FUNGICIDE TREATMENTS FOR CONTROL OF TOBACCO BLUE MOLD. L. J. Herr, Dept. of Plant Pathology, The Ohio State University/OARDC, Wooster, OH 44691.

In the first of two greenhouse tests metalaxyl (Ridomil 2E) drench (2.2 kg a.i./h) and foliar spray (8 ug a.i./ml) were compared with phosethyl (Aliette 80W) drench (220 kg a.i./h) and foliar spray (800 ug a.i./ml). Treatments were applied 3 days before (preventive) or 4, 7 or 9 days after (curative) inoculation of 'Ky 14' tobacco plants (5 to 7 leaves) with Peronospora tabacina (10^5 sporangia/ml). In the second test, preventive (3 days before) and curative (6 days after inoculation) metalaxyl drenches (2.2 kg a.i./h), were applied to plants (5-7 leaves) inoculated with 5 x 10^3, 2.5 x 10^4 or 10^5 sporangia/ml. Disease ratings (DR) were based on a 0-5 scale (0=healthy and 5=dead plants). In both tests, curative control was mediocre to poor (DR's=1-4, 1-3, respectively) and was dependent upon the concentration of sporangia and the fungicide applied. Preventive treatment with metalaxyl gave the best control of blue mold (DR=0) in both tests.

A173

EFFECT OF STEROL-INHIBITING FUNGICIDES ON APPLE MILDEW CONTROL, YIELD, AND FRUIT GROWTH FACTORS. Robert A. Spotts, Mid-Columbia Experiment Station, Hood River, OR 97031.

The effects of triadimefon, etaconazole, and karathane on apple powdery mildew control, fruit set, size, shape, and yield were studied on mature Newtown apple trees on seedling rootstocks in 3 successive years. Treatments were applied to 0.2 ha blocks at 561 l/ha at 1 cm green, pink, petal fall, and first cover. Triadimefon, etaconazole, and karathane rates were 0.14, 0.10, and 0.55 kg a.i./ha, respectively. Cyprex at 2.2 kg a.i./ha was applied for apple scab control with triadimefon, karathane, and to check trees wich received no mildewcide. Orchard cultural practices, insect, and weed control were applied uniformly to all trees. Triadimefon and etaconazole reduced leaf and bud mildew infection when compared with both karathane and check treatments. None of the mildewcides affected fruit set, size, or shape. Yield of trees treated with etaconazole and karathane were not different from each other but were greater than yield of triadimefon or check trees.

A174

SYNERGY BETWEEN METALAXYL AND MANCOZEB IN CONTROLLING LATE BLIGHT ON POTATOES. Yair Samoucha and Yigal Cohen, Department of Life Sciences, Bar-Ilan University, Israel 52100.

Efficacy of metalaxyl, mancozeb and mixtures of metalaxyl and mancozeb in controlling late blight (Phytophthora infestans) in potatoes (cv. Croft) was examined in growth chambers and the field. Synergy between fungicides was calculated using the formula Sf(synergy factor) = M/[P+0.01S(100-P)] where M, P, and S represent percentage control incited by mixtures, mancozeb and metalaxyl, respectively. In growth chambers, Sf values for a mixture of 3.12 µg a.i./ml metalaxyl and 25 µg a.i./ml mancozeb were: 1.22, 2.44, 2.02, 2.19, 4.33, and 5.35 for isolates 1, 2, 3, 4, 5 and 6 respectively. In field experiments, Sf values for mixtures of (µg a.i./ml metalaxyl + mancozeb) 2.5 +10, 2.5 + 20, 5 + 20, 5 + 40, 10 + 40, and 10 + 80 in plants inoculated with isolate 1 were 6.58, 5.81, 4.07, 3.73, 2.82, and 1.69 respectively. Sf values for mixtures of 3.12 + 25, 6.25 + 50, 12.5 + 100, 3.12 + 50, 6.25 + 100, and 12.5 + 200 µg a.i./ml metalaxyl + mancozeb in plants inoculated with isolate 5 were 7.12, 2.96, 1.94, 2.93, 2.02 and 1.24, respectively.

A175

CHARACTERISTICS OF PATHOGENS CAUSING PATCH DISEASES OF POA PRATENSIS IN NEW YORK. Richard W. Smiley, M. Craven Fowler and R. T. Kane, Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Phialophora graminicola (Pg), Leptosphaeria korrae (Lk) and similar unidentified fungi were identified as primary causal agents of a group of distinctively patterned patch diseases previously known as Fusarium blight syndrome of Kentucky bluegrass. Ectotrophic growth and pathogenesis by these soil-borne fungi closely resemble those of Gaeumannomyces graminis. Lk is more virulent and has a wider host range than Pg. Both tolerate a wide range of pH. Pg causes summer patch disease, is favored by warm to hot, wet conditions, and is sensitive to many modern fungicides. Lk causes necrotic ring spot, is favored by mild, dry to wet environments, and is sensitive to fewer fungicides. The characteristic ring shapes and longevity of individual patches appears to result from disease-decline phenomena.

A176

"FUSARIUM BLIGHT SYNDROME" RE-DESCRIBED AS A GROUP OF PATCH DISEASES CAUSED BY PHIALOPHORA GRAMINICOLA, LEPTOSPHAERIA KORRAE, OR RELATED SPECIES. Richard W. Smiley, Dept. of Plant Pathology, Cornell University, Ithaca, NY 14853.

P. graminicola (Pg), L. korrae (Lk), and similar soil-borne pathogens cause the distinctly patterned group of patch diseases previously known as Fusarium blight or Fusarium blight syndrome. Bioassays of presymptomatic patches, proofs of pathogenicity (including development of patches in the field), and studies of environments, fungicides and host ranges each support this finding. The name Fusarium blight is therefore rejected for well-patterned circular to arc-shaped patch diseases. Fusarium spp. should be recognized as causal agents only of diffusely distributed or irregularly patchy leaf spots and crown and root rots. The name summer patch is proposed for diseases caused by Pg. Necrotic ring spot (sensu Worf) is the name for Lk-incited diseases of cool-season grasses.

A177

A NEW DISEASE OF BLUEGRASS TURF AND ITS CONTROL IN THE PACIFIC

NORTHWEST. G. A Chastagner, R. L. Goss, J. M. Staley, and W. Hammer, Wash. State Univ., West. Wash. Res. and Ext. Ctr., Puyallup, WA 98371.

A serious disease of bluegrass turf with symptoms resembling take-all patch of bentgrass has been observed on recently established turf in eastern and western Washington since 1979. A fungus, tentatively identified as Leptosphaeria sp., is consistently associated with diseased plants which have dark ectotrophic mycelium on roots and crowns. Pseudothecia are occasionally found on roots and have been produced in culture. Ascospores measure 135 (100-188) by 4-5 µm with 7 (5-11) septa. Fenarimol and propiconazole at 1 µg a.i./ml were effective in suppressing growth of this fungus in vitro. Monthly applications of fenarimol at 29 and 56 g a.i./93 m² and propiconazole at 56 g a.i./93 m² during May, June and July provided excellent disease control. Applications of triadimefon at 28 and 56 g a.i./93 m² and sulfur at 0.9 kg a.i./93 m² were ineffective.

A178

THE CAUSE OF THE SPRING DEAD SPOT DISEASE (SDS) OF CYNODON DACTYLON (L) PERS. IN CALIFORNIA. R. M. Endo, H. D. Ohr and E. M. Krausman, Department of Plant Pathology, University of California, Riverside 92521.

The SDS disease appears as circular dead patches in the spring as the bermudagrass recovers from winter dormancy. A black to brown rot affects all the cells and tissues of the roots, stolons, rhizomes and lower culm bases. The causal fungus grows as hyphae on the surface of these organs. The presence of the dry rot, of external, round, brown sclerotia on the surfaces of the stolons, rhizomes and lower culm bases and of internal, elongated, brown sclerotia in the cortex of the roots are diagnostic for the disease. Koch's postulates were successfully completed with an ascospore isolate of Leptosphaeria korrae and two sterile isolates of an unidentified fungus which resembles L. korrae. The disease developed much more severely at 13 than at 24°C. Although L. korrae occasionally produces ascospores within pseudothecia, it does not produce conidia. The bermudagrass cultivars Tifgreen, Common and Santa Ana are susceptible.

A179

A NEW COOL SEASON PATCH DISEASE OF KENTUCKY BLUEGRASS TURF IN THE NORTHEASTERN UNITED STATES. Noel Jackson, Department of Plant Pathology/Entomology, University of Rhode Island, Kingston, RI 02881.

Take-all patch caused by Gaeumannomyces graminis (Sacc) Arx & Olivier var. avenae (E.M. Turner) Dennis, continues as an increasing & serious problem on Agrostis turf in New England. Prior to 1982 only one instance of this fungus attacking Poa pratensis turf had been confirmed in the region, and the damage was negligible. However, during the fall of 1982, symptoms similar to severe take-all patch occurred on bluegrass lawns in several New England locations and on Long Island. Pseudothecia found on infected plants and pseudothecia induced on sterile oat grains from cultures of the same fungus isolated from Poa pratensis, Festuca rubra, Agrostis spp. and Poa annua proved to be ascocarps of Leptosphaeria korrae Walker & Smith, an incitant of spring dead spot of bermuda grass in Australia. Further new outbreaks were recorded in the spring & fall of 1983.

A180

YELLOW RING DISEASE OF POA PRATENSIS. H. T. Wilkinson, Department of Plant Pathology, University of Illinois, Urbana-Champaign, Illinois 61801.

During 1978-83 in Illinois, Iowa, Wisconsin, Indiana, and New Jersey, yellowed rings (10 cm wide and 0.1-1.5 m diam.) were observed in P. pratensis turf. The yellowed rings were visible during the months of June-October. Trechispora alnicola was identified as the predominant microorganism isolated from diseased grass plants. Isolations were made by disinfesting the surface of plesionecrotic roots and crowns using 0.5% sodium hypochlorite and seeding them onto thatch medium (200 g P. pratensis thatch + 17 g agar per liter of water) adjusted to pH of 4.5 using HCl. Trechispora alnicola cultured on thatch medium was used to inoculate P. pratensis and Agrostis palustris plants. Root and crown infections were produced on, and T. alnicola was recovered from, P. pratensis; no infections occurred on A. palustris. Inoculated P. pratensis developed the characteristic yellowed leaf symptoms observed in the field. This disease of P. pratensis caused by T. alnicola has been named yellow ring.

A181

THE SELECTION OF BACTERIA ANTAGONISTIC TO PYTHIUM SPP. PATHOGENIC TO TURFGRASS. H.T. Wilkinson and R. Avenius. Department of Plant Pathology, University of Illinois, Urbana-Champaign, Illinois 61801.

A procedure was developed to identify bacteria suppressive to cottony blight (CB) of turfgrass caused by P. spp. Bacteria isolated from the rhizosphere of Triticum sativum were predominantly of the genus Pseudomonas. Each of 25 isolates displaying antagonism of Pythium growth on culture medium were assayed for suppressiveness to CB. Bacterial isolates with rifampicin insensitivity were suspended in 10^{-3} mM $MgSO_4$ and sprayed onto grass previously inoculated with a single P. sp. Disease development and epiphytic colonization were measured to determine the suppressiveness of the bacteria. The level of in vitro antagonism by a single bacterial isolate varied among the P. spp.: the level of in vitro antagonism on a single P. sp. varied greatly among bacterial isolates. Most antagonistic bacteria failed to suppress disease development. Several isolates reduced disease development by 25% compared to treatments lacking bacteria.

A182

COMPARISON OF FUNGICIDES FOR CONTROL OF PYTHIUM BLIGHT ON FESTUCA RUBRA. F.M. Ashbaugh and P.O. Larsen, Dept. of Plant Pathology, The Ohio State University, Columbus, Ohio 43210.

Chloroneb, ethazole, fosetyl aluminum, metalaxyl and propamocarb were compared for preventive and eradicant efficacy and systemic movement. Fungicides were sprayed on foliage at label rates, or soil injected at 1, 2, 4 and 8 times the label rates into pots of 'Pennlawn' red fescue. Plants were inoculated with Pythium aphanidermatum-infested millet seed, and incubated at 30C in $\geq$ 95% R.H. Chloroneb, metalaxyl and propamocarb gave excellent control when sprayed foliarly immediately prior to inoculation, but fosetyl aluminum and ethazole were not effective. Metalaxyl and propamocarb demonstrated control when sprayed up to 48 hrs after inoculation, whereas the other three chemicals did not provide control. In addition, metalaxyl and propamocarb exhibited residual activity up to 14 days after foliar application, but chloroneb failed to control disease 3 days after application. Chloroneb, fosetyl aluminum and ethazole did not provide control when soil injected; metalaxyl and propamocarb controlled disease at 0.77mg ai and 18-36mg ai/300 ml soil, respectively.

A183

A RAPID STAINING METHOD FOR DETECTION OF ENDOPHYTIC FUNGI IN TURF GRASSES. DHANONJOY C. SAHA, M. A. JACKSON, AND R. L. TATE, III., NEW JERSEY AGRIC. EXPT. STA., RUTGERS UNIV., P. O. BOX 231 NEW BRUNSWICK, NJ 08903. PUBLI. NO. K-15001-1-84.

Endophytic fungi of turf and forage grasses have been known for many years. Recently, there has been increased interest in these fungi because of the strong relationship between the presence of these fungi in grasses and resistance to certain insect pests. In order to facilitate screening a large number of plants in breeding programs, it was desirable to have a rapid staining procedure. Previously developed methods were time consuming and required careful boiling of the plant tissue. Our investigations indicated a simple, effective method of staining could be developed using rose bengal stain. The rose bengal staining procedure gave excellent visual results over a range of pH and stain concentrations. Minimal staining time (3 to 6 min.) gave good fungal differentiation even in thick tissues and no additional fixing was necessary. This simple, rapid staining technique should facilitate mass screening of plant material for the presence of endophytic fungi.

A184

PREVALENCE OF LOLIUM ENDOPHYTE IN SEED LOTS AND PLANTS OF MANHATTAN PERENNIAL RYEGRASS. Philip M. Halisky and C. Reed Funk. Cook College, New Jersey Agricultural Experiment Station, Rutgers University, New Brunswick, New Jersey, 08903.

Seed lots and plants of 'Manhattan' perennial ryegrass (Lolium perenne) were examined microscopically for the presence of Lolium endophyte (Epichloe typhina) using an alkali-soak-stain procedure. Commercial seed from early harvests of Manhattan ryegrass (1967-71) contained an average of 45 percent endophyte infection. In contrast, seed harvested in 1982-83 in both North America and Europe averaged only 6 percent infection. Loss of endophyte viability in seed used to establish recent seed production fields would appear to account for this marked reduction in endophyte content. A low-maintenance Manhattan turf seeded in 1972 showed an increase in frequency of endophyte-infected plants from below 45 percent to 91 percent during an 11-year

period. This increase indicates that endophyte-infected plants exhibited a selective advantage under the conditions of this test.

A185

A NEW DISEASE OF TURF GRASS IN CALIFORNIA CAUSED BY A UNIQUE RHIZOCTONIA. Gerard Adams and R.G. Grogan. Department of Plant Pathology, University of California, Davis, CA. 95616.

Conventional fungicide treatments failed to control yellow patch on certain California golf course greens in 1983. The pathogen isolated from diseased patches, in mid-summer and winter, resembled Rhizoctonia oryzae Ryker & Gooch. The turf Rhizoctonia and R. oryzae have 4-nuclei per cell, identical hyphal characteristics and form submerged orange sclerotia on PDA, Corn meal and water agar media. The sclerotia of the turf fungus, however, are also numerous on the agar surface and always turn a dark brown. The new Rhizoctonia from turf forms soft and waxy sclerotia unlike the hard sclerotia of Rhizoctonia zeae Voorhees. Apparently, the turf fungus is a variant strain of R. oryzae. It has a lower temperature minimum and optimum than R. oryzae isolated from rice in California and Louisiana. Koch's postulates have been satisfied and a Graminiae host-range test is in progress.

A186

ANTIGENIC RELATEDNESS OF THE NORTH AMERICAN TORONTO BENTGRASS BACTERIUM TO XANTHOMONAS CAMPESTRIS PV. GRAMINIS FROM EUROPE. D. L. Roberts and J. M. Vargas, Jr., Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

Ouchterlony gel double diffusion was used to characterize antigenic properties of a Xanthomonas spp. recently identified as causing a wilt disease of Toronto creeping bentgrass (Agrostis palustris Huds. cv. Toronto) in the midwestern United States. Antiserum prepared against an Illinois isolate cross-reacted with isolates from locations in Ohio, Michigan and Wisconsin, and with several X. campestris pv. graminis isolates known to cause wilts of forage grasses in Europe. No reaction occurred with common North American Xanthomonas spp. Serological relatedness to X. campestris pv. graminis from Europe suggests the 'Toronto' bacterium may have originated from this source.

A187

SEXUAL REPRODUCTION OF PHYTOPHTHORA CACTORUM IN A CHEMICALLY DEFINED MEDIUM CONTAINING PHOSPHOLIPIDS BUT NO STEROLS. W. H. Ko, Department of Plant Pathology, University of Hawaii, Beaumont Agricultural Research Center, Hilo, HI 96720

Phytophthora cactorum produced abundant oospores in a chemically defined medium containing 1000 ppm chromatographically purified soybean phosphatidylcholine (lecithin) in the absence of sterols. No oospores were produced in the medium when lecithin was omitted. Type III and Type V preparations of lecithin from egg yolk were less effective in inducing oospore formation. When lecithin was replaced by the same concentration of soybean phosphatidylethanolamine (cephalin) the medium was still capable of supporting oospore formation by P. cactorum. Sexual reproduction in P. cactorum also occurred in the basal medium supplemented with 1000 ppm synthetic lecithin, dioleoyl-phosphatidylcholine. These results support the previous report (Ann. Phytopathol. Soc. Japan 49:316-321, 1983) that sterols are not essential for sexual reproduction in Phytophthora.

A188

PATTERNS OF ROOT GROWTH AND INOCULUM PRODUCTION IN THE CATHARANTHUS ROSEUS - PHYTOPHTHORA PARASITICA PATHOSYSTEM. J.T. English and D.J. Mitchell, Dept. Plant Pathology, Univ. Florida, Gainesville, FL 32611.

After 2 wk growth in autoclaved soil infested with Phytophthora parasitica, sporangia and chlamydospores were produced on 23.0% of first order roots and 18.5% of second order roots of Catharanthus roseus, based on a morphometric root analysis system (Fitter, A.H. 1982. Plant Cell Env. 5:313-322). The average numbers of spores per colonized first, second, and third order root were 3.6, 2.3, and 0.0, respectively. Infected seedlings exhibited large increases in both the number and average length of each order root as compared to noninfected seedlings; the branching ratios, based on relative abundance of each root order were 6.1 and 3.9 for infected and healthy seedlings, respectively. Similar trends in root growth and pathogen reproduction were noted in the Nicotiana tabacum - P. parasitica var. nicotianae pathosystem, with the exception that considerably greater numbers of spores were formed on first order roots than on second order roots (21.5 and 7.0, respectively).

A189

THE EFFECT OF PLANT STAGE AND ROOT GROWTH ON INCIDENCE OF SCLEROTINIA WILT OF SUNFLOWER. B. D. Nelson, Dept. of Plant Pathology, North Dakota State University, Fargo, 58105.

Incidence of Sclerotinia wilt of sunflower increases dramatically following host flowering (R-5 stage). Possible explanations are (1) a change in growth stage increases host susceptibility and (2) a greater root volume and decreased root activity during flowering favors infection and spread of pathogen. To investigate these ideas, sunflowers (Hybrid 894) in the R1 (bud stage) and R5 stages were inoculated. Significantly (p=.05) fewer inoculated R-1 plants wilted; however, there was no difference between growth stages in days from inoculation to complete wilting. Lateral root length and diameter in the field reached a maximum at the R-5 stage, then declined and dead and dying roots were detected. Root contacts between plants were first observed in the R-1 stage. Results suggested root growth could be a major factor affecting disease incidence through rootsclerotia contact and plant to plant spread of the fungus via roots. Also, elimination of infected tissue by actively growing roots may reduce disease incidence of plants in vegetative or bud stages.

A190

FACTORS AFFECTING MYCELIOGENIC GERMINATION OF SCLEROTIA OF SCLEROTINIA SCLEROTIORUM. H. C. Huang, Agriculture Canada Research Station, Lethbridge, Alberta, Canada T1J 4B1.

Sclerotia of Sclerotinia sclerotiorum collected from various hosts in Western Canada varied in their ability to germinate myceliogenically. The percentage of sclerotia capable of undergoing myceliogenic germination without the addition of exogenous nutrients was high in samples from safflower and mungbeans but was generally low from sunflower. Such a drastic difference in myceliogenic germination among samples was not due to the existence of distinct strains. Rather, it was due to incomplete melanization of sclerotia in the samples. Myceliogenic germination occurred readily in the light-colored sclerotia as well as in the black sclerotia with light brown patches or with an injured rind. In addition to the difference in germination behavior, the incompletely melanized sclerotia were also contaminated with microorganisms more frequently than were the completely blackened ones. The impact of myceliogenic germination of sclerotia on the epidemiology of sclerotinia wilt in sunflower is discussed.

A191

RELATIONSHIP OF DISEASE INCIDENCE TO INOCULUM DENSITY IN SCLEROTIUM ROLFSII ROOT ROT OF PROCESSING CARROTS. Z. K. Punja, V. L. Smith, and S. F. Jenkins, Dept. of Plant Pathology, North Carolina State University, Raleigh 27695.

Mycelium from eruptively germinating (competent) sclerotia of S. rolfsii infected mature carrot roots in the greenhouse from 3 cm away at the soil surface. At depths of 1, 3, 5 and 8 cm, sclerotia infected from 2.5, 1.5, 1.0 and 0 cm, respectively. The shape of the volume of soil around the root delimited by these effective depths and distances (competence volume, C-Vol.) resembled the frustum of a cone. The C-Vols. for 8-cm-long roots of radii 1.5 and 4.0 cm (at 3 and 5 months after planting, respectively) were 190 and 403 cm^3 (average = 300 cm^3). Inoculum densities (ID) of 1, 2 or 3 sclerotia/C-Vol. resulted in 50, 80 or 100% infection, respectively, of carrot roots in the greenhouse. In the field, beds with initial ID on 1 June 1983 of 1, 3, 5, 7, 9 and 12 sclerotia/300 cm^3 had 20, 30, 35, 39, 41 and 43% disease, respectively, on 23 August. Disease increase as a function of time was linear and resulted from an increase in the number of disease foci and mycelial spread from root to root.

A192

RELATING SOIL DENSITIES OF FUSARIUM, PYTHIUM, RHIZOCTONIA, AND THIELAVIOPSIS TO DISEASE SEVERITY AND YIELD OF SNAP BEANS IN FIELD MICROPLOTS. G. S. Abawi and A. C. Cobb, Dept. of Plant Pathology, NYSAES, Cornell University, Geneva, NY 14456.

Four densities each of F. solani f. sp. phaseoli (FSP; 0, 200, 2000 and 4000 propagules [pg]/g soil), P. ultimum (PU; 0, 20, 200 and 1015 pg/g), R. solani (RS; 0, 8, 42 and 84 pg/100 g), and T. basicola (TB; 0, 200, 2000 and 20,000 pg/g) were tested in 1981, '82 and '83. Yield of Bush Blue Lake 47 was reduced the most by RS and TB. In 1983, density of RS was correlated with emergence ($r = -0.68^{**}$), root wt ($r = -0.63^{**}$), top wt ($r = -0.54^{**}$), pod wt ($r = -0.46^{**}$), and rot severity on stems ($r = 0.57^{**}$) and roots ($r = 0.59^{**}$). Densities of TB were correlated with root wt ($r = -0.67^{**}$); top wt ($r = -0.57^{**}$), pod wt ($r = -0.51^{**}$), and rot severity on stems ($r = 0.39^{*}$) and roots ($r = 0.58^{**}$). Densities of FSP were correlated only with root wt ($r = -0.54^{**}$), and rot severity on stems ($r = 0.67^{**}$) and roots ($r = 0.65^{**}$). No significant correlation was found with PU probably because warm, dry conditions prevailed.

A193

ENHANCEMENT OF CROWN ROT OF APPLE TREES WITH NITROGEN FERTILIZERS AND COMPOSTS. R.S. Utkhede, Agriculture Canada Research Station, Summerland, British Columbia V0H 1Z0

The recommended rates of nitrogen fertilizers, ammonium sulfate, ammonium nitrate, calcium nitrate and urea, two composts, cattle manure, and sewage sludge were applied to evaluate the effects of different sources of nitrogen and composts on the incidence of crown rot in apple trees caused by Phytophthora cactorum. All nitrogen fertilizer treatments showed significantly higher percent infection compared with untreated control. No significant differences in percent infection was observed between untreated control and hardwood compost, softwood compost, and cattle manure treatments. Total spur length and trunk diameter were not affected by any of the fertilizer and other treatments. These results suggest that the crown rot infection in apple trees is enhanced by the application of nitrogen fertilizers in any form at the recommended rates.

A194

RESPONSES OF SOIL INHABITING MICROORGANISMS TO VOLATILE EXUDATES FROM GERMINATING PEA SEEDS. J. M. Norton and G. E. Harman, Dept. of Horticultural Sciences, Cornell University, NYSAES, Geneva, NY 14456

In this work we determined the responses of several important groups of soil inhabiting microorganisms to volatiles from germinating pea seeds of differing quality. Populations of microorganisms indigenous to the Arkport sandy loam were exposed to the volatiles and monitored by the use of selective media. In response to volatiles from aged seed bacterial and fungal populations showed significant increases. For example, Fusarium spp. and Pseudomonas spp. showed increases of 79% and 2200% respectively over their original population levels. Conversely Pythium spp. decreased markedly. The relationship between microbial population changes and the seed rotting potential of soil was assessed using a cucumber seed bioassay. In other experiments, volatiles from aged pea seeds elicited germination and preferential growth from Rhizoctonia solani Kuhn or Sclerotium rolfsii Sacc. sclerotia that had been added to natural soil.

A195

VERTICAL AND HORIZONTAL DISTRIBUTION OF PHYMATOTRICHUM SCLEROTIA IN TEXAS SOILS. S. D. Lyda, Department of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843.

Soil cores (15 x 91 cm) were collected from fields with a history of Phymatotrichum root rot in 22 counties of Texas. Each core was extricated with a tractor-mounted, hydraulic soil sampler, extruded from the cylinder, segmented into three sections (15 x 30 cm), and wet sieved through a 40-mesh screen. Sclerotia were recovered using a sucrose flotation technique, counted, and checked for viability. Sclerotia were found in 21% of the cores with 93.3% of the total numbers found in the second and third segments (30-90 cm deep). Sclerotia were found in 20 out of 100 cores taken every 3 m in a gridded area (30.5 x 30.5 m) at the Blackland Research Center. Less than 0.1% of the sclerotia were found in the top 30 cm-segment, whereas 79.2% were found 60-90 cm deep. The same trend was observed for 10 cores taken in 10 locations (total 100 cores) in a different field at this station. The horizontal and vertical distribution of Phymatotrichum sclerotia follows clustered distributions.

A196

IRRIGATION MANAGEMENT AND CORN STALK ROT. Donald R. Sumner and James E. Hook, Departments of Plant Pathology and Agronomy, University of Georgia, Coastal Plain Station, Tifton, GA 31793

Corn was grown for three years on Tifton loamy sand (TLS) and Bonifay sand (BS) with different methods of irrigation management. Irrigation management for optimum yield maintained soil water content >50% available water in the root zone. With other irrigation methods, or without irrigation >70% depletion of available soil water in the root zone occurred during various growth stages. In both soils, stalk rot induced by Fusarium moniliforme increased as the number of days with adequate subsoil moisture increased during grain fill. There was significantly greater stalk rot on BS than on TLS following peanut or soybean regardless of irrigation management. In 1983 on BS stalk rot and yield increased as the number of days of drought stress decreased during the reproductive stage. Irrigation management had no significant influence on populations of Pythium spp., Rhizoctonia solani, and binucleate Rhizoctonia-like fungi in soils.

A197

INFLUENCE OF SOIL WATER POTENTIAL ON THE SURVIVAL AND SAPROPHYTIC ACTIVITY OF RHIZOCTONIA SOLANI AG 4 IN NATURAL SOIL. R.C. Ploetz and D.J. Mitchell. Univ. Florida, Plant Pathol. Dept., Gainesville, 32611.

The survival and saprophytic activity of Rhizoctonia solani AG 4 in a natural Arredondo fine sand was investigated in closed systems with constant water potentials(Ψ). Survival of four isolates with a localized arrangement of inoculum was monitered after 0,4, 10,25, and 55 days of incubation. After 55 days, maximum survival of all isolates was recorded at -2 and -10 bars; lower rates of survival were recorded at 0, -50, and -200mb. The survival of one of the isolates was also tested with a dispersed arrangement of inoculum; survival was monitered after 0,3,6, and 12 days of incubation. After 12 days, survival was greater at -15 bars than at -50mb, -100mb, -3.5 bars, -8 bars, -100 bars, or -1500 bars. After 12 days of incubation, colonization of rye stem segments was greater in infested soil held at -15 bars, than at -50mb, -100mb, -3.5 bars, or -8 bars; colonization did not occur at -1500 bars. The linear effects of Ψ and sampling date were highly significant in all three experiments($p=0.0001$).

A198

THE EFFECTS OF SOIL MATRIC POTENTIAL AND SOIL TEXTURE ON SPORANGIAL FORMATION BY PHYTOPHTHORA PARASITICA VAR NICOTIANAE. J. R. Sidebottom and H. D. Shew, Department of Plant Pathology, North Carolina State University, Raleigh 27695-7616.

Mycelial mats of Phytophthora parasitica var nicotianae grown on nylon mesh were incubated in the sand fraction of a sandy loam at constant matric potentials ranging from 0 to -200 millibars (mb) on Buchner funnel tension plates. After 2 days, few to no sporangia were produced at 0 and -10mb; >150 sporangia/mm^2 were produced between -40 to -200mb. Sporangial production was erratic at -20 and -30mb, with either few to no sporangia or >150 sporangia/mm^2 produced. Sporangial production was also observed in five soil materials held at constant potentials of +2 (flood), 0 (saturation), -5, -10, -20, and -40mb. The texture of these materials influenced the matric potential inhibitory to sporangial production. Inhibition occurred at -20mb in fine sand and sandy loam, -10mb in the sand fraction of a sandy loam, and at potentials >-5mb in coarse sand and silt loam.

A199

THE EFFECT OF TEMPERATURE ON PYTHIUM ROOT ROT OF HYDROPONICALLY-GROWN SPINACH. S.E. Gold and M.E. Stanghellini, Department of Plant Pathology, University of Arizona, Tucson, 85721

The effect of nutrient solution temperature on the cyclic occurrence of Pythium aphanidermatum (PA) and P. dissotocum (PD) as causal agents of root rot of hydroponic-grown spinach was investigated. Both species grew equally well and produced zoospores in comparable numbers between 17 and 27 C. Zoospores of both species penetrated spinach roots, without appressoria formation, within 15 min. after inoculation at 23 C, the transitional temperature which governed the predominance of either PA or PD. Both species also caused significant yield reductions at all temperatures tested (17, 21 and 27 C) except PA at 17 C. Severe root rot and/or plant death occurred within 4 and 7 days after inoculation with PA and PD, respectively. Differences in virulence at specific temperatures would give a temporary competitive advantage to the favored species with respect to rapidity of host colonization and subsequent fungus reproduction.

A200

THE INFLUENCE OF TRIFLURALIN ON THE BEAN ROOT ROT PATHOGEN, FUSARIUM SOLANI F. SP. PHASEOLI. C. Bareta Walker and Jack Altman, Colorado State University, Fort Collins, CO 80523

The influence of trifluralin and its formulation blank on Fusarium solani f. sp. phaseoli was studied in vitro using herbicide-amended agar at rates equivalent to field concentrations. Sporulation of the pathogen was significantly increased when grown on trifluralin amended agar. No effect was observed on vegetative growth when compared to water (control) and formulation blank treatments and colony characteristics such as aerial growth and color were similar in all treatments. Trifluralin also stimulated chlamydospore germination in field soil under laboratory conditions. Chlamydospores with more than one germ tube were occasionally observed in trifluralin treatments while none were seen in non-herbicide or formulation blank treatments.

A201

THE EFFECT OF CONSERVATION TILLAGE AND ROTATION WITH GRAIN SORGHUM ON SOYBEAN DISEASES. Minkaila Ayinla and W. M. Powell,

Dept. of Plant Pathology, Univ. of Georgia, Athens, GA 30602.

Incidence of soybean diseases in a study involving rotation with grain sorghum and different tillage systems (none, reduced, and conventional) was determined at the Southern Piedmont Conservation Research Center, Watkinsville, Georgia, during 1982 and 1983. Pod and stem blight (Diaporthe phaseolorum var. sojae) and purple stain (Cercospora kikuchii), present in both years, were not affected by tillage or rotation systems. Downy mildew (Peronospora manshurica) occurred in 1982 and was significantly increased by no tillage. Rotation of two preceding years with grain sorghum favored downy mildew development more than a continuous soybean culture. In 1983 a lethal disease complex involving stem canker (D. phaseolorum var. caulivorum) and southern blight (Sclerotium rolfsii) was significantly more severe in reduced tillage systems than in conventional tillage plots.

A202

CHARACTERIZATION OF SOUTHEASTERN BIOTYPES OF DIAPORTHE PHASEOLORUM VAR. CAULIVORA, THE CAUSAL ORGANISM OF SOYBEAN STEM CANKER. Gareth Morgan-Jones and Paul A. Backman, Department of Botany, Plant Pathology and Microbiology, Alabama Agricultural Experiment Station, Auburn University, Alabama 36849.

Within the Diaporthe phaseolorum (Cke. & Ell.) Sacc., species concept considerable genetic diversity reflected in both morphological and physiological differences at varietal level exists. Infravarietal strains of var. caulivora Athow and Caldwell, the causal organism of soybean stem canker, originating from disparate geographical sources were found to occur. Southeastern biotypes differ from northern isolates in cultural characteristics in vitro including colony appearance and color, growth rate, temperature relationships, stroma size and perithecium and ascospore morphology. The data indicate that a separate, distinct variety of D. phaseolorum may exist in the southeastern U.S.

A203

A SELECTIVE MEDIUM FOR DIAPORTHE PHASEOLORUM VAR. CAULIVORA. D. V. Phillips, Dept. of Plant Pathology, Georgia Experiment Station, Experiment, GA 30212.

Epidemiological studies of stem canker of soybean caused by Diaporthe phaseolorum var. caulivora (DPC) are complicated by the common presence of D. phaseolorum var. sojae (DPS) in plants and seed. Preliminary differentiation between DPC and DPS (or their Phomopsis anamorphs) by cultural characteristics and extent of stroma formation requires 7-14 days on most media. Confirmation requires much longer. A selective medium containing 4 fungicides (DPC medium A) was developed which totally inhibits growth of DPS, but allows DPC to grow at approximately half its normal rate. From a point source DPC will form a distinctive colony in 3 days and completely cover an 85 mm diameter plate of DPC Medium A in 7 days at 25 C. Over 60 pathogenic cultures of DPC grew well while over 30 typical DPS cultures were totally inhibited for at least 14 days on DPC Medium A. The reduced growth rate on DPC Medium A of most fungi found on soybean plants and seed allows the recovery of DPC without surface sterilization.

A204

VIRULENCE OF PERONOSPORA TABACINA DURING SERIAL PASSAGE THROUGH NICOTIANA TABACUM AND N. REPANDA. M. Reuveni, W. C. Nesmith and M. R. Siegel, Plant Pathology Department, University of Kentucky, Lexington, KY 40546.

Inoculation of Nicotiana tabacum (Ky 14) and Nicotiana repanda with Peronospora tabacina (Ky 79) previously established that the isolate was more pathogenic to Ky 14 than N. repanda. Sporangia ($2x10^2$,$2x10^3$ & $2x10^4$/ml) collected from sporulating lesions of each host (7 wk old, 4-5 leaves/plant) were passed serially through both hosts. Disease severity was evaluated seven days after inoculation. For all concentrations of inoculum collected from N. repanda, the disease severity was 4-5 times greater on Ky 14 than on N. repanda, but when the inoculum harvested from Ky 14 was used at the same concentrations, the disease severity was only 1-2 times greater. These differences were consistent for inoculum collected through six successive passages, and the differences were not affected by changing the temperature and/or wetness duration during the infection period. Inoculum obtained from chlorotic, local lesions or systemically infected leaves of N. repanda produced similar results.

A205

SYMPTOMOLOGY AND SPORANGIA PRODUCTION OF PERONOSPORA TABACINA OF NICOTIANA TABACUM AND N. REPANDA. M. Reuveni, W. C. Nesmith and M. R. Siegel, Plant Pathology Department, University of Kentucky, Lexington, KY 40546.

Nicotiana tabacum L. (Ky 14) and N. repanda plants of similar age and size (7 wks old, 4-5 leaves/plant) produced in growth chambers were inoculated with various sporangial concentrations ($2x10^2$-$2x10^4$) of Peronospora tabacina Adam. Greater disease severity characterized by extensive chlorosis, wilting and necrosis of leaves within 7-10 days followed by plant death occured on Ky 14. On N. repanda inoculated leaves curled and developed distinct chlorotic lesions that became systemic with time, but necrosis was limited. New growth was systemically infected producing extensive chlorosis and leaf curling accompanied by extensive sporulation. Sporangia production (morphology, germination and numbers) was greatly affected by the host. Sporangia harvested from N. repanda were more abundant, had more cytoplasm, were more variable in size and had lower germination than those from Ky 14.

A206

A RAPID TECHNIQUE TO ASSESS PATHOGENICITY OF SCLEROTINIA MINOR ON PEANUT. T. B. Brenneman, P. M. Phipps and R. J. Stipes, Dept. of Plant Path., Physiol. and Weed Sci., VPI & SU, Blacksburg, VA 24061.

A technique has been developed utilizing excised peanut stems to assess quantitatively pathogenicity of Sclerotinia minor isolates. Stem segments 8 cm long were wounded, inoculated with mycelial plugs of S. minor, and incubated in moist chambers at 20 C. Water-soaked lesions were visible after 1 d, and the rate of lesion expansion was used to compare pathogenicity of isolates either sensitive or resistant to iprodione and vinclozolin. The mean rate of lesion expansion for three fungicide-sensitive strains was 17.9 mm/d as opposed to 15.5 mm/d for six fungicide-resistant isolates. Host tissue also differed in susceptibility; 3 d after inoculation, mean lesion lengths for all isolates were 58.8 mm, 46.8 mm and 38.9 mm for the terminal, median and basal segments, respectively. This is a promising new method for the rapid comparison of isolate pathogenicity and can be adapted to evaluate fungicides.

A207

EFFECTS OF LEAF MISTING AND SPORE CONCENTRATION ON LATE LEAFSPOT DEVELOPMENT ON DETACHED PEANUT LEAVES. **B. B. Shew and M. K. Beute, Department of Plant Pathology, North Carolina State University, Raleigh 27695-7616**

Detached leaves from 6- to 8-wk old NC 3033 peanut plants maintained in 10 x 1 cm glass tubes containing a complete nutrient solution were inoculated with spore suspensions of Cercosporidium personatum ranging from 12,500 to 100,000 conidia/ml. Misting for 4,8,16,24, or 32 days after inoculation caused lesion numbers to increase with increasing mist periods up to 24 days. In separate tests, lesions developed after only 24 hr of continuously moist conditions. Leaves kept dry for 1,2,3, or 4 days following inoculation developed lesions when misted beginning 4 days after inoculation. In all tests, lesion numbers increased with increasing spore concentrations.

A208

FUNGI THAT CAUSE CANE CANKERS ON THORNLESS BLACKBERRY IN OHIO. G. A. Kuter and M. A. Ellis. Department of Plant Pathology, The Ohio State University/OARDC, Wooster, OH 44691.

Various cultivars of thornless blackberry (Rubus spp. hybrids, subgenus Eubatus) have been introduced into Ohio and planted commercially on a small scale. Although the potential of these cultivars ('Hull Thornless', 'Dirksen Thornless' and 'Thornfree') is great, stem cankers appearing primarily on second year canes have reduced yields and caused several plantings to be destroyed. Three fungi, Gnomonia rubi (Rehm) Winter, Leptosphaeria coniothyrium Sacc. and Botryosphaeria obtusa (Schw.) Shoemaker were consistently isolated from cankers and produced characteristic symptoms in both field and greenhouse inoculations. All three fungi have been previously associated with Rubus spp. however, pathogenicity of G. rubi and B. obtusa had not previously been shown for blackberries.

A209

TOLERANCE OF FOUR APPLE ROOTSTOCKS TO XYLARIA ROOT ROT. A. J. Latham, Dept. Botany, Plant Pathology and Microbiology,

W. A. Dozier, Jr., and J. W. Knowles, Dept. Horticulture, Ala. Agric. Exp. Stn., Auburn University, AL 36849.

Apple tree losses to root rots were recorded as they occurred in an orchard of Red Delicious planted in 1965 on the Piedmont Substation, Camp Hill. The orchard was established to evaluate various rootstocks. Rootstocks tested were M 104, M 106, M 111 and seedling; each rootstock was planted in 5-tree plots with eight replicate blocks. By 1983, the percentage losses to Xylaria mali for the various rootstocks were as follows: M 104, 55; M 106, 23; M 111, 39; and seedling 14. Trees remaining in the orchard were removed and infected roots were plated on PDA for pathogen determination. Percentages of trees representing the various rootstocks in the residual stand that were infected with X. mali were M 104, 30; M 106, 20; M 111, 36; and seedling, 26. Rootstocks least tolerant to X. mali were M 104 and M 111.

A210

DRY ROT DISEASE OF CITRUS--PREDISPOSING ROOTS TO INFECTION BY FUSARIUM SOLANI. Gary S. Bender and John A. Menge, Department of Plant Pathology, University of California, Riverside, 92521.

Dry rot of citrus is a disease characterized by death of the bark and brown staining of the scaffold roots and crown below the budunion. Attempts were made to induce the disease on Troyer citrange, rough lemon, macrophylla and sour orange seedlings by inoculating with Fusarium solani, the fungus most commonly isolated from dry rot-affected trees, but no disease resulted. A reduction in vigor of the tap root as a predisposing factor for Fusarium infection was evaluated by girdling 6-month-old Troyer citrange tap roots 1 cm below the soil line. One half of the girdled and one half of the non-girdled tap roots were inoculated 10 cm below the soil line with infected twigs. Gridled inoculated trees developed typical dry rot below the girdle within 4 weeks; non-girdled inoculated seedlings did not become infected. Symptoms included vascular plugging, reduction in adventitious roots and feeder roots, and brown staining of the wood. Girdled inoculated root dry wts. were reduced ($P < .01$) compared to controls.

A211

NATURAL AND EXPERIMENTAL PRODUCTION OF OOSPORES OF BREMIA LACTUCAE IN NEW YORK. J. E. Yuen and J. W. Lorbeer. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Oospores of Bremia lactucae were observed occasionally in infected leaves of lettuce plants collected from commercial lettuce fields in Oswego County, New York. Oospores were formed under laboratory conditions in cotyledons of the lettuce cultivar Ithaca after dual inoculation with single-sporangium isolates of opposite mating type and known virulence factor(s) from New York collections of B. lactucae. Only two mating types have thus far been detected in New York. Only one mating type was detected in each field sampled, with a single exception, where both mating types were found.

A212

PHYTOPHTHORA KATSURAE FRUIT ROT OF COCONUT IN HAWAII. **J.J. Ooka, and J.Y. Uchida. Department of Plant Pathology, University of Hawaii, Honolulu, HI 96822; and B.T. Yamamoto, Kauai Community College, Lihue, HI 96766.**

A coconut rot was observed on Kauai, Hawaii in 1982. The rot, which starts as water soaked lesions at the stem end, affects all fruit stages. The lesions coalesce and become tan then black and sunken. The disease was more prevalent during or shortly following prolonged wet periods. Phytophthora katsurae was consistently isolated from the lesions. Sporangial suspensions of a P. katsurae isolate from Lawai, and one from Wailua were prepared in sterile water and adjusted to 5000/ml. One ml of suspension was placed at the stem end of the fruit and the inoculated fruit placed in a moist chamber at 25 C for 24 hr. Incubation continued at 25 C and 70% RH for 7 days. Control fruit were treated in a like manner with sterile water. After 48 hr small water soaked lesions were visible. The lesions were tan on the third day, and by the fourth day began turning black, symptoms identical to those observed in the field. Phytophthora katsurae was recovered from these lesions.

A213

EARLY EVENTS OF GENE EXPRESSION IN COMPATIBLE MAIZE-HELMINTHOSPORIUM SPP. INTERACTIONS: POLYSOME SHIFT AND SPECIFIC TRANSLATIONAL ALTERATION. Cathy H. Wu,, H.L. Warren and C.Y. Tsai, Department of Botany & Plant Pathology, Purdue University, West Lafayette, IN 47907

To study the biochemical events at the early stage of compatible interactions, 7-day-old seedlings of maize (Zea mays L.) inbred W64A were inoculated with Helminthosporium maydis races T and O, and H. carbonum, then leaves were harvested and analyzed after 3 to 48 h. Polysome shift was obvious at 12 h after inoculation. Accompanied with the polysomal shift was a reduction in translational efficiency when polysomes were translated in vitro using wheat germ S176. These early events in compatible plant-pathogen interactions are similar to those observed in heat or anaerobiosis-stressed plants. Gel electrophoresis profiles of in vitro translation products revealed changes in the intensity of several proteins after infection. A specific diminution of four major proteins, from 23 to 33 kilodaltons, occurred in all three disease interactions but was not observed in paraquat treated seedlings.

A214

LACK OF PATHOGENICITY BY ESCHERICHIA COLI CONTAINING PLASMIDS WITH GENES MEDIATING PECTOLYTIC ENZYME PRODUCTION CLONED FROM ERWINIA CAROTOVORA SUBSP. CAROTOVORA STRAIN EC14. C. Allen, D. P. Roberts, M. Ford, V. K. Stromberg, P. M. Berman*, G. H. Lacy, and M. S. Mount*. Dept. Plant Pathol., Physiol. & Weed Sci., VPI&SU, Blacksburg, VA 24061 and *Dept. Plant Pathol., U. Mass., Amherst, MA 01003

Pectate lyase (PL) macerates plant tissue during soft rot disease. To test if E. coli strains containing cloned genes for PL production act as plant pathogens, standardized wounds in potato slices were filled with 30 µl of dilutions of suspensions containing up to 10^7cfu/ml and incubated aerobically (48 h, 28C, 100% RH). Strains of E. coli containing hybrid pBR322-E. carotovora EC14 DNA plasmids mediating production of two PLs were compared with EC14, E. coli containing pBR322, E. coli containing plasmids with E. chrysanthemi PL genes (pVS-1, pPTE-3, pPTE-7) and PL-producing Klebsiella pneumoniae. Under these conditions soft rot was caused only by EC14.

A215

TRANSLATIONAL REGULATION BY RIBOSOMAL PROTEINS IN COMPATIBLE MAIZE-HELMINTHOSPORIUM SPP. INTERACTIONS. Cathy H. Wu, H.L. Warren and C.Y. Tsai, Department of Botany & Plant Pathology, Purdue University, West Lafayette, IN 47907

A shift in polysome profile and reduction in translational efficiency were observed in maize seedlings as early as 12 h after inoculation with Helminthosporium maydis races T or O or H. carbonum. Because RNase activity did not increase significantly, these changes in protein synthetic machinery are not the result of RNA degradation. When ribosomal proteins of infected maize leaves were analyzed on SDS acrylamide gels, one new, intensed band appeared at mol wt of 92,000, and a 31,000 protein occurred possibly at the expense of a larger protein with a mol wt of 32,000. Therefore, the alteration in ribosomal proteins may explain the polysome shift and changes in translational efficiency which is an important mechanism to disarm the normal metabolism of the host in compatible interactions.

A216

HEAT PROTECTION OF SORGHUM AGAINST EFFECTS OF PERICONIA TOXIN. Elbert A. Traylor and Larry D. Dunkle. Dept. of Botany and Plant Pathology. Purdue University. W. Lafayette, IN 47907.

Incubation of sorghum seedlings at elevated temperatures prior to treatment with the host-specific toxin from Periconia circinata (PC) protected the seedlings against toxin-induced loss of electrolytes and milo disease symptoms. In roots of susceptible seedlings, treatment with PC-toxin increased the rate of synthesis of four 16 kD proteins, each with a different pI. A heat shock of 40 C for 1 hr reduced the synthesis of those toxin-induced proteins and increased the rate of synthesis of a set of heat shock proteins. The results suggest that the 16 kD proteins are involved in development of disease symptoms or that the heat shock proteins otherwise protect seedlings against toxin-induced stresses.

A217

REVISED STRUCTURE OF PHASEOLOTOXIN. **Oliver C. H. Kwok, R. E. Moore and Suresh S. Patil, Dept. of Plant Pathology and Dept. of Chemistry, University of Hawaii, Honolulu, Hawaii 96822.**

Phaseolotoxin and five minor homologs were isolated from culture of Pseudomonas syringae pv. phaseolicola, the causal agent of halo blight of beans, by charcoal adsorption, QAE ion exchange chromatography, ion exchange and reverse phase HPLC. ^1H, ^{13}C and ^{31}P NMR studies of phaseolotoxin confirmed the presence of the previously reported tripeptide moiety and its

point of attachment to phosphorus. Fast-atom-bombardment mass spectroscopy of phaseolotoxin, however, revealed that the inorganic portion possessed an additional NH and one less O. ^{31}P NMR spectroscopy of ^{15}N-labelled phaseolotoxin provided conclusive evidence for the presence of a phosphorus directly bonded to three nitrogen atoms instead of one. Thus, the structure of phaseolotoxin should be revised to $^{-}O_3SNHP(O)$-$(NH_2)NH(CH_2)_3CH(NH_3^+)CO$-alanylhomoarginine. Two toxic compounds were isolated from infected bean plants. The major compound, which we named octicidin, was identical to the metabolite produced by treating phaseolotoxin with peptidase.

A218

ETHYLENE PRODUCTION BY MYCOSPHAERELLA CITRI AND GREASY SPOT-INFECTED CITRUS LEAVES. J. H. Graham, J. O. Whiteside and C. R. Barmore, Univ. of Florida, Citrus Research and Education Center, Lake Alfred, FL 33850.

Mycosphaerella citri, the greasy spot fungus, produced ethylene in a glucose - mineral salts medium ammended with methionine. Rough lemon and grapefruit leaves inoculated with M. citri produced ethylene. Ethylene production by rough lemon leaves began 3 wk after inoculation when M. citri had penetrated into the air spaces of the spongy mesophyll and symptoms of leaf chlorosis were first apparent. Ethylene production remained low for 5 wk during symptom development, but increased prior to leaf abscission. Spray oil, which reduces greasy spot-induced leaf chlorosis and abscission, reduced ethylene production and symptoms development when applied at 3 and 5 wk after inoculation. Oil is not fungitoxic but appeared to alter the physiological effect of ethylene on leaves. Prior application of oil reduced chlorophyllase activity but not respiration of excised grapefruit leaves exposed to 1-2.5 ppm ethylene continuously for 168 h.

A219

EFFECT OF CORN ROOT INFECTION BY FUSARIUM MONILIFORME ON HYDRAULIC RESISTANCE. R. W. Schneider, Department of Plant Pathology and Crop Physiology, Louisiana State University, Baton Rouge 70803

Analysis with a water ransport model of the diurnal cycles of diffusive resistance and water potential of field grown plants infected nonsystemically with Fusarium moniliforme following predisposition by an early-season water deficit had transient increases in whole plant hydraulic resistance (r). Nonsystemically infected plants grown under steady-state conditions at relatively low vapor pressure deficits (VPD) had significantly higher and constant r compared to noninfected and infected plants that had not been predisposed by a water deficit. However, infected, predisposed plants grown under higher VPDs showed the same cycling of r as was observed in the field. These results indicate that the source of increased r dissipated as a function of water flow through the plant. The site of increased r and a proposed mechanism will be discussed.

A220

WILT-INDUCING-FACTORS FROM COTTON INFECTED WITH PHYMATOTRICHUM OMNIVORUM (SHEAR) DUGGER. P.J. Cotty and I.J. Misaghi, Department of Plant Pathology, University of Arizona, Tucson, Arizona, 85721.

A wilt-inducing factor(s) (WIF) similar to those produced by Phymatotrichum omnivorum in vitro was isolated from field-grown cotton plants infected with the fungus. Cortex and bark tissues were removed from lyophylized basal stem segments of healthy and infected cotton plants. The remaining xylem and pith were ground in a Wiley Mill. Ground material was extracted with 0.1 N NaOH (pH 13). The aqueous fraction was centrifuged and passed through a 0.22 um filter. WIF was precipitated by adjusting the pH to 4.0 and recovered by centrifugation. Dissolution at pH 13 and precipitation with methanol and/or pH adjustment was repeated 3 to 5 times. Diseased plants, but not healthy, yielded a brown, high-molecular-weight (above 1 million daltons) substance(s). A 30 mg WIF/ml solution significantly reduced transpiration of excised 20-day-old cotton plants (2 true leaf stage) within 2 hr and induced wilting within 4 hr.

A221

PRODUCTION BY XANTHOMONAS ALBILINEANS OF AN ANTIBIOTIC INHIBITING DNA SYNTHESIS IN ESCHERICHIA COLI. Robert G. Birch and Suresh S. Patil, Dept. of Plant Pathology, Univ. of Hawaii, Honolulu, Hawaii 96822.

Chlorosis-inducing isolates of Xanthomonas albilineans, the sugarcane leaf scald pathogen, produced a mixture of antibacterial compounds in culture. The mixture, which eluted as a single strongly retarded peak from Sephadex LH-20 in methanol, was bactericidal to Escherichia coli. Inhibition of E. coli was not reversed by added nutrients, and affected cells were not lysed but many accumulated polyphosphate granules. The major antibacterial component, isolated in crystalline form after HPLC, is given the trivial name albicidin. Near the minimum inhibitory concentration, albicidin caused a rapid and complete block to DNA synthesis, followed by partial inhibition of RNA and protein synthesis. Spontaneous antibiotic-resistant mutants of E. coli showed no cross-resistance between albicidin and inhibitors of either subunit of DNA gyrase. Binding of albicidin to purified E. coli DNA does not seem to occur.

A222

DEVELOPMENT OF INFECTION CUSHIONS OF RHIZOCTONIA SOLANI ON ARTIFICIAL SURFACES. V. N. Armentrout and A. J. Downer, Biological Sciences Dept., Cal State Polytechnic University, Pomona, CA 91768, U.S.A.

Hyphae of Rhizoctonia solani aligned with grooves on polystyrene replicas of cotton hypocotyl surfaces, displayed shortened internodes, and produced "feet" (T-shaped lateral branches). Cushion-like masses were formed after 48 hours. These replicas were more efficient than flat polystyrene surfaces at inducing internode shortening. Internodes were shorter (compared to agar cultures) on flat surfaces of polystyrene, cellophane, and Parafilm. Bilobed branches were observed on Parafilm, whereas appressorium-like swellings were formed at the ends of lateral branches on cellophane or flat polystryene. Behavior of the fungus on scratched Parafilm resembled that on replicas of the hypocotyl surface, with hyphal alignment and foot formation. Nutrient (sucrose or hypocotyl exudate) was required for development of these early stages of infection cushion formation on all surfaces.

A223

CHANGES IN ASPARAGINE/ASPARTIC ACID RATIOS IN THE LEAVES OF THE GROUNDNUT (= PEANUT), ARACHIS HYPOGAEA IN RELATION TO INFECTION BY GROUNDNUT CHLORITIC SPOT VIRUS. **Vedam Chandrasekharam and Manam V. Bharani Kumar, Research Centre in Biology, Department of Zoology, S.G.S. Arts College, Tirupati, India 517501**

The ratios of aspartic acid (ASP) and its omega-amide asparagine (ASP-NH2) have been found to exhibit an interesting trend in the leaves of Arachis hypogaea, in relation to infection by the groundnut chlorotic spot virus. In the early stages of infection ASP-NH2/ASP ratio, will be very high. At the time of symptom-expression, this ratio makes a sharp decline, indicating utilization of the omega-amide in virus-oriented bio-syntheses. A rise in this ratio in the recovery phase, designated as 'asparagine-lock', is presumed to be causal for the "famine of nitrogen amidst plenty of it" leading to 'oligogynophoria'.

A224

CHANGES IN CONCENTRATION OF RIBONUCLEIC ACID IN THE LEAVES OF ARACHIS HYPOGAEA L. UNDER GROUNDNUT CHLOROTIC SPOT VIRUS-INFECTION. **Talisetty Haragopal, Department of Botany, S.V.Jr. College, Tirupati, India 517501**

The profile of changes in concentration of ribonucleic acid (RNA) was followed in the leaves of the groundnut (= peanut) Arachis hypogaea L. under infection with the groundnut chlorotic spot virus. The changes in concentration of RNA were remarkable and the maximal elevation of RNA coincided with the stage of symptom-appearance. Thus the present virus may be deemed to be ribo-nucleo-virus. Dedicated to Prof. Theodore O. Diener.

A225

SPECIFIC METABOLIC ORIENTATION OF THE GROUNDNUT CHLOROTIC SPOT VIRUS TOWARDS A TRANSLOCATED NITROGENOUS COMPOUND OF THE HOST, ARACHIS HYPOGAEA L. **Talisetty Haragopal, Department of Botany, S.V.Jr. College, Tirupati, India 517501**

In the free amino acid pool of leaves of Arachis hypogaea under infection by the groundnut chlorotic spot virus (GCSV) gamma-methylene glutamic acid (gamma-MGA) shows significant quantitative altera-

tions correlatable with infection and symptom-development. Symptom-fading coincides with a stage of growth of the host when the level of gamma-methylene glutamine (gamma-MG), the translocated source of gamma-MGA of leaves, is reduced considerably. The data, prima facie, argue for a nexus between translocation of gamma-MG and susceptibility of the host to GCSV.

A226

IDENTIFICATION OF COMMON BEAN GERMPLASM WITH LOW BEAN COMMON MOSAIC VIRUS SEED TRANSMISSIBILITY. J.C. Faria. National Rice and Beans Research Center, Goiânia, Goiás, Brazil.

Field and greenhouse inoculation tests conducted with BCMV strain "GO-1" utilizing 200 entries obtained from CNPAF germplasm bank showed that 18 did not present any visible symptoms. Out of 182 entries exhibiting symptoms 15 presently cultivated varieties were chosen for detailed transmissibility tests. The lowest seed-transmitting line (Costa Rica-GF 0014) showed 2.8% transmission as compared to 82.6% in cultivar Manteiga (GF 0942). The commonly planted cultivar Rico 23 exhibited 24.6% BCMV transmissibility. Low incidence of BCMV seed transmission may prove to be of great value to reduce primary inoculum level especially for low income bean growers raising their own seed.

A227

FIELD EVALUATIONS OF MAIZE INBREDS AND POPULATION SELECTIONS FOR RESISTANCE TO EXSEROHILUM TURCICUM, RACE 2. W.L.Pedersen, Dept. of Plant Pathology, University of IL, Urbana, IL 61801.

Eighty-five inbreds and nine selections from BS8, an Iowa broad-base synthetic, were evaluated for chlorotic-type lesion resistance to race 2 of Exserohilum turcicum, the causal agent of Northern leaf blight of maize. Plants were inoculated with an indigenous isolate of E. turcicum, virulent on germplasm with the Ht1 resistance gene, at Williamsburg, IA (1981-1982) using ground leaf tissue and at Urbana,IL and Mt. Joy, PA (1982) using a conidial suspension (1×10^4/ml). Eleven inbreds and three selections were rated resistant at all locations. Two inbreds and one selection were rated resistant at PA and IL, but susceptible at IA. Only one inbred was rated resistant at IA and IL, but susceptible at PA. These results indicate that screening for disease resistance should be done at multiple locations with more than one isolate.

A228

EVALUATION OF THE RESIDUAL EFFECTS OF THE Ht_1 GENE WITH RESPECT TO EXSEROHILUM TURCICUM RACE 2 IN MAIZE INBREDS AND HYBRIDS. S. Leath and W. L. Pedersen, Department of Plant Pathology, University of Illinois, Urbana, IL 61801

Field studies in 1982 and 1983 showed that four near-isogenic maize hybrid pairs, A619xA632, Mo17xA634, Mo17xN28 and B73xMo17, either homozygous recessive or dominant for the Ht_1 gene, differed in resistance to race 2 of Exserohilum turcicum. Differences existed between and within pairs. These four near-isogenic pairs also were evaluated under greenhouse conditions for resistance to Bipolaris maydis, B. zeicola and Colletotrichum graminicola. Inbreds A634, Va26 and B84 either dominant or homozygous recessive for the Ht_1 gene, were obtained by selfing plants from BC_7 generations. These sets and their recurrent parents were evaluated for resistance to E. turcicum, race 2. Disease evaluations in both studies were based on lesion size, disease efficiency and incubation period. Lesion size data indicated that differences in resistance to E. turcicum race 2, within pairs, may not be solely due to the Ht_1 gene.

A229

QUANTITATIVE CHARACTERISTICS OF RESISTANCE TO GIBBERELLA ZEAE EAR ROT IN SELECTED FIELD CORN CROSSES. E. H. Gendloff, E. C. Rossman*, and L. P. Hart, Departments of Botany and Plant Pathology and *Crop and Soil Sciences, Michigan State University, East Lansing, MI 48824

Ears of parents, F_1's, F_2's and backcrosses of all four combinations of susceptible (B79 or Mo17HT) by resistant (Pa347 or A509) field corn inbreds were inoculated with toothpicks infested with Gibberella zeae U5373. Resistance data were analyzed for environmental effects as well as mean, additive, dominance, and the three epistatic (aa, ad, and dd) gene effects. Analysis by t-test showed that mean and additive gene effects were significant in all four combinations. The significance of dominance and epistatic effects was variable among the crosses.

A230

Analytic Breeding of Alfalfa for Resistance to Phytophthora Root Rot. M. J. Havey and D. P. Maxwell, Depart. of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Analytic breeding involves ploidy reduction of tetraploid (4x) alfalfa to the diploid (2x) level, breeding and selection at the 2x level, and retetraploidization. Phytophthora root rot (PRR) resistance factors were transferred from a 4x plant to the 2x level by use of a triploid bridge. Inheritance of these factors was studied, taking advantage of disomic ratios. Two modes of inheritance were found: resistance conditioned by a dominant allele at either of two independently segregating loci and resistance conditioned by a dominant allele at both of two independently segregating complementary loci. Resistance to PRR was also identified in 2x Medicago falcata and was inherited as a dominant allele at either of two independently segregating loci. The relationships between the loci were studied in hybrids between M. falcata and the derived diploids of M. sativa. Retetraploidization will be achieved by direct 2x-4x crosses, using genetic markers to identify hybrid progeny.

A231

FIELD TESTING AND PRELIMINARY PROGENY EVALUATION OF ALFALFA REGENERATED FROM CELL LINES RESISTANT TO THE TOXINS PRODUCED BY FUSARIUM OXYSPORUM F. SP. MEDICAGINIS. C. L. Hartman*, T. R. Knous, Dept. of Plant Science and T. J. McCoy, USDA/ARS, University of Nevada, Reno, NV 89557

Plants regenerated from alfalfa cell lines selected in vitro for resistance to the toxins produced by F. oxysporum were field tested for disease response. Regenerated plants from unselected cell lines were included in the disease nursery. Disease resistance in plants from selected cell lines were equal or greater than the resistant check. Some plants from non-selected cell lines had some degree of resistance, but most remained highly susceptible. The resistance response in vitro is genetically transmissible and appears to be due to a dominant mutation, although the exact genetic control has not been established.

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A232

THE POTENTIAL OF A MASS SCREENING TECHNIQUE FOR SELECTION OF PISTACHIO ROOTSTOCK MATERIAL RESISTANT TO VERTICILLIUM DAHLIAE. L. J. Ashworth, Jr., University of California, Berkeley 94720.

Pistacia atlantica seedlings were used as the rootstock for over 90% of California plantings before it was discovered to be highly susceptible to V. dahliae. A source of P. integerrima recently proved to be useful in heavily infested soils but the need exists for other resistant rootstock sources. Mass plantings of four species were made in a field with an I.D. of about 20 microsclerotia/g soil of V. dahliae in 1982. Populations in 1983 were P. atlantica, 2,044 trees with 7.8% hybrids; P. integerrima, 3,144 trees with 30.8% hybrids; P. terebinthus, 23,748 trees with 4.2% hybrids; and P. vera, 13,444 trees with 1.1% hybrids. Mortality among homotypic seedlings was 21% for P. atlantica, 2.2% for P. integerrima, 5.4% for P. terebinthus, and 8.1% for P. vera. High susceptibility of hybrid seedlings was confined to those crosses of P. atlantica with other species, suggesting that susceptibility may be dominant over resistance. Appropriate crosses between mother trees and pollen sources were made in 1984 to test this hypothesis.

A233

MAINTAINING IMMUNITY AND HIGH RESISTANCE IN COTTON TO RACES OF XANTHOMONAS COMPESTRIS PV MALVACEARUM. L. S. Bird, K. M. El-Zik, P. M. Thaxton, M. Howell, and R. G. Percy, Dept. of Plant Pathology and Microbiology, Tex. Agr. Exp. Sta., College Sta., TX 77843.

Nineteen races of the bacterial blight pathogen of cotton are recognized. Immunity to these races was developed by 1964 using inoculum of 4 U.S. races to select effective gene combinations. Use of this procedure in the multi-adversity resistance (MAR) program gave immunity in a number of modern cultivars. New isolates HV-1, 3 and 7 from the Upper Volta and one from the Sudan of Africa are now virulent on cottons which had been immune for 20 years. Immunity to the 4 new African isolates was recovered when HV-3 and 7 and U.S. races 1,2,7 and 18 were used to select parental material prior to crossing and selecting in the F_1 and F_2. No immune plants were recovered with this inoculum when selection in similar material was applied only in the F_2 and F_3. Direct selection using a few races of the pathogen continues to be effective for recovering immunity to many races in MAR germplasm.

A234

RESISTANCE TO PHYMATOTRICHUM OMNIVORUM IN COTTON. L.S. Bird, K.M. El-Zik, P.M. Thaxton, R.G. Percy and G.R. Lazo. Dept. of Plant Pathology & Microbiology, Tex. Agr. Exp. Sta., College Sta., TX. 77843.

Phymatotrichum root rot of cotton occurs in Southwest U.S.A. and in Mexico. Differences in response among cultivars and genetic improvement for resistance have been noted and this is an expectation within the multi-adversity resistance (MAR) genetic system. Cultivars were evaluated in field nurseries with uniformly infested soil in a 1982 and in two 1983 tests at Temple. Coefficients of variation were higher in paired comparisons among resistant strains than in comparisons with and among susceptible ones for dead plants and rate of maturity. At maturity CAMAS, SP21S and CABCS' (resistant) with 30, 36 and 40% dead plants respectively were significantly (F-test) different from CABU'CS and CAHUS (susceptible) with 61 and 62% dead plants respectively. CAMD-E was intermediate, 50% dead plants, and equal to CABCS', CABU'CS and CAHUS but different from CAMAS and SP21S. The MAR system is developing cottons differing in response to root rot.

A235

ASSESSING RESISTANCE OF FOUR AVOCADO ROOTSTOCKS TO PHYTOPHTHORA CINNAMOMI USING A LABORATORY SCREENING TECHNIQUE. T. E. Dolan and M. D. Coffey, Department of Plant Pathology, University of California, Riverside 92521.

Clonally propagated selections D7, G6, G1033, and G755c were screened in a laboratory procedure. The G1033 and G755c are new selections and we compared their performance with that of D7 and G6 rootstocks showing low levels of field tolerance to Phytophthora cinnamomi (PC). Sections of young etiolated stem tissue were point-inoculated with 1000 PC zoospores. After three days incubation each segment was examined and mean lesion length (MLL) as well as percent recovery (PR) is recorded. Mean lesion length and PR for D7 and G6 are 26 mm., 84% and 25 mm., 57% respectively. Selection G755c shows a high level of resistance with a MLL of 2.0 mm. and 14% PR. This observation is supported by the early success of G775c in field trials. Selection G1033 shows a level of resistance superior to D7 and G6 with MLL of 6.2 mm. and 27.7% PI. Preliminary screening of intact roots from each selection suggests there is a good correlation of root resistance/susceptibility with stem tissue behavior.

A236

EVALUATION OF THE COMPONENTS OF RATE-REDUCING RESISTANCE IN VICIA FABA TO UROMYCES VICIAE-FABAE. M.K. Bhalla and C.C. Bernier Dept. of Plant Science, Univ. of Manitoba, Winnipeg,MB R3T 2N2.

The rate-reducing ability of 8 faba bean accessions was evaluated under field conditions in 1982 and 1983. Components of resistance to 2 single-pustule isolates of Uromyces viciae-fabae were measured at weekly intervals following inoculation at 3 plant ages. Incubation period, latent period, infection frequency, lesion size, range in lesion size, lesion cover, spore production and necrosis were recorded on 7 accessions which had consistently low AUDPC values in 3-4 years of preliminary field screening and on a single fast-rusting accession. Plots were arranged at 2 sites at Winnipeg in a RCBD with 6 replications. The most important component in the slow-rusting response was infection frequency and on this basis accessions could be ranked in terms of their relative slow-rusting ability. This character appears to have remained constant when evaluated with both isolates over 2 seasons. Latent period was significant in at least 3 accessions. Rust development decreased with plant age until pod-fill, then increased.

A237

BELLY ROT RESISTANCE IN CUCUMIS SATIVUS. R. L. Clark and C. C. Block. Regional Plant Introduction Station, Ames, IA 50011

High resistance (immunity?) to belly rot (Rhizoctonia solani) has been found in PI 165509, Cucumis sativus var. sikkinensis from India. Resistance appears, on the basis of F2 populations, to be due to a single, dominant gene. Black Diamond and National Pickle were used as susceptible parents. Inoculations were made by placing field grown fruits on infested sand benches topped with sterile field soil. Inoculum was prepared by growing R. solani on a substrate of 10g corn meal, 250ml white sand, 250ml perlite, and 100ml distilled water for 7-10 days. Approximately 15g of this inoculum was spread over 900cm^2 of the sand bench and allowed to incubate 24 hr before use. Susceptible fruits rotted within 3-5 days, resistant fruits held up for at least 10 days. Fruits of PI 165509 held up indefinitely. Four other lines from India (PI 197085, 197086, 197087, 197088) also have good levels of belly rot resistance.

A238

MEASUREMENT OF RELATIVE RESISTANCE OF SOYBEAN CULTIVARS TO STEM CANKER. B. L. Keeling, USDA, ARS, Stoneville, Ms 38776

The relative resistance of 10 soybean [Glycine max (L.) Merr.] cultivars to stem canker disease caused by Diaporthe phaseolorum (Che. & Ell.) Sacc. var. caulivora Athow & Caldwell was measured. Sixty days after seeding cultivars in sandy loam field plots, 20 plants per cultivar, replicated 3 times, were inoculated by inserting a toothpick infested with the pathogen into the soybean stem 10 cm below the apical meristem. Stems were split and lesion development measured 30 days after inoculation. Tested cultivars were separated into 5 reaction classes: 1) resistant- Tracy, Braxton (lesions 5-6 mm); 2) moderately resistant- Jeff, Centennial, Hood (lesions 14-52 mm); 3) intermediate- Semmes (lesions 50-70 mm); 4) moderately susceptible- Bedford, Forrest, Bragg (lesions 107-168 mm); 5) susceptible- J77-339 (lesions 269-380 mm). Reactions obtained by artificial inoculation agree with results obtained in field trials under natural disease development.

A239

RESISTANCE IN SOYBEANS TO DIAPORTHE PHASEOLORUM VAR. SOJAE. R. D. Berger and K. Hinson, Depts. of Plant Pathology and Agronomy, Univ. of Florida, Gainesville 32611

Nearly 1000 breeding lines and 35 cvs of soybean were rated in the field for resistance to several diseases. None of the 35 cvs was ranked in the top 100 entrees for resistance to Diaporthe phaseolorum var. sojae (Dps). In harvested seed from the 21 most resistant lines, incidence of Dps and other pathogens was 0-5%; in >800 susceptible lines, incidence of infection was 100% and many seeds were severely shriveled. Eleven of the 21 resistant lines had a common lineage to PI's 227687 and 229358 (chosen for their insect resistance). Two other Dps-resistant lines were descendants of the Brazilian cv Santa Maria. The Dps-resistant lines also have resistance to several insects, root knot nematodes, and other diseases (purple seed stain, downy mildew, anthracnose, frog eye, bacterial pustule, and mosaic). Some of the resistant lines also had 5-10% more yield than commercial cvs in separate tests.

A240

COMPLETE TOLERANCE AND IMMUNITY TO TOMATO BIG BUD DISEASE IN A LYCOPERSICON PERUVIANUM LINE AND ITS TOMATO HYBRID PROGENY. P.E. Thomas and Sher Hassan, IAREC, P.O. Box 30, Prosser, WA 99350.

Plants of Lycopersicon peruvianum, PI line 128655, and certain of its F_4 tomato hybrid progeny (L. peruvianum x L. esculentum) were graft inoculated with tissue from a field-grown tomato plant, collected near Prosser, WA in 1981, expressing typical symptoms of tomato big bud disease. None of the L. peruvianum or hybrid plants developed symptoms, but the big bud-infected scions grafted to the plants continued to express typical symptoms. The L. peruvianum and hybrid plants were indexed for presence of the big bud organism by graft inoculation to susceptible tomato plants. Big bud was recovered from about 30% of the plants. We previously demonstrated that the same L. peruvianum line and hybrid progeny also contained plants with tolerance and immunity to beet curly top, tomato yellow top, and potato leafroll viruses.

A241

REACTION OF INTERMEDIATE WHEATGRASS TO LEAF SPOT DISEASES. J. M. Krupinsky and J. D. Berdahl, ARS-USDA, Northern Great Plains Research Center, Mandan, ND 58554-0459.

Intermediate wheatgrass (Agropyron intermedium) was evaluated for reaction to various leaf spot diseases under natural field conditions. The twelve cultivars or strains included were Greenleaf, Mandan 759, Slate, Trigo, PI 345526, Neb 314054, SC I3702, SC I3712, SD 50, SD 2-15, SD 4-14, and SD 10-14. In 1980, 4,800 spaced plants were established and evaluated for three years. Each plant was rated for leaf spot diseases on a 1-9 scale. SD 2-15 was rated as having the highest level of resistance and Trigo the lowest for all three years. Neither flowering date nor plant height was associated with resistant or susceptible reaction types. There was a significant difference among years and among cultivars and strains evaluated. Cochliobolus sativus was considered to be the most important pathogen. Pyrenophora trichostoma, and Leptosphaeria nodorum also were widely distributed in the plots.

A242

EVALUATION OF FUNGICIDE ADVISORY SYSTEMS USING HISTORICAL WEATHER DATA. C. S. Johnson and M. K. Beute, Department of

Plant Pathology, North Carolina State University, Raleigh, NC 27695-7616.

Reduction in frequency of fungicide sprays based on the North Carolina and Virginia leafspot spray advisory systems was estimated from a 30 year historical record of daily precipitation. Two consecutive days with rainfall of no less than 0.10 inches or single rain events of at least 0.25 inches were used as criteria for the estimated issuance of a spray advisory. The number of sprays initiated on the basis of either criterion was similar to the number applied in field tests of the North Carolina and Virginia advisories conducted in 1981-1983. Results indicated that fewer sprays would have been applied during each of the last 30 years if applications were made based on an advisory model rather than a calendar schedule. Between 1 and 3, or 1 and 5, sprays per year would have been saved using the two consecutive rain-days or single rain-event criterion, respectively.

A243

EFFECTS OF HOST AGGREGATION AND DISTRIBUTION OF INITIAL INOCULUM ON THE EFFICACY OF HOST MIXTURES FOR DISEASE MANAGEMENT. C. C. Mundt and K. J. Leonard, Dept. of Plant Pathology, North Carolina State Univ., Raleigh, NC 27695-7616.

In previous studies with one initial disease focus per plot (Phytopathology 72:1006), oat crown rust increased as rapidly in multilines with genotypes randomly distributed as in plots with plants of the same genotype aggregated into blocks to simulate multilines of crops with larger plants. In a subsequent study using a general inoculation, we have found that aggregating genotypes markedly increases the rate of crown rust development in mixtures. Results from a modified version of EPIMUL, a computer simulator, indicate that for steep dispersal gradients (as with the small grain rusts), host aggregation will reduce the efficacy of host mixtures if the initial inoculum is uniformily or randomly distributed, but not if it is concentrated in widely dispersed foci. Our results are relevant to the use of host mixtures for crops with large plants and to the use of other diversification strategies such as strip-cropping.

A244

MANAGEMENT STRATEGIES FOR CERCOSPORA LEAFSPOT OF SUGAR BEET BASED ON DISEASE SEVERITY/CROP LOSS RELATIONSHIPS AND DISEASE PROGRESS RATES. W. W. Shane and P. S. Teng. Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Cercospora leafspot caused by Cercospora beticola Sacc. is the major pest of sugar beet in Minnesota and North Dakota. Crop losses were detected in research plots with 2% leaf severities at the end of August and 3% by harvest in 1983. Late season epidemics decreased sugar purity of roots while long-term epidemics decreased purity, tonnage, and sugar content. Disease severities increased as much as two-fold in one week on susceptible cultivars. A disease severity "safety line" management guide was devised based on crop loss thresholds, worst-case scenarios for disease increase and time left until harvest. Fungicide applications are recommended to avert crop losses if disease severities climb above the "safety line". Additional guidance for borderline cases is provided by a cercospora leafspot model which uses temperature and relative humidity information to indicate the potential for infection.

A245

PUCCINIA STRIIFORMIS F.SP. HORDEI (PSH): CAUSE OF BARLEY YELLOW RUST EPIDEMIC IN SOUTH AMERICA. H.J. Dubin, CIMMYT, Box 2600, Quito, Ecuador, R.W. Stubbs, IPO, Box 42, Wageningen, Holland.

During 1975, a new disease of barley was identified in the savannah of Bogota, Colombia causing severe damage. Field observations and experimental evidence showed the pathogen to be PSH, race 24. PSH moved to Ecuador, Peru, and Bolivia between 1976-1978; into Chile in 1981 and Argentina in 1982. Behavior of the pathogen leads to the conclusion that it is exotic to South America. The race identification coincided with the prevalent race in Europe, race 24. Between 1975-1983 field trap nursery data showed variants appearing virulent on cultivars Varunda, Mazurka and I-5. The cultivars Emir, Bigo, and Abyssinian 14 have maintained their resistance. In 1981 collections from Hordeum muticum in Peru proved to be PSH, race 24, indicating that wild barley's in the Andes could serve as reservoirs. Yield reductions due to PSH in Ecuador have been estimated at 29%. Appropriate PSH resistance is present in such cultivars as: Quibenras (Colombia), Dorada (Ecuador), UNA 80 (Peru), and IBTA 80 (Bolivia).

A246

PRODUCTION OF STERILE LEAF SURFACES FOR PHYLLOPLANE COLONIZATION STUDIES. Linda Kinkel and J. H. Andrews, Department of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706

A method for sterilizing apple leaves in situ was developed to facilitate a study of microbial colonization of the phylloplane. Leaf blades from greenhouse and orchard trees were immersed in acidified ClO_2, NaOCl, H_2O_2, ethanol, propanol, and sodium thiosulfate at 2-3 concentrations each for 30, 60, 75, 180, and 300 seconds. Relative sterility and leaf damage were assessed. A 75-second immersion in 15% H_2O_2 was optimal, resulting in sterile leaves with minimum damage. H_2O_2 treated and untreated leaves were compared at intervals for differences in gross morphology, abscission time, internal structure, stomatal function, and epiphytic microbial populations. Abaxial bronzing was the major effect of peroxide treatment; no other significant differences between treated and untreated leaves were noted.

A247

INTERACTIONS OF MYCOSPHAERELLA GRAMINICOLA AND PUCCINIA STRIIFORMIS ON WHEAT. Ricardo Madariaga B. and A. L. Scharen, USDA, ARS, Dept. of Plant Pathology, Montana State University, Bozeman, MT 59717.

Stripe rust, caused by Puccinia striiformis, and septoria tritici blotch, caused by Mycosphaerella graminicola, frequently occur together on wheat. Spring wheat seedlings of known reaction to the two pathogens were inoculated at different time intervals under conditions that favored both diseases. Leaf area affected by each pathogen, occurrence of pycnidia,and dry weight of leaves were recorded. A smaller amount of leaf area was affected by each pathogen when they shared the same tissue than when either was alone. Leaves having sporulating rust pustules were heavier than leaves with both pathogens, leaves with septoria tritici blotch alone, or leaves with no disease. Magnitude of the weight differences depended upon relative resistance of the host cultivar to both pathogens. The effect of stripe rust was always diminished by the presence of septoria tritici blotch.

A248

INOCULUM PRODUCTION OF CRONARTIUM QUERCUUM F. SP. FUSIFORME ON OAK. M.C. Klapproth and R.A. Schmidt. School of Forest Resources and Conservation, Univ. of Fl., Gainesville, FL 32611.

When leaves of water oak (Quercus nigra) seedlings were inoculated at seven leaf ages with inoculum densities of ca. 50 to 650 aeciospores/cm^2, the mean number of uredinia and telia/cm^2 increased linearly on three- to 15-day-old leaves. Infection efficiencies for uredinia ranged from <0.003 (ages 3 and 15 days) to 0.014 (age 6 days); infection efficiencies for telia ranged from <0.001 (ages 3 and 21 days) to 0.177 (age 15 days). Mean uredinium area, urediniospores/uredinium, and infectious period decreased significantly as leaf age increased. The latent period for uredinia varied from 5.4 days at 22.5 C to 18.7 days at 10 C; latent period for telia ranged from 7.5 days at 22.5 C to 23.9 days at 10 C. Mean number of uredinia and telia/cm^2 decreased ca. 35 and 85 percent when plants were exposed to 50 and 20 percent of maximum dew (ca. 10 mg/cm^2), respectively, for an 11-hour period.

A249

BIOLOGY OF ALTERNARIA ALTERNATA, THE CAUSAL FUNGUS OF BLACK POD DISEASE OF WHITE BEANS IN SOUTHWESTERN ONTARIO. J. C. Tu, Research Station, Agriculture Canada, Harrow, Ontario NOR 1GO.

Alternaria alternata survived over winter in infected plant debris, buried in the soil or kept in a field screenhouse, and in infested seeds stored in the laboratory at room temperature. During the growing season, A. alternata was isolated from leaves of most species of weeds in the bean field. It was also found in bean plants at all stages of growth. Population densities of A. alternata on the leaves of bean plants grown from disinfested and non-disinfested seeds were similar. Population densities of A. alternata on the leaves of weeds and beans increased as the growing season advanced. Increase in the A. alternata population paralleled with natural senescence of leaves. Sugars and ninhydrin positive substances (NPS) in the leaf wash increased with plant age. At a given growth stage, the concentration of sugars and NPS found in leaf washes of cultivars susceptible to A. alternata were higher than those substances in washes of tolerant cultivars.

A250

SEASONAL DISPERSAL OF Alternaria solani SPORES OVER RUSSET

BURBANK POTATOES IN WISCONSIN. J.W. Pscheidt and W.R. Stevenson, Dept. of Plant Pathology, Univ. of Wis., Madison, WI 53706.

Seasonal dispersal of A. solani was characterized by trapping airborne spores with rotorod spore samplers during the 1980-83 growing seasons. In mid July of 1980 and 1981, a single sampler positioned at canopy height detected a dramatic increase in the concentration of airborne spores. Few, if any spores were detected prior to this time while after this time, concentrations were never as great. During 1982 and 1983, several samplers were operated simultaneously at various heights and locations within field plots to assess whether initial increases in spore concentrations were due to local or outside sources. As before, the initial increase of airborne spores occurred during mid July, however, at much lower concentrations. Concentration gradients obtained at this time indicated that sources of airborne spores were located within and outside of the experimental plots. High concentrations analogus to the initial increase of previous years did not occur until late Aug. and early Sept. The overall seasonal dispersal pattern of spores originating from the experimental plots was proportional to the amount of diseased tissue present.

A251

DISEASE PROGRESS OF POWDERY MILDEW (LEVEILLULA TAURICA) ON TOMATOES IN CALIFORNIA. J. C. Correll and V. J. Elliott, Dept. of Plant Pathology, Univ. of California, Berkeley, CA 94720.

Field plots were established in commercial tomato fields in Stanislaus and Merced Counties of California. Disease, caused by Leveillula taurica (Lev.) Arn. (=Oidiopsis taurica (Lev.) Salm.), was assessed every 7 days after disease onset up until harvest. Disease was recorded as the mean number of lesions per 100 leaflets. Disease onset occurred 45-55 days after transplanting in all fields observed. Plots in Stanislaus Co. (cv. 'Royal Flush') had final mean disease severities of 169.4 and 148.3 lesions per 100 leaflets. Logistic rate of infection for both plots was 0.09. Field plots in Merced Co. (cv. 'Jackpot') had final mean disease ratings of 93.8 and 65.8 lesions per 100 leaflets with logistic rates of infection of 0.13 and 0.17, respectively. Mean maximum and minimum daily temperatures, at canopy height, for Stanislaus and Merced Counties during the sample period were 25.7/12.4 and 31.4/15.2 C, respectively. Although disease severity was relatively low in all plots monitored (2-5%), some fields adjacent to the monitored areas had higher levels of disease at harvest time with the highest level observed being 584.1 lesions per 100 leaflets (15%).

A252

COMPONENTS OF PARTIAL RESISTANCE OF PEANUT TO EARLY LEAFSPOT. M. D. Ricker and M.K.Beute, Dept. Plant Path.,NCSU,Raleigh,NC 27695.

Twenty peanut (Arachis hypogaea) lines representing different levels of resistance to early leafspot were evaluated for five components of rate-reducing resistance: lesion number (LN), latent period (LP), time until leaflet defoliation (DEF), lesion diameter (LD) and sporulation. Leaf cuttings were inoculated with 10,000, 25,000, 55,000, and 100,000 Cercospora arachidicola spores per ml and maintained in moisture chambers for 95 days. Differences among genotypes and inoculum densities were observed for LN, LP, and DEF. Linear regression showed that inoculum concentration explained 71 to 99% of the variation in average LN per genotype. LD and sporulation were measured in a separate test with a suspension of 9000 spores per ml. Differences in sporulation occurred among genotypes when sporulation was measured per lesion (SPL), rather than per unit area of lesion. Differences in LD were not observed, although there was a correlation between SPL and LD. Rankings of genotypes by LN, LP, DEF, and SPL indicated (P=0.05) that genotypes with a greater LN had a shorter LP and defoliated more quickly.

A253

COFFEE RUST: TIMING AND FREQUENCY OF FUNGICID APPLICATION BASED ON NSRMP - PREDICTION - MODEL. Kushalappa, A.C.[1], Hernandez, T.[1], Chaves, G.M.[1], Melles, C.A.[2] & Miranda, J.M.[3]. [1]Dept.º Fitopatologia, UFV, 36570 - Viçosa; [2]EPAMIG, São Sebastião do Paraíso; [3]EPAMIG, Ponte Nova, MG, Brasil.

The net survival ratio for monocyclic process (NSRMP) values were calculated as a product of parameters: inoculum, yield and monocyclic process equivalent for environment (MPEE), determined at two locations during 1983-84 (Fitopatologia Brasileira 9 (in press) - 1984). A protective fungicide, copper oxycloride, was applied when NSRMP $\geq$ 0.00015. The MPEE values were either quantified (complex-model) or preestablished values were used (simple-model). In one location three applications were required based on the complex-model and four when based on the simple, whereas in the other location no application was required. Local recommendations required four applications be made at fixed times.

A254

CULTURE METHOD FOR LARGE SCALE PRODUCTION OF CHARACTERISTIC CONIDIA OF COCHLIOBOLUS CARBONUS AND C. HETEROSTROPHUS. O.H. Calvert, A.S. Foudin, G. Fry and G.F. Krause, Dept. of Plant Pathology, USDA, APHIS, PPQ, and Dept. of Agronomy, Univ. of Missouri, Columbia 65211, respectively.

The purpose of this research was to develop a technique to economically produce gram quantities of Cochliobolus carbonus and C. heterostrophus conidia with consistancy for other research uses that were viable, morphologically normal, and free from other fungi and bacteria. Typical conidia were produced on lima bean agar (LBA) made from 20 g/l finely-ground, dry baby lima bean seed and 7.5 g/l USP crude agar. LBA cultures were inoculated with a loop of conidia, incubated at 25 C under fluorescent light (100-200 lux 24 hr) for 14 d, and then dried at 32-35 C to a thin film. Conidia were aseptically vacuumed at 200 mm of mercury from the dried medium with a cyclone spore collector. We obtained ~25 g of conidia of each species from 300 LBA petri dish cultures (~82 mg conidia/plate culture). Cultures on this LBA medium produced 60 times more conidia than on potato dextrose agar medium and at the lowest practical cost of any of several substrates tried.

A255

PLOT SIZE EFFECTS ON DISEASE PROGRESS AND YIELD OF WHEAT INFECTED BY Mycosphaerella graminicola AND BARLEY INFECTED BY Pyrenophora teres. J. Burleigh and M. Loubane, Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

Areas under disease progress curves (AUDPC) for Pyrenophora teres and Mycosphaerella graminicola and grain weights were subjected to pertinent orthogonal comparisons to determine if AUDPC and crop loss estimates are functions of plot size. AUDPC from plots 40 x 40 m and infected with P. teres were statistically greater than AUDPC from plots 20 x 20 and 10 x 10 m, yet grain weights were similar. Initial severities were similar but final severities at hard dough were 23 to 36% greater in plots 40 x 40 than in plots 20 x 20 and 10 x 10. AUDPC from plots 40 x 40 and infected with M. graminicola also were statistically greater than AUDPC from plots 20 x 20 and 10 x 10 but final severities were only 2 to 10% greater in plots 40 x 40 than in plots 20 x 20 and 10 x 10. Grain weights were not significantly different. AUDPC from plots 20 x 20 and 10 x 10 were not significantly different for P. teres or M. graminicola.

A256

An apparent synergistic effect on statice (Limonium sinuatum) inoculated with a nonpathogenic Colletotrichum gloeosporioides and a pathogenic Pseudomonas caryophylli. Arthur W. Engelhard and J. B. Jones. GCREC-Bradenton, FL 34203.

Colletotrichum gloeosporioides incites a crown rot and Pseudomonas caryophylli a crown and leaf rot of statice. When statice plants were inoculated with C. gloeosporioides from Anthurium by placing a 3 mm^2 piece of agar on which the fungus was growing on a wound on the crown, no disease developed after 41 days. When plants were injected in the crown with P. caryophylli from statice, an average of 13 and 41 percent disease developed after 21 and 41 days, respectively. When the plants were inoculated with both C. gloeosporioides and P. caryophylli by the above methods, 96 and 100 percent disease developed after 21 and 41 days, respectively. Similar results were obtained in 3 replicated tests. Experiments included the cultivars Lavender Queen, Midnight Blue and Zluta. An apparent synergistic effect was demonstrated when statice was inoculated simultaneously with a non-pathogenic isolate of C. gloeosporioides and a pathogenic isolate of P. caryophylli.

A257

EFFECT OF COMBINED OR ALTERNATING USE OF FUNGICIDES ON RESISTANCE AND CONTROL OF FIRE ON TULIPS. G. A Chastagner, Washington State University, Puyallup, WA 98371.

Using fungicides with different mechanisms of action in alternation and/or combinations is advocated to delay or prevent the development of fungicide resistance and subsequent disease control failures. The effectiveness of repeat, alternate, and combination applications of benomyl (0.6 kg a.i./ha) and iprodione (1.1 kg a.i./ha) at 14-day intervals in controlling fire, caused by Botrytis tulipae, on field grown tulips was monitored for 4 years. Applications of benomyl and iprodione provided effective control during the first two seasons, regardless of application method. During the third and fourth seasons, repeat applications of benomyl failed to control disease because of the development of resistance. Repeat applications of iprodione and the alternate and combination applications of benomyl and iprodione continued to provide effective disease control.

A258

ASSOCIATION OF THE GREEN ALGA CEPHALEUROS WITH THE BLACK LEAF-

SPOT OF MAGNOLIA GRANDIFLORA. G. E. Holcomb and M. C. Henk. Dept. of Plant Path. and Crop Physiol., La. Agr. Exp. Sta., La. State Univ. Agr. Ctr., and Botany Dept., La. State Univ., Baton Rouge, LA 70803.

The parasitic alga Cephaleuros was observed sporulating on the surfaces of the common, black leafspot of Magnolia grandiflora. This leafspot (to 2 cm dia.) was reported to be caused by Glomerella cingulata in 1947 (Plant Dis. Rep. 31:298). We observed the production of algal sporangia or gametangia with gametes on all leafspots examined. Vegetative filaments of the alga were observed throughout the leaf tissues. The alga was isolated from these leafspots but contaminating fungi including Colletotrichum commonly invaded the empty gametangia. Specimens of this disease from the National Fungus Collection revealed the leafspots to be the same as those we collected. Algal fruiting structures were seen on specimens from the National Collection. A specimen from the Collection that had been inoculated with Glomerella revealed symptoms completely different from the originals. We suggest this disease is caused by the alga.

A259

FUNGICIDE ACTIVITY AND CULTIVAR REACTION FOR PHOMOPSIS CANKER OF RUSSIAN-OLIVE. H. L. Morton, Univ. Mich., Ann Arbor, MI 48109-1115.

Nine fungicides and the c.v. 'King Red' were studied for the control of phomopsis canker (Phomopsis elaeagni Arnold & Carter) on Russian-olive (Elaeagnus angustifolia L.). In-vitro activity was measured by incorporating fungicides into PDA. At manufacturer's recommended rates (X), Benlate and Dithane M-45 were fungicidal; Baycor, Beam, Elanco 222, Rohm & Haas 5781F, Triforine EC and Zyban were fungistatic; Bayleton was inhibitory. At 0.1 and 0.01X Bayleton, Beam and Triforine were no longer inhibitory. Benlate and Dithane were fungicidal at 0.1, but not at 0.01X. The c.v. and species were inoculated by an infested grain/bark slit technique. In the greenhouse 10 isolates were used. After 1 mo., 32 and 5 of 100 c.v. and species seedlings, respectively, died. In growth chambers, 3 isolates were used to inoculate pairs of matched-caliper trees. After 1 mo., mortality occurred in 24 and 8 of 30 c.v. and species, respectively. No checks became infected.

A260

EFFECTS OF OXYTETRACYCLINE ON GERANIUM FASCIATION. S. H. Kim, W. A. Woodward and S. K. Faul. Pennsylvania Department of Agriculture, 2301 N. Cameron Street, Harrisburg, PA 17110-9408

The chemical control of geranium fasciation was studied because geranium growers occasionally claimed that Corynebacterium fascians was introduced to their greenhouse via culture-indexed plants. Entire rooted cuttings of culture-indexed varieties Penny, Veronica, Spring Time and Yours Truly were dip-inoculated with a C. fascians suspension, and potted in 10 cm pots. Oxytetracycline (Mycoshield™), 200 ppm, was sprayed to run-off on the inoculated plants at weekly intervals during March-August, 1983 in a greenhouse. Due to the decaying nature of the leafy gall symptom and its reoccurrence, the disease was evaluated at 20 day intervals during June-November. The percent of plants with leafy galls on the sprayed plants during June-August and September-November were 0 and 2.4, respectively; whereas, the unsprayed plants exhibited 7.8 and 17.9, respectively. The phytotoxicity of general chlorosis was quickly corrected by discontinuing the treatment and applying additional fertilizer.

A261

BIOLOGICAL AND CHEMICAL CONTROL OF FUSARIUM WILT OF CHRYSANTHEMUM B.C. Raju and C.R. Semer, IV, Technical Division, Yoder Bros., Inc PO Box 68, Alva, FL 33920

Various soil micro-organisms were evaluated for the biocontrol of chrysanthemum wilt caused by F. oxysporum f. sp. chrysanthemi. Tests were conducted under greenhouse and field conditions using fumigated sandy soils. Soil was infested with Fusarium and the trials were conducted with 5 cultivars most sensitive to Fusarium wilt. Of the several Trichoderma isolates tested, T. viride isolate T-1-R9 and T95 provided disease reduction of 50-70%. However, when T. viride was introduced into non-fumigated soils, no disease control was observed. Several isolates of Aspergillus ochraceus and Penicillium restrictum were ineffective against the wilt pathogen and in some cases increased wilt. Pseudomonas putida, isolate E159 was also effective in wilt control and provided reduction in disease occurrence. Initial level of biocontrol agent introduced into the fumigated soils played a major role in disease reduction. Under Florida conditions, no significant difference in disease control was seen between benomyl (Benlate) and thiophanate M (Topsin).

A262

THE ASSOCIATION OF THIELAVIOPSIS BASICOLA WITH DECLINE OF BLUE HOLLY IN KENTUCKY. Paul A. Bachi, University of Kentucky Research and Education Center, Princeton, KY 42445, John R. Hartman, Department of Plant Pathology, and Robert E. McNiel, Department of Horticulture and Landscape Architecture, University of Kentucky, Lexington, KY 40546.

Blue (Ilex x meserveae) Hollies, Ilex aquifolium x I. rugosa, have been widely planted in landscapes in Kentucky. During recent years blue hollies have been observed in various stages of decline ranging from leaf scorch on a few branches to death of entire plants. T. basicola was observed in and isolated consistently from root systems of affected plants. A survey of container grown and landscape blue hollies in Henderson, Louisville and Lexington revealed that ca. 85% of hollies showing decline symptoms had roots infected with T. basicola. Infected cultivars included: 'Blue Prince', 'Blue Princess', 'Mesid', and 'Blue Angel'. Agricultural soils in Kentucky infested with T. basicola may represent a source of local inoculum, however, roots of container-grown blue hollies shipped from some out-of-state sources were found to be already infected by T. basicola.

A263

A FIRST REPORT OF CERCOSPORA BLIGHT ON CRYPTOMERIA JAPONICA IN THE UNITED STATES. R.L. Wick and R.C. Lambe. Department of Plant Pathology, Physiology and Weed Science. VPI&SU, Blacksburg, VA 24061.

Cercospora blight of Cryptomeria japonica, caused by Cercospora sequoiae (= C. cryptomeriae), is a destructive forest nursery disease in Japan. A blighted C. japonica specimen was received by the VPI&SU Plant Clinic and determined by the senior author to be colonized by C. sequoiae. Isolates of the fungus were tested for pathogenicity on Cryptomeria. Lesions on succulent stems and needles were dark brown. Stromata, conidiophores and conidia were produced on the lesions. Conidiophores were fasicled and measured 40-107 µm x 4-6 µm. Conidia were brown, 33-80 µm x 4-6 µm, echinulate and 3-8 septate. On oatmeal agar, cultures were gray to slightly brown and grew slowly (less than 1 cm/2 weeks). Conidia rarely formed on oatmeal agar but formed sparsely on carrot leaf decoction agar. This is the first published report of an occurrence of Cercospora blight on Cryptomeria in the U.S.

A264

TRIFORINE, A NEW FUNGICIDE FOR LEAF SPOT CONTROL ON PHOTINIA FRASERI. G. S. COBB, A. K. HAGAN, C. H. GILLIAM, AND J. M. MULLEN. Auburn University, AL 36849.

Triforine was evaluated as a preventative treatment for leafspot caused by Entomosporium maculatum on P. fraseri. Treatments were applied with a compressed air sprayer to container grown photinia arranged in a randomized complete block. Diseased photinia were elevated above each replicate to maintain inoculum pressure. Triforine applied weekly at 0.4 lb a.i./100 gal maintained excellent disease control. Of the other fungicides screened, only triadimefon was as efficacious as triforine. As the interval between triforine applications was lengthened from 1 to 2 wk, leafspot incidence and severity increased. Lengthening spray intervals beyond 2 wk resulted in poor disease control. Weekly triforine applications at 0.1 lb a.i./100 gal maintained good leafspot control. Disease control improved as rates were increased to 0.4 lb a.i./100 gal to the point that infection of new foliage by E. maculatum was stopped.

A265

EFFECT OF ENVIRONMENTAL FACTORS AND LEAF AGE ON INFECTION OF PHOTINIA FRASERI BY ENTOMOSPORIUM MACULATUM. A.B.A.M. Baudoin, Dept. of Plant Path., Physiol. & Weed Sci., VPI&SU, Blacksburg, VA 24061.

Entomosporium maculatum causes leafspotting and defoliation of Photinia fraseri and several other members of the Rosaceae. Young, expanding leaves of photinia were susceptible to infection, with spots appearing 6-7 days after inoculation. Mature leaves were rarely infected. Under greenhouse conditions, leaves became resistant upon reaching full expansion, within 20 days of their initial appearance. Infection was most rapid at 20 C and was only slightly reduced at 15 and 25 C; little infection occurred at 30 C. At 20 C in the growth chamber or outdoors, a 9-h and a 12-h leaf wetness period was required for light infection and for moderate infection, respectively. Interrupted leaf wetness periods were less effective than continuous wetness for the same length of time. Exposing plants to sunny, outdoor conditions for 1 day between inoculation and the leaf wetness period reduced the number of leafspots by 30-70%.

A266

Preinoculation factors affecting severity of Pseudomonas leaf spot of Schefflera arboricola. A. R. Chase and J. B. Jones. IFAS University of Florida, Agricultural Research Center-Apopka,32703 and Agricultural Research and Education Center-Bradenton, 33508.

Bacterial leaf spot of Schefflera arboricola is caused by Pseudomonas cichorii. In the first study, plants were grown at the following fertilizer levels for 2 mo. prior to inoculation: 2200, 4300, 6500, 8600, 11000, or 13000 kg N/ha/yr (2200 = recommended level). Plants were unaffected by fertilizer level as determined by color, number of leaves, top weight and height. Fertilizer level increases resulted in linear decreases in both lesion number and diameter. Also, there was a linear decrease in lesion diameter and number as leaf age decreased. In a second study, plants were exposed to 100% relative humidity for various times: 0, 2, 4, 8, 12, and 24 hr. Number of lesions increased linearly as duration of exposure increased. In a third study, plants were grown for 6 wk in different light levels prior to inoculation:1000, 2000 and 4000 ft. candles. Fresh weight of tops and plant height were unaffected by treatment. Lesion diameter and number were greatest on plants grown at 2000 ftc.

A267

EVALUATION OF ROSE CULTIVARS FOR REACTION TO BLACK SPOT AND POWDERY MILDEW. J.E. Watkins, J.A. Houfek and D.H. Steinegger, Dept. of Plant Pathology, University of Nebraska, Lincoln, NE 68583; Dept. of Plant Pathology, Kansas State University, Manhattan, KS 66506; and Dept. of Horticulture, University of Nebraska, Lincoln, NE 68583.

Thirty-eight hybrid tea, floribunda or grandiflora rose cultivars were evaluated under field conditions for reaction to black spot (Diplocarpon rosae) and powdery mildew (Sphaerotheca pannosa). Black spot was least severe on the cultivars 'Sunsprite', 'Spanish Sun', 'Crysler Imperial', 'Charlotte Armstrong' and 'Goldilocks'. 'White Knight', 'Miss All American Beauty', 'Mr. Lincoln', 'Double Delight' and 'Cathedral' were low intermediate in their reaction to black spot. Of the remaining 28 cultivars, 'Paradise', 'First Edition', 'Honor', 'Sundowner', 'Eutin', 'Color Magic', 'Oregold', 'Viva', 'Charisma' and 'Friendship' showed severe black spot. The most susceptible cultivars were defoliated by mid-summer. Most cultivars showed good resistance to powdery mildew.

A268

ASH DIEBACK DISEASE DEVELOPMENT IN NEW YORK STATE: 1962-1980. S. B. Silverborg, J. D. Castello and P. D. Manion, State University of New York, College of Environmental Science and Forestry, Department of Environmental and Forest Biology, Syracuse, NY 13210.

Four white ash dieback observation plots were established in 1962 in New York state and monitored for disease development periodically through 1974. In 1968, nine additional plots were established throughout the state and monitored periodically through 1980. In total, 175 trees in 13 plots were checked for disease development between 1962 and 1980. In the first set of plots, mortality increased in a simple interest linear fashion at 8.4% per year. Mortality levelled off at approximately 50% in these plots. In the second set of plots, mortality increased in a simple interest linear fashion at 4.1% per year. In 1980, mortality was approximately 40% in these plots. The incidence of ash dieback, in the past 50 years as reported in the literature, and the linear rate of ash dieback as determined by these plots was not obviously related to spring or yearly drought conditions as assessed by the Palmer Drought Index.

A269

WIND, ROCKS, ROOT DISEASE AND MORTALITY OF SUBALPINE RED SPRUCE AND BALSAM FIR. T. C. Harrington and D. M. Rizzo, Dept. of Botany and Plant Pathology, Univ. of New Hampshire, Durham 03824 and P. J. Marchand, Div. of Environmental Studies, Johnson State College, VT 05656.

Excessive winds on steep, high elevation sites in the northern Appalachian Mtns. may result in windthrow, broken tops, and mechanical damage to shoots by rime ice and to roots by root movements. Root damage appears to be restricted to rocky soils and shallow-rooted trees whose crowns are exposed directly to wind. Severed roots and abrasion wounds were abundant on overstory red spruce (Picea rubens) and fir (Abies balsamea) along the margin of canopy gaps. Fungi (eg, root and butt-rot hymenomycetes) colonized the wounds and further damaged the roots. Mechanical damage to shoots and loss of roots due to root movements and pathogens may help to explain the mortality of spruce on some windy sites with red spruce decline and the mortality of balsam fir in fir waves (J. Ecology 64:899). Acid depositions, drought, or other stresses may exacerbate the damage, but they do not appear to be the primay cause of mortality on these sites.

A270

ASSOCIATION OF WOOD DECAY FUNGI WITH DECLINE AND MORTALITY OF APPLE TREES IN MINNESOTA. D.R. Bergdahl, Dept. of Forestry, Univ. of Vermont, Burlington, VT 05405 and D.W. French, Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

A 10-year study indicated that wood decay fungi were causal agents of apple tree decline and mortality in two orchards in Minnesota (Monticello and Stillwater). Of 140 6-year-old trees surveyed in 1972, 93% were healthy, 6% declining, and 1% dead and extensively decayed. Percentages were 5,23,23 and 2,19,79 for 1976 and 1982, respectively. Symptoms were more advanced in 8-year-old trees in 1972, but there was little difference in symptom development between age classes by 1982. Irpex lacteus was most common on cankered trees in both orchards. Field inoculations of healthy 3-year-old Malus domestica (cultivar Connell Red) confirmed I. lacteus, Coriolus versicolor and Schizophyllum commune caused symptoms and signs associated with apple tree decline and mortality.

A271

DECAY OF BIRCH BY Hirschioporous pargamenus (Fr.) Bond. et Sing. (Polyporus pargamenus Fr.); A COMPARISON OF LABORATORY AND FIELD DECAY. Lewis Otjen. Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Hirschioporous pargamenus caused a white pocket rot in the sapwood of dead paper birch (Betula papyrifera Marsh.) in the forest. Sound birch blocks inoculated with H. pargamenus and incubated for three months at 27°C and 70% relative humidity had patterns of decay macroscopically similar to that which occurred naturally in the forest. Chemical analyses showed lignin to be selectively removed within pocketed areas. H. pargamenus selectively delignified wood in patterns similar to those of other white pocket rot fungi but differed in that delignification was not as complete. Thin areas of tan wood separated pockets of decay, and contained in their centers a thin web of hyphae not seen in other white pocket rots. The formation of pockets in wood and the process of selective lignin removal was shown to result from the action of a single fungus.

A272

DECAY OF ALTERED WOOD RELATED TO DECAY OF SAPWOOD BY A NEW, MODIFIED AGAR-BLOCK TEST. K. T. Smith and W.C. Shortle. University of New Hampshire, Durham, NH 03824.

Incubation of small pieces of wood (1x8x20 mm, grain direction) upon mycelia of a decay fungus permits the rapid comparison of decay resistance of unaltered sapwood to wood altered by aging and injury/infection. Percent loss of oven-dried weight (ODW) for sapwood (SW) ranged from 34-55% at 24 days incubation with the decay fungus Coriolus versicolor; for heartwood (HW) 6-50%. Decay resistance of heartwood expressed as proportional losses in ODW, HW:SW, were black locust 0.2, black cherry 0.4, red oak 0.7, and yellow birch (older SW in place of HW) 1.2. The fungus is grown prior to wood incubation in slants (16x150 mm tubes) containing 12.5 ml malt-yeast extract agar. Advantages of this test include: planar samples make moisture and oxygen more available to the decay system, the small sample size permits comparisons of localized zones, short incubation times and limited space are required, and the materials are commonly available.

A273

RELATION OF WOUND OCCLUSION TO DISCOLORED WOOD COLUMNS IN RED MAPLE. Curt Leben. Department of Plant Pathology, The Ohio State University/OARDC, Wooster, OH 44691.

Stems (15-25 cm dbh) were wounded in spring or fall with horizontal, 1 cm wide chain saw kerfs, 4-6 cm deep and 10-16 cm long. Kerfs were divided by metal to form "treatment" and "control" sides (Can. J. For. Res. 12:115-117. 1982). Data are for control sides, which were left open to the air for two growing seasons. Wound occlusion [VO=vertical occlusion, at the wound side; and bark dieback (BD), the failure of horizontal occlusion above and below the wound] and length of discolored wood column (DWC) were recorded. For 93 trees (one wound each), correlations between cm BD and cm DWC were r=0.58 and 0.56 above and below the wound (n.s.d.), with the DWC being 2-3 times the length of BD. Correlations between VO rating and cm DWC above and below the

wound were 0.41 and 0.38 (n.s.d.). Less BD, greater VO, and shorter DWC resulted from spring wounds (36 trees) than from fall wounds (57 trees). With red maple it is suggested that wood hydrostatic pressure is a major determinant of volume of DWC and hence of wood susceptible to decay.

A274

ARMILLARIA ISOLATES, THEIR CHARACTERIZATION AND THEIR HOST SPECIFICITY. D. Lin, M.T. Dumas and M. Hubbes. Faculty of Forestry, University of Toronto, Toronto, Ontario, Canada M5S 1A1

Armillaria mellea causes root and butt rot in a wide variety of softwoods and hardwoods in Canada and throughout the world. Differences in pathogenicity as well as host specificity show that A. mellea may be composed of several species. Attempts to characterize different isolates from different hosts grown on synthetic and semi-synthetic media by their morphological features failed; however, characterization was possible by isoenzyme pattern of esterase.
When the isolates were tested on stem wood discs of Pinus strobus, the mycelial growth on sapwood was greater than on heartwood. Fresh wood discs were more inhibitory to fungal growth than the sterilized ones.

A275

IDENTITY OF THE ARMILLARIA MELLEA COMPLEX IN ALBERTA. K.I. Mallett[1] and Y. Hiratsuka[2]. [1]Department of Plant Science, University of Alberta, Edmonton, Alberta, Canada T5G 2P5. [2]Canadian Forestry Service, Northern Forest Research Centre, 5320 122 St., Edmonton, Alberta, Canada T6H 3S5.

Two forms of the Armillaria mellea complex, distinguishable by compatability tests, have been found in Alberta. These forms were differentiated using cultural characteristics when grown on potato dextrose agar, carrot agar, malt agar and malt peptone dextrose agar. The characteristics used for separation purposes included thallus growth rate, colony type and rhizomorph branching pattern. One form was consistently isolated from Populus and Betula sp. that were growing on mesic to wet sites and the other from Pinus sp. growing on well drained sites.

A276

ASPEN STUMP COLONIZATION, RHIZOMORPH PRODUCTION, AND SUCKER INFECTION BY ARMILLARIA MELLEA. G. R. Stanosz and R. F. Patton, Dept. Plant Pathology, Univ. Wisconsin-Madison, 53706.

In two Wisconsin stands A. mellea had infected 94% of 100 and 100% of 85 quaking aspen (Populus tremuloides) stumps 2 and 5 years, respectively, after clearcutting. The fungus had colonized >50% of the collar area in 13% of the 2- and 98% of the 5-year-old stumps. The quantities (dry weight) of rhizomorphs from screened soil samples collected around stumps in stands 6-13 years after harvest were 15-37 times the quantity of rhizomorphs from around stumps 3 years after harvest. Of fifty apparently healthy dominant or codominant suckers in each of three stands 3, 9, or 15 years after cutting, 24%, 34%, and 74%, respectively, had root lesions with mycelial fans and decay typical of that caused by A. mellea. The pathogen was isolated from >90% of these lesioned suckers. Thus, A. mellea may be a significant factor affecting the management of aspen sucker stands.

A277

INFECTION OF OUTPLANTED DOUGLAS-FIR SEEDLINGS BY VERTICICLADIELLA WAGENERI. F. W. Cobb, Jr. and D. Adams*, Dept. of Plant Pathology, Univ. of Calif., Berekeley, CA 94720 and *Calif. Dept. of Forestry, Sacramento, CA.

Twelve-meter square plots with about 400 Douglas fir seedlings each were established around ten stumps each of redwood, lightly-infected, heavily-infected and apparently healthy Douglas-fir in an 11-acre clearcut and around ten stumps each of infected and apparently healthy Douglas-firs in a partial cut. Seedlings were planted 4-8 months after the harvest and were checked periodically for foliage symptoms and mortality caused by V. wageneri. Infection was first detected 6 mos after planting. Nineteen mos after planting, average mortality due to infection (by treatment) was: Clearcut, redwood 3.0%, healthy Douglas-fir 3.2%, lightly-infected D-f 21.8%, heavily-infected D-f 13.5%; Partial cut, healthy D-f 5.8%, infected D-f 12.0%. Mortality exceeded 40% on several plots and ranged to 56%. Differences among treatments were significant, except for that between redwood and healthy Douglas-fir on the clearcut. However, differences between harvest methods or stump infection levels were not large enough to be useful in disease management.

A278

ECONOMIC EFFECTIVENESS OF OPERATIONAL THERAPEUTIC PRUNING FOR CONTROL OF DUTCH ELM DISEASE. F. A. Baker and D. W. French, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Therapeutic pruning has not been considered effective as a measure for control of Dutch elm disease (DED) because of low success rates. Trees in this study were identified as candidates for pruning during routine inspections for DED. Pruning was done by homeowners or by contractors. Costs were determined from the 1983 contract for tree removal in one of the cities participating in this study. Two trees were pruned in 1981, 14 in 1982, and 11 in 1983. Of the 27 pruned trees, 2 died in the season of pruning, 4 in the first growing season, and 1 in the third growing season after after pruning. Had trees not been pruned, removal costs would have been $4301. Pruning costs were $900, and removal costs for pruned trees which died were $751. The pruning program has saved these cities $2650 to date. In this operational study, the cost of pruning trees to free them of disease is less than the cost of removing the diseased trees, even when failures are considered.

A279

A MODIFIED APPARATUS FOR TRUNK OR ROOT FLARE INJECTION OF ELMS, Daniel F. Plourde, Francis W. Holmes, Dept. Plant Pathology, Shade Tree Laboratories, Univ. of Massachusetts, Amherst, MA 01003 and William E. Phair, Harvard, MA 01451.

A better method to inject systemic fungicides and other materials into the current-season xylem can improve distribution and diminish wound effects. Many injectors now in use require deep injection holes (up to 5cm) and block the newest wood which is critical for uptake in a ring-porous species. Modifications made to a 4 mm (i.d.) plastic "T" include the use of a spacer between the "T" and the bark surface and driving a 7d duplex nail through the body of the injector in the direction of fluid flow. The injector is held firmly against the tree by the nail while plastic and metal washers prevent leaks. Holes drilled 1.0 cm deep and 1.1 cm wide with a wood bit are larger than the injector tip so that material can flood back to the outermost xylem. When four injectors were used per stem (9.5-17.2 cm dbh) up to 1.5L of 2% acid fuchsin was taken up within a 24 hour period from a gravity feed system.

A280

DISEASE RESISTANCE SCREENING OF SELECTED ELM SPECIES AND CULTIVARS. M. M. Chen, E. B. Smalley and R. P. Guries, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Ten elm species and cultivars were evaluated for resistance to six strains of Ceratocystis ulmi of varying levels of aggressiveness. English elm (U. procera) and American elm (U. americana) were very susceptible to all strains of C. ulmi. Hybrids between "Sapporo Autumn Gold" and English and red elm (U. rubra) European white elm (U. laevis), 'Regal' elm and one collection of Siberian elm (U. pumila) from Xinjiang were moderately resistant to some strains of C. ulmi. Interactions between these elms and C. ulmi strains were very pronounced. "Christine Buisman" elm and a collection of Siberian elm from Liaoning were highly resistant to C. ulmi. In general, the aggressive strains of C. ulmi were more pathogenic to these elms, but differences related to geographic origin of elms and strains of C. ulmi were obvious. In particular, different sources of Siberian elm exhibited different levels of resistance to our strains of C. ulmi.

A281

DISTRIBUTION OF CERATOCYSTIS FAGACEARUM MATING TYPES IN TEXAS. D. N. Appel and C. Frost Drees, Dept. of Plant Pathology and Microbiology, Texas Ag. Exp. St., College St., Texas 77843.

Oak wilt, caused by Ceratocystis fagacearum, has been identified in 31 Texas counties and is the major oak disease in the state. The compatabilities of 100 single spore isolates from 22 counties were tested using standard spermatization techniques. The isolates were obtained from 57 live oaks (Quercus viginiana, Q. fusiformis), 15 Spanish oaks (Q. texana), and 1 blackjack oak (Q. marilandica). Both A and B mating types were found throughout the known range of the disease with a total of 50 type A and 44 type B isolates. Six of the isolates could not be typed. In some counties where both mating types are found, fertile perithecia are frequently observed in fungal mats on Spanish oaks. Ample opportunity exists for sexual recombination by C. fagacearum in Texas.

A282

EFFECT OF HOST RESISTANCE ON THE VIRULENCE OF ERWINIA

AMYLOVORA. J. L. Norelli, H. S. Aldwinckle and S. V. Beer, Dept. of Plant Pathology, New York State Agr. Expt. Stn., Cornell Univ., Geneva, NY 14456 and Ithaca, NY 14853.

Malus pumila cvs. Idared and Delicious, and M. robusta cv. Robusta 5 were inoculated with strains Ea 273 and Ea 266 of E. amylovora. Bacteria were isolated in mass from diseased tissue and used to inoculate another plant of the same cv. After ten serial passages the virulence of the strains was assessed on Idared, Delicious, McIntosh, Quinte and Robusta 5. Passage of Ea 273 and Ea 266 through Idared (very susceptible) resulted in a reduction in virulence. Passage of Ea 273 through Delicious (moderately resistant) had no significant effect on virulence. Passage of Ea 273 through Robusta 5 (very resistant) resulted in a differential increase in virulence to Robusta 5. Although there was no significant change in the strain's virulence to Delicious, Idared or McIntosh (susceptible), there was a significant increase in virulence to Quinte (resistant).

A283

IN VITRO TESTING OF VARIOUS CHEMICALS FOR BACTERICIDAL ACTIVITY AGAINST ERWINIA AMYLOVORA. T. van der Zwet, USDA, ARS, Appalachian Fruit Research Station, Kearneysville, WV 25430.

Epiphytic Erwinia amylovora has been readily isolated from the surface of mature, apparently healthy apple fruit collected in the orchard. Preliminary tests were undertaken to test the effect of various chemicals on the growth of E. amylovora in vitro and to the fruit surface of 'Rome Beauty' and 'Red Delicious' apples. One ml of a cell suspension of E. amylovora at 10^4, 10^6, and 10^8 cells/ml were added to 10 ml of sodium hypochlorite (5, 10 and 20%), acetic acid (0.1, 0.5 and 1.0 M), and ethyl alcohol (50 and 100%). After 1, 3, 5, and 10 min., 0.5 ml of these suspensions were plated on nutrient-yeast-dextrose agar. The plates were incubated at 26° C for 24, 48, and 72 hrs. E. amylovora did not survive any of the concentrations of the chemicals tested, even at only 1 min. exposure. Fruit surfaces remained unaffected after 10 min. dips in the highest concentrations of the chemicals. Two stop scald chemicals, ethoxyquin and diphenylamine, tested up to 4000 ppm, had no effect against the growth of E. amylovora.

A284

DIURNAL CHANGES IN POPULATION SIZES AND ICE NUCLEATION ACTIVITY OF PSEUDOMONAS SYRINGAE ON SNAP BEAN (PHASEOLUS VULGARIS L.) LEAFLETS. S. S. Hirano and C. D. Upper*, Dept. of Plant Pathology and *ARS, USDA, University of Wisconsin, Madison 53706

Population sizes and ice nucleation activity of P. syringae (Ps) on snap bean leaflets from field plots were determined at 2-hr intervals during each of three 26-hr periods. Individual leaflets (100 per sampling time) were assayed for ice nuclei with a tube nucleation test. Population sizes of Ps were determined by dilution plating of homogenates of 30 individual nucleation-tested leaflets per sampling time. The frequency with which leaflets bore ice nuclei active at -2.0, -2.2, and -2.5 C varied greatly within each of the 26-hr periods. These large diurnal changes were inversely correlated with the diurnal changes in air temperature and reflected changes in nucleation frequencies rather than changes in population sizes of Ps. Population sizes of Ps can increase rapidly. In one 26-hr period, the median population increased 28-fold. Populations of Ps increased even during daylight periods in spite of the absence of moisture on the leaf surfaces.

A285

POPULATION TRENDS OF EPIPHYTIC CORYNEBACTERIUM NEBRASKENSE ON CORN/POPCORN GENOTYPES. M. L. Schuster and C. C. Smith, Horticulture Department, University of Nebraska, Lincoln, NE 68583

Comparisons were made of epiphytic populations of Corynebacterium nebraskense Schuster, Hoff, Mandel and Lazar 1972, the incitant of leaf freckles and wilt, on different maize and popcorn germ plasms. There was no consistent relationship between the epiphytic populations and the resistant or susceptible genotypes. Maize and popcorn hybrids and inbreds can maintain high populations of C. nebraskense and thus be involved in epidemiology. The plants can act as colonization sites for the bacterial pathogen.

A286

DETECTION OF X-DISEASE IN PLANT HOSTS BY ENZYME-LINKED IMMUNOSORBENT ASSAY. B. C. Kirkpatrick and D. G. Garrott, Department of Plant Pathology, University of California, Berkeley, CA 94790.

Antisera were produced by injecting rabbits with extracts prepared from Colladonus montanus leafhoppers, infected with peach yellow leaf roll (PYLR) strain of X-disease. These antisera could discriminate between X-diseased and healthy plant hosts of several species when used in an enzyme-linked immuno-sorbent assay (ELISA). Both the cherry-infecting Green Valley strain and the PYLR strain could be readily detected in root and leaf tissue from presymptomatic celery plants. Symptomatic peduncles from field grown sweet cherry trees on 'Mazzard' or 'Mahaleb' rootstocks tested positively. Absorbance values for symptomatic cherry peduncles peaked at fruit maturity. The ELISA procedures used in these experiments did not reliably discriminate between healthy and diseased leaves or roots from peach or cherry trees. Celery infected with Spiroplasma citri or aster yellows did not react positively to X-disease antisera.

A287

CITRUS BACTERIAL CANKER DISEASE IN YEMEN ARAB REPUBLIC. J. E. Dimitman. Cal State Poly Univ., Pomona, CA 91768 & Werner Gassert Yemen-German Plant Protection Project, Box 26, Sanaa Y.A.R.

Citrus bacterial canker disease (CBCD) caused by Xanthomonas campestris pv. citri was first observed on four citrus trees in the Surdud State Farm during the month of Sept. 1982. Within one month, over 400 citrus trees were infected and subsequently the entire block was burned. The trees which were originally found to be infected were part of a consignment of trees from India planted in 1981. Despite eradication and a spray program in the surrounding areas, the disease spread to the Government nursery at Al Garabah some two km to the east. In Feb. 1984, 21,000 young nursery trees, 1000 three-year old Mexican lime seedlings, as well as 350 two-year old orange trees were burned. The disease has been found on Brazilian sour orange and Mexican lime trees but has not been observed on sweet orange, Mandarin or Lisbon lemon. The results of the eradication program indicate the need for adequate quarantine and inspection procedures and haste in treatment and eradication. Efforts are being made to determine which of the three forms of CBCD exists in Y.A.R.

A288

TRANSLOCATION AND MULTIPLICATION OF SPIROPLASMA CITRI IN TURNIP. J. Fletcher and C. E. Eastman. Dept. of Plant Pathology, Oklahoma State University, Stillwater, OK 74078 and Illinois Natural History Survey, Champaign, IL 61820.

Spiroplasma citri, introduced into the third-oldest leaf of turnip plants (Bassica rapa) via leafhoppers (Circulifer tenellus), was first detectable by isolation from roots (day 4) then from the youngest and the inoculated leaves (day 8). All tested tissues except senescing leaves yielded spiroplasmas (day 12), although symptoms of S. citri infection were not seen until day 13. Titer changes were measured in turnip plants inoculated with S. citri via whole-plant exposure to pairs of inoculative C. tenellus. A protocol of enzyme-linked immunosorbent assay (ELISA) was optimized to quantify populations of the pathogen in host plant tissues. Titer of S. citri in young leaves, as determined by ELISA, was first detectable (day 7) at 2 X 10^9 CFU/g. It rose to a peak (day 12-20) of 0.23-5.0 X 10^{11} CFU/g, then declined. In these tests symptoms were visible on day 10.

A289

ANTIBACTERIAL ACTIVITY OF A PROTEIN(S) FROM TOMATO. Tibor Ersek, Arthur L. Karr and Robert N. Goodman, Department of Plant Pathology, University of Missouri-Columbia, Columbia, MO 65211.

Tomato seeds and leaves contain an acid extractable protein capable of agglutinating cells of Pseudomonas syringae pv. tomato, P. s. pv. glycinea and P. fluorescens. In addition, the protein exhibits bactericidal activity against the first two bacteria, but not P. fluorescens, when cells are exposed to protein solution for 30 to 60 min. The optimal bactericidal activity is defined by the ratio of protein to bacterial cells. Both agglutination and killing take place at pH values below 4 while the agglutination activity is diminished at higher pH values. As little as 5 min exposure of the cells to the protein at pH 3.7 is sufficient to kill even if the pH is increased thereafter. Our results suggest that agglutination and antibacterial activities of the protein are separate phenomena. Both may play a role in disease resistance. The relationship of this protein to the group of thionins will be discussed.

A290

LACK OF EVIDENCE FOR IN SITU FLUORESCENT PIGMENT PRODUCTION BY P. S. SYRINGAE ON LEAF SURFACES. S. E. Lindow,

J. E. Loper, and M. N. Schroth. Dept. of Plant Pathology, University of California, Berkeley, CA 94720.

The fluorescent pigment/siderophore of Pseudomonas syringae pv. syringae (Pss) is a chromophore effective in protecting cells from UV exposure in vitro. Pss strain B728a produces the fluorescent pigment and grows on an iron deficient medium (King's B with 200 ug ml^{-1} EDDA) whereas strain I-1, a nonfluorescent derivative of B728a obtained after EMS mutagenesis, does not. Upon in vitro exposure to UV (254 nm), B728a was less sensitive than I-1 (LD_{50} = 142.4 ± 1.4 and 100.8 ± 1.8 erg/mm^2, respectively). Growth rates, stationary populations, number of brown spot lesions, and UV sensitivity of B728a and I-1 did not differ significantly on leaf surfaces of greenhouse-grown bean plants. Survival of these strains was also indistinguishable on leaf surfaces of field grown plants. These indirect studies indicate no evidence of in situ fluorescent siderophore production by Pss nor of its contribution to the growth, survival or pathogenicity of Pss on bean leaves.

A291

FLUORESCENT SIDEROPHORE PRODUCTION AND IRON ACQUISITION BY PSEUDOMONAS SYRINGAE PV. SYRINGAE. Y. S. Cody and D. C. Gross. Dept. of Plant Pathology, WA State Univ., Pullman, WA 99164.

Under low iron cultural conditions Pseudomonas syringae pv. syringae strain B301D produces a water-soluble fluorescent siderophore, pyoverdine$_{pss}$, which may be essential for iron acquisition by the pathogen in the host environment. Pyoverdine$_{pss}$ and ferric pyoverdine$_{pss}$ have absorption maxima of 377 nm and 400 nm, respectively. Both forms of the siderophore are effective in reversing iron starvation of P. syringae. The uptake of iron from ferric pyoverdine$_{pss}$ may require an outer membrane protein functioning as a siderophore receptor. Low iron growth conditions derepress synthesis of outer membrane protein 4 (MW 69,000) as well as pyoverdine$_{pss}$. Mutants of B301D were selected that were deficient in the ability to obtain iron from the ferric siderophore. Mutants were identified by screening for the inability to reverse EDDA-induced iron starvation utilizing purified ferric pyoverdine$_{pss}$. These mutants were altered in their outer membrane protein profile, including a quantitative difference in iron-regulated protein 4.

A292

IMMUNOCHEMICAL CHARACTERIZATION OF SPECIFIC ANTIGENIC DETERMINANT OF MEMBRANE PROTEIN COMPLEX OF XANTHOMONAS CAMPESTRIS PV. CAMPESTRIS. **N. Thaveechai and N. W.Schaad, Dept. Pl., Soil and Ent., Univ. of Idaho, Moscow, ID 83843.**

A LiCl extracted membrane protein complex (MPC) of X. campestris results in a single, specific line of precipitin (SLP) in Ouchterlony double diffusion (ODD). The specific antigenic (SA) determinant(s) was characterized. Crude (c) MPC contained sugar, protein and LPS with a M.W. > 2,000 K. Treatments of cMPC with proteases, periodate, acid, alkali, and heat failed to destroy the SLP but showed additional lines in ODD and immunoelectrophoresis (IEP). SDS-PAGE of cMPC resulted in major peptide bands of 125, 100, 44, 34, 32, and 23.5 K and SDS-PAGE crossed-IEP revealed SA determinant present in the first three bands. The major bands were isolated from SDS-PAGE and presence of the SA determinant in the first three bands confirmed by identity reaction with cMPC in ODD. Bands with SA determinant contained sugar and protein but no LPS. These results suggest that the SA determinant of the SLP is a sugar-protein molecule and occurs in three peptide bands.

A293

IDENTIFICATION OF A PHYTOTOXIN PRODUCED BY PSEUDOMONAS CORRUGATA, CAUSAL AGENT OF TOMATO PITH NECROSIS. J. V. Leary, and W. Chun, Department of Plant Pathology, University of California, Riverside, CA 92521.

Pseudomonas corrugata produces a toxin in culture that causes rapid necrosis when infiltrated into tomato leaves. Culture fluids inhibit growth of Micrococcus luteus and Bacillus sp. The toxin is produced from 5 to 37 C but not at 39 C although vigorous growth occurred. Cultures grown at 39 C, when returned to lower temperatures resumed toxin production. Preliminary purification attempts indicate that the toxin is between 10 and 14 kd. The toxin is soluble in 50% methanol. It is neutral in charge between pH 5 and 9. The phytotoxic activity was not affected by strong acid or base. The toxin is heat labile at 100 C for 2 minutes but stable at room temperature. Studies on the relationship of the toxin in the pathogenesis of tomato pith necrosis is in progress. Further purification and characterization is being continued.

A294

PHYSIOLOGICAL CHARACTERISTICS OF THE FASTIDIOUS, GRAM-NEGATIVE, XYLEM-LIMITED BACTERIA FROM PLANTS. J.M. Wells and B.C. Raju, USDA, ARS, NER, Rutgers Univ., New Brunswick, NJ 08903 and Yoder Bros., Alva, FL 33920.

Physiological characteristics were determined for 13 strains of xylem-limited, fastidious bacteria causing Pierce's disease of grapevines, phony disease of peach, plum leaf scorch, elm leaf scorch, stunt of ragweed and periwinkle wilt. All were Gram-negative and not acid fast, and negative for oxidase, coagulase, β-galactosidase, urease, phosphatase, and for production of H_2S and indole. All were positive for catalase, gelatinase and utilization of hippurate. All strains except phony peach bacterium were positive for β-lactamase. Growth effects of carbohydrate amendments were determined on a complex medium containing peptone and bovine serum albumin. Growth of all strains was stimulated by L-cystine and L-arginine, and inhibited by citrate, choline, esculin, fructose, gluconate, glutamate, α-ketoglutarate, malonate, succinate and rhamnose. Mannose did not affect growth rates.

A295

A VIDEO-MICROPROCESSOR LINKED TECHNIQUE FOR SCORING BACTERIAL GROWTH IN LIQUID MEDIA. C.E. Morris, S.J. Ventura, & D.I. Rouse Department of Plant Pathology, UW, Madison, WI 53706

To assess nutrient use patterns of a large number of epiphytic bacterial isolates, a video camera and microprocessor were used to record growth in liquid minimal media. Single carbon and nitrogen source media in the 96 wells of microtitration plates were inoculated with isolates and growth was recorded after 7 days. The video signal of the plate image was sent to an analog to digital converter housed in a microcomputer. Signal strengths of 9 pixels in each well were stored as digital grey scale values (GSV). The system was calibrated with suspensions of known turbidity and population density of 10 isolates representing a range of growth forms of the epiphytes. Growth can be expressed as the average GSV, the number of doublings relative to controls with no nutrient source, or the presence or absence of growth relative to controls. Pigment production and clumped growth do not interfere with assessment of growth as they would for a spectrophotometric system. Rapid and unbiased assessment of growth make this system superior to conventional replica-plating.

A296

EXTRACELLULAR POLYSACCHARIDE MUTANTS OF CORYNEBACTERIUM MICHIGANENSE pv INSIDIOSUM. M. Paschke, N. K. Van Alfen, Department of Biology, Utah State University, Logan, UT 84322.

Mutants of Corynebacterium michiganense pv insidiosum (CMI) with an impaired extracellular polysaccharide (EPS)-producing ability were obtained by UV irradiation in order to assess the role of EPS in virulence. Liquid culture was used to compare mutant EPS production to that of wild type. Wild type EPS consists of three components. The mutants obtained by UV produced only one of these components or lacked all components of EPS. Bioassay results showed that the mutants were able to grow within susceptible host plants, but vascular discoloration typical of a compatible reaction did not occur. Numbers of bacteria recovered from inoculated alfalfa were the same as those obtained from naturally infected field plants. Wild type CMI infected plants contained approximately 2 x 10^9 cells/gram, while mutant CMI infected plants contained from 7 x 10^7 to 2 x 10^9 cells/gram. Mutant and wild type bacterial populations had no effect on plant growth during this time period.

A297

THRESHOLD LEVEL OF FUNGICIDES NEEDED FOR PROTECTION OF APPLE AGAINST SCAB. Michael Szkolnik, Dept. of Plant Pathology, New York State Agricultural Experiment Station, Geneva, NY 14456.

Studies were made on potted Rome Beauty, Niagara, Macoun and Empire apple trees in the greenhouse to determine the amount of fungicide which would provide protection of foliage against scab (Spilocaea pomi) during one infection period. The trees received a precision application of 10 mg of fresh spray per cm^2 of leaf surface and allowed to dry. They were inoculated with 70,000 conidia/ml and given an infection period of 30 hr in the mist chamber at 18 C. The threshold of fungicides needed for 90-98% protection against scab compared with water-sprayed checks are given here at the following active ug/ml spray rates: dodine, 2; captafol, 5; metiram (Polyram), 10; mancozeb, 20; CGA 64251 (Vangard), 20; thiram, 40; Glyodin, 80; fenarimol, 120; benomyl, 160; captan, 200; and over 300 for bitertanol, triadimefon, and triforine. These threshold levels for protection may be considerably different from those needed for high levels of control in the after-infection (eradicative), presymptom, and postsymptom modes of fungicidal action.

A298

EFFECT OF TWO COMMERCIAL FUNGICIDES ON INCIDENCE OF Diaporthe phaseolorum var. caulivora ON SUSCEPTIBLE SOYBEAN CULTIVARS. R. P. Pacumbaba, V. T. Sapra, and L. K. Prom. Dept. of Natural Resource and Environmental Studies, Alabama A & M University, Normal 35762.

Manzate 200 applied at 3 lbs/acre and Dyrene applied at 6 lbs/acre were tested in the field to control soybean stem canker (Diaporthe phaseolorum var. caulivora) on susceptible soybean cultivars (Bragg and RA 701). Initial results indicated that Manzate 200 and Dyrene, when applied once before inoculation of the stem canker organism, had significantly better control of the disease for both cultivars than when the fungicides were applied after inoculation of the pathogen. Yields of both cultivars were also significantly higher when the fungicides were applied before than after introduction of the organism. The experiment is in progress.

A299

RH-3866, A NEW FUNGICIDE FOR CONTROL OF LOCULOASCOMYCETES, POWDERY MILDEWS AND BASIDIOMYCETES. James A. Quinn and T.T. Fujimoto, Rohm and Haas Co., Research Laboratories, Spring House PA 19477.

RH-3866 is a systemic fungicide useful for controlling diseases caused by several groups of important fungi. It has EC50 values of < 5µg/ml in poison agar tests against many members of the loculoascomycetidae (e.g. Microsphaera spp., Pyrenophora spp., and Cochliobolus spp.) and against Rhizoctonia solani. In greenhouse pot tests it is also highly active against powdery mildews and rusts (EC90 < 10 ppm against Puccinia recondita, P. graminis, Uromyces phaseoli, Erysiphe graminis, Sphaerotheca fuliginea and Erysiphe polygoni when inoculated 1 day before or on the day of treatment). The compound is moderately active against the hymenoascomycetidae including Gibberella spp. Ceratocystis ulmi, and Monilinia fructicola (EC50 in poison agar <10 µg/ml). The compound has excellent curative activity against presymptomatic infections and it has outstanding residual activity. It is probably a sterol synthesis inhibitor.

A300

Efficacy of chlorothalonil in controlling spring black stem (Phoma medicaginis var. medicaginis) and common leaf spot (Pseudopeziza medicaginis) of alfalfa. F.A. Gray and J. L. Horton, Plant Science Division, University of Wyoming, Laramie, 82071.

Severity of spring black stem (SPBS), incited by Phoma medicaginis var. medicaginis, was decreased and forage yield increased in the cultivar Agate with bi-weekly applications of chlorothalonil. Disease severity and upward invasion of P. medicaginis into the plant canopy were both reduced with increasing fungicide rates of 0, .55, 1.12 and 2.25 kg a.i./ha. Severity of SPBS and common leaf spot (CLS) incited by Pseudopeziza medicaginis, as well as leaf defoliation, were decreased and seed yield increased with increasing fungicide rates in the susceptible cultivar Ranger. Severity of SPBS and CLS was less in the resistant cultivar Ramsey. Diseases were easily controlled but there was no increase in seed yield. In vitro studies showed germination of Phoma medicaginis spores was inhibited by a concentration as low as 1 ppm chlorothalonil. Mycelium, although somewhat inhibited at 10 ppm, continued to grow in concentrations up to 10,000 ppm.

A301

EFFECT OF 14 FUNGICIDES ON GERMINATION OF SECONDARY SPORIDIA OF THE KARNAL BUNT PATHOGEN, NEOVOSSIA INDICA. N.L. Cashion, Centro Internacional de Mejoramiento de Maiz y Trigo, Londres 40, Apdo. Postal 6-641, 06600 Mexico, D. F. Mexico

The fungicidal properties of 14 compounds were tested for their effect on germination of secondary sporidia of Neovossia indica using the cellophane bioassay described by Neely and Himelick (Phytopathology 56:203-209). Maneb and mancozeb delayed spore germination at concentrations of 5 and 20 ug a.i./ml and were fungitoxic at 100 ug/ml. At 500 ug a.i./ml, copper hydroxide, Dithane S-31, and triphenyltin hydroxide stopped hyphal growth but not spore germination. Pentachloronitrobenzene, thiram, and captan were fungistatic at 5, 20, and 100 ug a.i./ml. Sporidial germination was not inhibited by bitertanol, chloroneb, copper carbonate, copper sulfate, dichloram, or hexachlorobenzene. Systemic fungicides that fail to inhibit spore germination could not be evaluated with this technique. Compounds showing fungicidal properties in the laboratory tests plus seven systemic fungicides were tested in replicated field trials in the spring of 1984 in Sonora, Mexico.

A302

TOXICITY OF NACCONOL 90F TO DECAY-CAUSING FUNGI OF FRESH MARKET TOMATOES. M. W. Hoy and J. M. Ogawa. Department of Plant Pathology, University of California, Davis, CA 95616.

The anionic surfactant Nacconol 90F (Na dodecylbenzene sulfonate) was fungistatic in vitro to mycelia and spores of Botrytis cinerea, Geotrichum candidum, Phytophthora parasitica and Rhizopus stolonifer. In laboratory tests, B. cinerea decay of injured fresh market tomatoes was significantly reduced by treating the fruit in 200 µg/ml Nacconol solutions for 3 min at 38 C followed by rinsing with fresh water. However, Nacconol amendments of 200 µg/ml did not enhance the ability of 100 or 400 µg/ml chlorine solutions in reducing the incidence of decay caused by B. cinerea. When the fresh water rinse was omitted, Nacconol amendments significantly improved the efficacy of 100 µg/ml, but not 400 µg/ml chlorine treatments. Phytotoxicity was not observed on non-rinsed fruits after treatment with 2000 µg/ml Nacconol alone or in combination with chlorine.

A303

CONTROL OF PRIMARY APPLE SCAB WITH RUBIGAN IN NEW YORK. C. H. Petzoldt, Lilly Research Laboratories, Greenfield, IN 46140

RUBIGAN (fenarimol) formulated as a 1EC material was evaluated under an experimental use permit (EUP) and in small plot research trials in New York during 1982 and 1983 for control of apple scab (Venturia inaequalis). A total of six small plot research trials and 25 large scale EUP trials were conducted. Rates for RUBIGAN ranged from 328 ml/ha (4.5 oz/A) to 877 ml/ha (12 oz/A). Applications were made utilizing commercial airblast sprayers calibrated to deliver 187 l/ha to 935 l/ha. Three different application schedules were evaluated for disease control during primary scab season as follows: 1) RUBIGAN 1EC alone on a 7-day interval; 2) RUBIGAN 1EC alone applied within 96 hours after the onset of an apple scab infection period with intervals no shorter than 7 days (post-infection schedule); and 3) RUBIGAN 1EC tank mixed with captan or mancozeb and applied on a modified post-infection schedule. Rates of captan and mancozeb in the tank were generally half the recommended labelled rates. Control of apple scab was excellent under all three programs.

A304

COMPARISON OF CHLOROTHALONIL APPLIED IN OVERHEAD SPRINKLER IRRIGATION AND CONVENTIONAL BOOM SPRAYER FOR CONTROL OF PEANUT LEAF SPOT DISEASES. R. H. Littrell, W. A. Rohde, and G. W. Harrison, Plant Pathology Dept.; USDA-ARS, Coastal Plain Station, Tifton, GA; and SDS Biotech Corp. Painesville, OH.

A center pivot irrigation system (CPIS) and conventional boom sprayer (CBS) applied 1.24 kg ai/ha chlorothalonil in 37,400 and 94 liters of water/ha, respectively, every 14 days beginning 35 days after planting and continuing until two weeks prior to digging. Severity of Cercosporidium personatum was determined 113 and 135 days after planting. Foliage from top, middle and lower canopies was analyzed for chlorothalonil residue immediately before and after treatment. The CBS treated plants had less percent defoliation than plants treated with the CPIS, however, no differences in percent necrotic tissue were detected between the two methods. More residue was found on foliage treated with the CBS and the most was found in the top canopy. In contrast, there were no differences in canopy levels when plants were treated with the CPIS. Pod yields tended to be greater than peanut treated with CBS.

A305

INFLUENCE OF LEVEL IN CANOPY AND POST-APPLICATION TIME ON THE EFFICACY OF TWO PROTECTANT FUNGICIDE TREATMENTS FOR TOMATO EARLY BLIGHT. D. J. O'Leary and P. B. Shoemaker, Department of Plant Pathology, North Carolina State University, Raleigh, NC 27695.

Tomato plants (cv. Flora-Dade) in the field at Fletcher, NC were sprayed with chlorothalonil (340 g ai/378.5 L), and mancozeb (363 g ai/378.5 L) + anilazine (227 g ai/378.5 L) using a backpack mistblower. Fungicide persistence was assessed approx. daily for 9 to 15 days after fungicide application by inoculating foliage in the upper and lower thirds of the canopy with an Alternaria solani conidium suspension (2,000 or 10,000/ml). A leaf infection index was calculated: II = (lesions on treated leaf/lesions on untreated leaf) X 100. Lower canopy II was less than upper canopy II, but significant ($P=0.05$) in only 3 of 6 experiments; e.g., in testing persistence of chlorothalonil for 9 days, upper canopy II rate was 3.9 while lower canopy rate was 0.2 II units/day. Chlorothalonil generally persisted longer than mancozeb + anilazine. Mean II of chlorothalonil (23%) was less than ($P=0.05$) mancozeb + anilazine II (38%), in one expt.

A306

CONTROL OF BLACKLEG DISEASE OF BROCCOLI THROUGH CULTURAL PRACTICES AND THE APPLICATION OF FUNGICIDES. A. S. Greathead. Monterey County Agricultural Extension, 118 Wilgart Way, Salinas, CA 93901.

Blackleg disease incited by Phoma lingam has caused increased losses in direct seeded broccoli in the Salinas Valley of California in recent years. Field trials were conducted in 1983 to test varieties for disease resistance, the effects of fungicides and the effect of plowing under crop refuse. Iprodione applied to the base of 3" high plants resulted in the highest level of control. The application of dichloran, benomyl, chlorothalonil, maneb + Zn, captan and vinclozolin resulted in intermediate control. Plowing under plant refuse from a seriously diseased crop with a specially designed plow resulted in a 67% reduction in disease in the succeeding broccoli crop when compared with unplowed areas. All commercial varieties tested showed a high level of disease incidence. Several experimental lines showed substantially lower levels of disease.

A307

EFFECTS OF SIZE OF LESIONS CAUSED BY ALTERNARIA PORRI ON THE EFFICACY OF TWO FUNGICIDES TO CONTROL PURPLE BLOTCH OF ONIONS. Marvin E. Miller, Texas Agricultural Experiment Station, Weslaco, Texas 78596.

Onions treated with mancozeb (Manzate 200), chlorothalonil (Bravo 500) and untreated controls required significantly different ($p = 0.05$) number of days until leaf damage levels caused by Alternaria porri reached 20%. However, days necessary for leaf damage levels to increase from 20 to 80% were not significantly different between treatments. In growth chambers, significant differences in damage levels were observed 4 days after inoculation between leaves with 1-3 lesions at leaf bases and leaves with more than 4 lesions; however, after 6 days there were no significant differences between treatments. The large purple blotch lesions girdled the leaves after 6 days and killed the tissue distal to the lesion. One to three lesions on leaves cause approximately the same damage level as 4 or more lesions, thus masking the effects fungicides have on controlling purple blotch.

A308

INFLUENCE OF SYSTEMIC CHEMICAL PRODUCTS ON THE INFECTION CAUSED BY TOBACCO MOSAIC VIRUS. Magda Carvajal. Dep. de Botánica, Inst. de Biología, UNAM. Ap. Postal 70-233, Delegación Coyoacán. 04510 México, D.F.

The aim of this research was to control the Tobacco Mosaic Virus (TMV) with two systemic fungicides[Vitavax(Vx), and Plantvax(Px)] and a plant hormone [Gibberellic acid (GA)], used as inhibitors of the local lesions caused by TMV in Nicotiana glutinosa.The best inhibition of TMV was obtained with Vx (1/10)on lower leaves; Vx+ Px + GA (Original dose)on lower leaves;Px (1/100) at soil; Vx + Px (1/1000) at soil and Vx + Px + GA (1/1000) on lower leaves.The GA alone was not active, but probably helps the penetration of the Vx through the roots and avoids the dwarfism caused by the Px. All the treatments were repeated in tomato plants and here the most effective were Px (1/100) at soil and Vx + GA (1/100)at soil also, but they caused plant toxicity.

A309

PSEUDOMONAS CORRUGATA-INDUCED PITH NECROSIS OCCURRENCE ON FIELD-GROWN TOMATOES IN LOUISIANA. W.P. Bond, and L.L. Black. Dept. Bio. Sciences, Southeastern La. Univ., Hammond, 70402; and Dept. Plant Path. & Crop Physiol., La. Agri. Expt. Sta., La. State Univ. Agri. Ctr., Baton Rouge, 70803, respectively.

A severe pith necrosis was observed on field-grown 'Floradel' tomatoes in Louisiana during the summer of 1983. Stems of infected plants appeared completely hollow due to breakdown and collapse of the pith. Other symptoms associated with the disease were elliptical dark brown to black surface lesions and profuse adventitious root formation. A nonfluorescent pseudomonad, similar in cultural and biochemical characteristics to Pseudomonas corrugata, was consistently isolated from diseased tissue. All isolates produced pith necrosis when inoculated into 4 wk old 'Bonny Best' tomato plants. When biochemical and physiological traits where compared, the Louisiana isolates appeared similar, if not identical, to known isolates of P. corrugata from Pennsylvania and Florida. Symptomotology, cultural and biochemical characteristics, pathogenicity and identity with known isolates, indicate that the tomato pith necrosis disease in Louisiana is incited by P. corrugata.

A310

CONTROL OF TOMATO POWDERY MILDEW. Demetrios G. Kontaxis, University of California Cooperative Extension, 1700 Oak Park Blvd., Pleasant Hill, CA. 94523.

Powdery mildew of tomato, Leveillula taurica, is erratic but widespread in California. One application of triadimephon or propiconazol at 0.350 and 0.336 Kg. a.i./ha/935 liters water, respectively, or one dusting with sulfur dust 98% at 44.8 Kg./ha/suppressed the fungus for at least 26 days. The triadimephon and sulfur-treated plant population was free of Powdery mildew 26 days after treatment. In the Benomyl-treated and the non-treated plots 4.5% and 11.6% of the plants were infected, respectively. The data were statistically significant. In another experiment, where the disease was severe, triadimephon considerably suppressed, but did not eliminate infection or fungal sporulation 8 days after airspray. Most conidiophores and conidia shriveled and collapsed and signs of powdery mildew developed on young sprayed leaves.

A311

OCCURRENCE AND DISTRIBITION OF POTATO VIRUSES M, S, AND X IN THE POTATO CULTIVAR ATLANTIC. R. W. Goth and R. E. Webb, Vegetable Lab., USDA, ARS, Beltsville, MD 20705.

The enzyme-linked immunosorbant assay (ELISA) and the infectivity assay using the PVX indicator plant Gomphrena globosa were used to assay individual tubers and plants from 10 samples of the potato cv Alantic collected in Maine, Colorado, and Nebraska for the presence of potato virus M (PVM), potato virus S (PVS) and potato virus X (PVX). Potato virus M was detected only in samples from Maine. In these samples the incidence of PVM ranged from 20 to 90 percent. All plants in all samples were infected with PVS. We did not detect PVX in any of the samples. We conclude that the cultivar Atlantic remains resistant to common strains of PVX.

A312

NATURE OF CULTURAL VARIABILITY IN FUSARIUM OXYSPORUM F. SP. APII. R. T. Awuah and J. W. Lorbeer. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Fusarium oxysporum f. sp. apii causing Fusarium yellows of celery was strongly mycelial when isolated from diseased plants and cultured on chloramphenicol-amended potato-dextrose agar (CPDA). Two-week-old mycelial isolates cultured on CPDA at 24-25 C (12 hr photoperiod; Sylvania fluorescent cool white light; 3,875 lux) produced abundant microconidia but few macroconidia. When single-microconidia were transferred from mycelial cultures and grown on CPDA, pionnotal cultures were produced. Mycelial cultures were more virulent and produced more chlamydospores than pionnotal cultures but were less stable than pionnotal cultures and produced fewer macroconidia. Radial growth and conidium dimensions were similar for both culture types. Mycelial and pionnotal cultures maintained their morphological identity when passed through the host and re-isolated on CPDA. Monoxenic storage of cultures in soil columns greatly reduced cultural variability.

A313

OCCURRENCE OF FUSARIUM YELLOWS OF CELERY IN NEW YORK AND ATTEMPTS TO CONTROL THE DISEASE. R. T. Awuah[1], J. W. Lorbeer[1], and L. A. Ellerbrock[2]. [1]Dept. of Plant Pathology and [2]Dept. of Veg. Crops, Cornell University, Ithaca, NY 14853.

A devastating disease of celery (Apium graveolens var. dulce) in Orange County, New York in recent years was proven to be Fusarium yellows caused by Fusarium oxysporum f. sp. apii Race 2. The pathogen was isolated consistently from roots, crowns, and petioles of diseased plants from several commercial fields in 1980. Pathogenicity of isolates was demonstrated in greenhouse infection studies by production of typical field symptoms: root decay, general chlorosis and stunting of plants, leaf curling and stiffening, internal discoloration, and death of plants. In 1980 greenhouse and field tests, all commercially grown celery cultivars tested were diseased, but several celeriac (A. graveolens var. rapaceum) plant introductions were resistant. Fungicidal drenches were ineffective for control. In 1983 field trials, the cultivars Bishop and Deacon were tolerant but not horticulturally acceptable.

A314

DEVELOPMENT OF FUSARIUM BASAL ROT AND MANAGEMENT OF PINK ROOT OF ONIONS. K.L. Everts and H.F. Schwartz, Dept. of Botany and Plant

Pathology, Colorado State Univ., Fort Collins, Colorado 80523.

Studies were made of soil-borne onion pathogens and factors affecting their development and management throughout major production regions of Colorado. Fusarium basal rot (Fusarium oxysporum f.sp. cepae)and Pink Root (Pyrenochaeta terrestris) can seriously reduce onion productivity, especially in the presence of maggots (Hylemya species) or if mechanical wounding occurs. Fusarium basal rot incidence was significantly increased from 14% without maggots to 20% with maggots, and to 42% when wounding occurred in infested soil. Soil solarization, seed coat or granular applications of a biocontrol agent (Pseudomonas putida), or a seedbed drench with benomyl did not reduce disease incidence in a field naturally infested with both pathogens. However, a seedbed drench of benomyl and metalaxyl increased the harvest index by 20% in a field heavily infested with P. terrestris. Additional studies are underway to substantiate these fungicide results, and to determine their effects on Pink Root infestation and disease development.

A315

DRY ROT OF GARLIC CAUSED BY BOTRYTIS PORRI BUCHW. P. A. Somerville, D. H. Hall and A. S. Greathead. Department of Plant Pathology, University of California, Davis, CA 95616.

Dry rot of garlic, caused by Botrytis porri, was observed in California during 1982 and 1983 and on garlic grown in Nevada and Oregon in 1983. This is believed to be the first report of the presence of this pathogen in North America. Infected plants were stunted and their outer leaves turned yellow, wilted and died prematurely. Conidial masses were often observed on the neck and bulb. Bulbs were shrivelled and light weight. Large, black, cerebriform sclerotia that formed on the outer scales were the principle diagnostic feature. In culture the mycelium is white-gray and forms a thick mat on rich media. Sporulation and formation of appressoria and sclerotia occurred primarily at the periphery of the culture dishes. Apothecia developed from sclerotia in the presence of moisture and light at 13°C, but have not been observed in the field. Infection of garlic cloves and onion slices occurred only after injury.

A316

FIELD REACTION OF PHASEOLUS VULGARIS TO SCLEROTINIA SCLEROTIORUM IN WISCONSIN. **D. J. Hagedorn, R. E. Rand, and W. R. Stevenson, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.**

Reaction of bean Plant Introductions (PI's), breeding lines and cultivars (cvs) to Sclerotinia sclerotiorum was studied in an infested field in 1982 and 1983. In '82 we planted 55 PIs and breeding lines in single 3.6 m rows 45 cm apart. Thirty-five of these lines and 15 cvs were tested in '83. Overhead irrigations aided disease severity. Data were obtained at late pod stage. A disease severity index (DI) was calculated, i.e. 0=all plants healthy, 100=all plants 100% molded. Eight bean lines had a DI <15 for both years, i.e. PIs 204717, 271999, 282078, Florida 72, Tacarigua, BTS 16, 821860 and 821857. PIs 204717 and 282078 gave DI readings <5 for both 1982 and 1983. Disease indices for most cvs were not significantly different, but were significant for 4 cvs including the most susceptible Peak (DI 44) and the most resistant Early Bird (DI 9).

A317

MULTIPLE REGRESSION ANALYSIS TO DETERMINE BEAN YIELDS IN WISCONSIN'S CENTRAL SANDS. **K. M. Kobriger and D. J. Hagedorn, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.**

The root rot complex of snap beans (Phaseolus vulgaris) is a serious yield constraint in Wisconsin's Central Sands production area. Data from 22 bean fields on root rot potential, planting date and crop history were used to determine for the first time the relationship of these variables to yield via multiple regression analysis. The equation was $Y = b_0 - b_1X_1 - b_2X_2 - b_3X_3$ where Y is yield, X_1 is root rot potential, X_2 is cropping history, and X_3 is planting date. High yearly correlations were found: '79 R^2=99.2, '80 R^2=70.3, '81 R^2=82.2 (df=0, 6 & 6). Pooled data correlated highly: R^2=56.6 (df=19). Correlation coefficient analysis showed positive correlation between variables within given years but not for pooled data. Results showed that early planting, many previous bean crops, and high root rot potential are important variables for predicting yield losses.

A318

STABILIZING RESISTANCE IN CUCUMIS MELO AGAINST DOWNY AND POWDERY MILDEWS IN ISRAEL AND THE USA. Y. Cohen[1], H. Eyal[1], and C. E. Thomas[2]. [1]Dept. Life Sciences, Bar-Ilan University, Ramat-Gan, Israel; [2]USDA, ARS, Vegetable Laboratory, Charleston, SC 29407.

High levels of resistance against downy (Pseudoperonospora cubensis) and powdery (Sphaerotheca fuliginea) mildews were found in Cucumis melo (cantaloup) PI 124111. Downy mildew resistance expressed as small (1-2 mm dia), circular, yellow, water-soaked lesions with extremely limited sporulation was effective against two races of the pathogen in the U. S. A. and the race in Israel. Powdery mildew resistance expressed as absence of lesions on true leaves, stems, and petioles was effective against races 1, 2, and 3 in the U. S. A. and races 1 and 2 in Israel. Six generations of selection and selfing within PI 124111 were required to stabilize resistance against both diseases. No linkage of the two resistances was observed since plants with resistance to either or to both diseases could be selected. The stabilized inbred of this PI is currently used in cooperative breeding programs. Research supported by BARD.

A319

CONTROL OF PHYTOPHTHORA ROT WITH METALAXYL IN ESTABLISHED ASPARAGUS. Peter G. Falloon*, L. M. Falloon+, R. J. Mullen†, B. L. Benson+, and R. G. Grogan*. *Departments of Plant Pathology and +Vegetable Crops, University of California, Davis, CA 95616, and †Cooperative Extension, San Joaquin County, Stockton, CA 95205.

Field plots of asparagus (Asparagus officinalis L.) established in Yolo loam at Davis in 1977 and inoculated with field soil containing Phytophthora megasperma var. sojae were sprayed with metalaxyl (Ridomil 2E) at rates of 0.56, 1.12 or 2.24 kg a.i./ha either on January 10 or February 20, 1983 or at 1.12 kg a.i./ha at both dates. Treatment with metalaxyl resulted in yield increases of between 35% and 141%. The results of this trial and a second similar trial on peat soil in the San Joaquin Delta showed that one application of metalaxyl at 1.12 kg a.i./ha applied 10 to 20 days before start of harvest resulted in the most cost-effective control of Phytophthora rot of asparagus. Data for 1984 also will be reported.

A320

A SURVEY OF CALIFORNIA ASPARAGUS FOR ASPARAGUS VIRUS I (AV I), ASPARAGUS VIRUS II (AV II) AND TOBACCO STREAK VIRUS (TSV). Peter G. Falloon, L. M. Falloon, and R. G. Grogan. Department of Plant Pathology, University of California, Davis, CA 95616.

Spear samples were collected from 41 asparagus (Asparagus officinalis L.) fields in California and from 100 male and 100 female plants in the U.C. 157 Foundation Seed block at Davis. Fern samples were collected from parent plants of the asparagus hybrids U.C. 157 and Ida-Lea. Sap from spears giving positive reactions following mechanical transmission to Chenopodium quinoa was tested using antisera of AV I, AV II and TSV in Ouchterlony double diffusion plates. AV II was the only virus found in 11 fields throughout California and in two female plants in the U.C. 157 Foundation Seed block. AV II was seed transmitted in U.C. 157 and Mary Washington. Both male and female parents of U.C. 157 and Ida-Lea were infected with AV II but tissue-cultured plants derived from them were free of the virus. A stock of parent plants of Ida-Lea free of AV I, AV II and TSV has been established. This is the first report of a virus occurring in asparagus in California.

A321

INFLUENCE OF CUTTING PRESSURE AND CAPTAFOL ON ASPARAGUS DECLINE. S.A. Johnston. Rutgers University, Rutgers Research & Development Center, Bridgeton, N.J. 08302.

The influence of cutting pressure and an annual soil application of captafol on the incidence of asparagus decline was investigated by measuring asparagus yield in a field of asparagus produced in a field with a history of asparagus production. One-year-old asparagus crowns were planted in 1979. Each year captafol was applied to selected plots as a soil application prior to spear emergence at the rate of 9 kg/ha. Three cutting pressure (cp) regimes were evaluated by varying the weeks of harvest during the 1980, 1981, 1982 and 1983 growing seasons (normal cp: 0, 2, 4, 8; moderate cp: 2, 4, 6, 8; heavy cp: 4, 6, 8, 8). In 1983 all treatments were harvested for a period of 8 weeks. Significantly greater yields resulted in normal cp regimes than in moderate or heavy cp regimes. Yields were greater in captafol treated plots than in nontreated plots. The greatest yield was obtained with the use of captafol and a normal cp regime. Asparagus decline is characterized by reduced yields.

A322

PARSNIP PETIOLE CANKER CAUSED BY PHOMA COMPLANATA.

R.F. Cerkauskas, Agriculture Canada, Vineland Research Station, Vineland Station, Ontario LOR 2EO

Parsnip petiole canker caused by Phoma complanata was found for the first time in Canada in the Bradford Marsh in Ontario in 1983. The disease was observed on plants growing on muck and mineral soils with incidence ranging from trace to 80% of plants affected. Black petiole lesions with immersed pycnidia bearing spore tendrils accompanied drooping and chlorosis of leaves followed by foliar necrosis. Dark tan, irregular leaf spots with chlorotic margins were also common. Symptoms in artificially inoculated plants were identical to those observed in natural infections. The fungus was readily reisolated on potato dextrose or oatmeal agar from inoculated but not control plants. The hyaline, single-celled conidia averaged 7.1 µ X 2.5 µ in length and width respectively. Pycnidia in culture were dark, unilocular, separate or aggregated with a single ostiole.

A323

EFFECTS OF FIELD LOCATION AND ROTATIONAL HISTORY ON PRATYLENCHUS PENETRANS POPULATIONS ON POTATO. D. Florini and R. Loria, Dept. of Plant Pathology, Cornell Univ., Long Island Horticultural Research Lab., Riverhead, NY 11901

Pratylenchus penetrans (Pp) populations were assessed in mid-summer and in late summer in paired, rotated and non-rotated potato fields on the north and south forks of Long Island. Rotated fields previously had been planted to rye or wheat. Five samples each were taken from a total of 8 fields in 1982 and 12 fields in 1983. Nematodes in soil and potato roots were extracted with Baermann pan and shaker techniques, respectively. Differences in Pp populations associated with field location and rotational history were usually significant ($P \leq .10$). Populations were consistently larger on the south fork of Long Island than on the north fork. Populations in rotated fields were generally greater than those in non-rotated fields. Differences in edaphic factors among fields are discussed in relation to Pp populations.

A324

EFFECT OF RESISTANT CULTIVARS AND A FUMIGANT NEMATICIDE ON THE CONTROL OF THE TOBACCO CYST NEMATODE (GLOBODERA TABACUM SOLANACEARUM) IN FLUE-CURED TOBACCO. D. A. Komm and T. R. Terrill, So. Piedmont Center, VPI&SU, Blackstone, VA 23824.

Several flue-cured tobacco cultivars and Soilbrom 90 (32ℓ/ha) were evaluated at two locations for two years for control of the tobacco cyst nematode (TCN) (Globodera tabacum solanacearum). Initial nematode levels were considered low and high for locations 1 and 2, respectively. Control of TCN was determined by measuring plant and root vigor, yield, plant height, stalk diameter, and nematode densities. Yields of cultivars treated with Soilbrom 90 (32 ℓ/ha) were higher than untreated cultivars except for location 1 in the first year. Soilbrom increased plant height, plant vigor, and root vigor over the untreated cultivars. 'Clemson PD-4', 'Va 81', and 'NC 567' significantly decreased 2nd stage larvae, cyst, and egg densities in fumigated and nonfumigated soils at both locations.

A325

EFFECTS OF ALDICARB AND TELONE II ON COTTON PRODUCTION. Earl B. Minton and Jack C. Bailey, USDA, ARS, P. O. Box 225, Stoneville, MS 38776.

Root-knot nematodes and early season insects, primarily thrips and plant bugs, can adversely affect cotton production. Some nematicides used today control both nematodes and insects, but their effectiveness in controlling these pests has not been well defined. The effects of aldicarb 15G (0.84 kg a.i./ha), a nematicide-insecticide, applied in the seed furrow at planting, and Telone II (28 l/ha), a nematicide only, injected in the seed bed 3 weeks before planting, were determined on five cotton cultivars. Populations of root-knot nematodes in untreated soil were at a medium level. Aldicarb and Telone II similarly reduced both the number of root-knot larvae in the soil and the severity of galling of the cotton roots. Low populations of thrips and plant bugs were recorded. The average lint yield of all cotton cultivars was 6% higher for aldicarb and 4% higher for Telone II than for the control. Cultivars reacted differently with these pesticides as aldicarb increased the yield with some and Telone II with the others.

A326

CONVENTIONAL AND CHEMIGATION METHODS FOR CONTROL OF TOBACCO BLACK SHANK AND ROOT KNOT NEMATODES WITH METALAXYL AND FENAMIPHOS. A. S. Csinos and A. W. Johnson, Associate Professor of Plant Pathology and Supervisory Research Nematologist, USDA, ARS, Coastal Plain Station, Tifton, GA 31793.

Two chemical application techniques were evaluated for the control of tobacco black-shank (TBS) and root-knot nematodes (RKN). A randomized complete block split plot design consisting of two tobacco cultivars (NC 95, susceptible to TBS but resistant to RKN and Coker 48, resistant to TBS but susceptible to RKN) of two rows each, 10 m long replicated four times was used. Application techniques were: conventional, a tractor powered rototiller with a spray boom; and an overhead irrigation simulator that applied chemicals in 0.25 cm of water. Metalaxyl (2.24 kg/ha) and fenamiphos (6.72 kg/ha) were applied alone and in combination using both application techniques. No significant differences in control were found between the two methods for either nematode or black shank. Application of the pesticides through irrigation water was as efficacious as the conventional method of application, but is more economical.

A327

CROP SEQUENCE AND SOYBEAN REACTION TO ROOT-KNOT NEMATODE. W. Birchfield, B. G. Harville and M. Lear. USDA, ARS, Dept. of Plant Pathology and Crop Physiology and Agronomy Dept., La. Agric. Exp. Sta., La. State Univ. Agr. Ctr., Baton Rouge, LA 70803.

Soybean resistance to root-knot nematode, Meloidogyne incognita Wartellei, is unknown in relation to growing soybeans in sequence with corn and grain sorghum. Thirteen soybean varieties/accessions were field tested for resistance to this nematode when soybeans were planted after corn, grain sorghum, and soybeans. Soybeans planted after soybeans developed the most root galls and soybeans following grain sorghum the least increase in root-knot larvae in the soil. Differences in soybean yields following the 3-crop sequence were not statistically significant. Yields were negatively correlated with foliage disease symptoms expressed as an index. Foliage necrosis and discoloration for assaying soybean varietal resistance to root-knot nematode may be more reliable than root galling/root-knot larval populations generally used.

A328

A NON DAMAGING ASSOCIATION OF ROOT-KNOT NEMATODE WITH PEACH. P. F. Bertrand and D. R. Evert. Departments of Extension Plant Pathology and Horticulture, The University of Georgia, Tifton, GA 31793.

Peach trees are commonly stunted when planted into high populations of root-knot nematode. In 1980 a 4 year old orchard (c.v. Junegold) showing no sign of stunted trees was found to contain some trees with heavy root-knot galling in the feeder roots. Infected and healthy trees were compared to see what effects could be attributed to root-knot nematode. Very little effect of root-knot was found in this orchard. Root-knot infection did not increase water stress in either a dry year (1981) or a wet year (1982). In 1981 there was no effect on yield. There was a reduction in fruit diameter ($p = 0.05$) but not packing size. There was no effect on size in 1982. The effects on leaf nutrient content were erratic and only occasionally significant ($p = 0.05$). Nutrients never approached deficient or toxic levels due to root-knot infection.

A329

EFFECTS OF GLYCEOLLIN ON HATCHING AND MOTILITY OF HETERODERA GLYCINES. P. C. TRIVEDI, K. R. BARKER, AND J. S. HUANG. Department of Plant Pathology, North Carolina State University, Raleigh, NC 27695-7616.

Phytoalexins are associated with the incompatible response of certain plants to fungi and bacteria. Their involvement in resistance to plant pathogenic nematodes is less certain. In vitro experiments were conducted to study the effects of the phytoalexin glyceollin on hatching and motility of the soybean cyst nematode, Heterodera glycines race 1. Glyceollin at 100 and 200 ug/ml in 1% ethanol adversely affected egg hatching and juvenile motility. These effects, however, could be reversed by rinsing eggs and juveniles in 1% ethanol. These results indicate that the phytoalexin glyceollin has a deleterious effect on H. glycines, and the effect is nematistatic rather than nematicidal.

A330

A COMPARISON OF FUNGI ASSOCIATED WITH HETERODERA GLYCINES CYSTS IN TWO ILLINOIS SOYBEAN FIELDS DURING 1983. L. M. Carris, D. A. Glawe, and D. I. Edwards. Dept. of Plant Pathology, and USDA-ARS, Univ. of Illinois, Urbana, IL 61801.

Populations of the soybean cyst nematode (SCN) and associated fungi in 2 Illinois fields were compared by sampling at 3-wk intervals, May-November, 1983. One field, near Dix, had typical high SCN population levels; the other field, near Sidney, had low SCN populations despite years of infestation. Cyst populations in the Dix field increased 9-fold during the growing season, while the population in the Sidney field remained stable. Over 50 different fungi were isolated from cysts from the 2 fields, the most common being species of Cylindrocarpon, Exophiala, Fusarium, Gliocladium, Phoma, Stagonospora, and Verticillium. Known SCN parasites, including Fusarium oxysporum, Cylindrocarpon destructans, Phoma spp., and Verticillium chlamydosporium were isolated more frequently from the low SCN population field, suggesting a possible involvement in suppressing SCN population levels.

A331

SPECIES OF VERTICILLIUM AND PAECILOMYCES AS PARASITES OF CYST AND ROOT-KNOT NEMATODES. Gareth Morgan-Jones and R. Rodriguez-Kabana, Department of Botany, Plant Pathology and Microbiology, Alabama Agricultural Experiment Station, Auburn University, Alabama 36849.

Four species of Verticillium, V. chlamydosporium, V. lamellicola, V. lecanii and V. leptobactrum, all belonging to section Prostrata of the genus, together with two species of Paecilomyces, P. lilacinus and P. variotii, have been found as parasites of cysts and eggs of Heterodera glycines, Meloidogyne arenaria and M. incognita in Alabama. Greenhouse studies have indicated that P. lilacinus and V. chlamydosporium can significantly suppress nematode populations. In vitro studies have shown that these fungi can readily destroy eggs of M. arenaria. Their mode of action is thought to involve both toxic metabolites and enzymatic degradation of eggs shells. Prospects for biological control using these fungi are thought to be good.

A332

RESPONSE OF RESISTANT AND SUSCEPTIBLE SOYBEANS TO CROPPING SYSTEMS IN AREA INFESTED WITH CYST NEMATODE. **L. D. Young and E. E. Hartwig, USDA-ARS, 605 Airways Blvd., Jackson, TN 38301 and P. O. Box 196, Stoneville, MS 38776, respectively.**

Management studies were conducted for 5 yr on soil infested with Heterodera glycines (Hg) race 4. Seven continuous-culture systems were six race 3 resistant lines with varying levels of resistance to race 4 (including 'Bedford' and race 4 susceptible 'Forrest') and a 70:30 Bedford:Forrest blend. Two rotations, Bedford with corn and Bedford with Forrest and Hg-susceptible 'Essex', were included. After 4 yr, Hg reproduction on Bedford in the greenhouse increased when the cultivar was planted in soil from continuous Bedford plots. However, the number of Hg recovered from soil in those plots did not increase. Number of cysts in soil of continuous Forrest plots was approximately 3 times the cysts in continuous Bedford plots. Five year continuous Bedford mean seed yield was 550 kg/ha greater than continuous Forrest. No system was superior in seed yield to growing Bedford continuously.

A333

PHYTOFERRITIN AND STARCH GRANULES IN DEVELOPING NODULES OF CYST-NEMATODE-INFECTED SOYBEANS. M. P. KO, P. Y. HUANG, J. S. HUANG, AND K. R. BARKER. Department of Plant Pathology, North Carolina State University, Raleigh, NC 27695-7616.

Structural changes in emerging soybean nodules as affected by race 1 of the soybean cyst nematode, Heterodera glycines, were investigated by light and electron microscopy. The most conspicuous features in the infected plants were the accumulation of starch granules and paracrystalline arrays of phytoferritin in the amyloplasts of the central nodular tissues. Although small starch granules occurred occasionally in similar tissues of control plants, phytoferritin was not observed. Starch and phytoferritin are respectively energy and iron reserves. Their accumulation in Rhizobium nodules of nematode-infected soybeans suggests that the metabolism of carbohydrates and iron-containing compounds is affected as a consequence of the presence of the nematode.

A334

COMPARISON OF TEMPERATURE AND CULTIVAR EFFECTS ON THE DEVELOPMENT OF HETERODERA GLYCINES IN SNAPBEAN AND TO SOYBEAN. **T. A. Melton, B. J. Jacobsen and G. R. Noel. 1984. Department of Plant Pathology and USDA/ARS, University of Illinois at Urbana-Champaign, Urbana, Illinois 61801.**

Development of the soybean cyst nematode was compared at 16, 20, 24, 28, and 32° C at 4 day intervals for 40 days on resistant (Fayette) and susceptible (Williams 79) soybean and on resistant (WIS RRR 36) and susceptible (Eagle) snapbean. Nematodes were stained with acid fuschin lactophenol and examined at 100x. Results indicate that rate of development is dependent on temperature in susceptible cultivars of each species. Degeneration due to temperature was noted at 32° and occurred rapidly in all four cultivars. Larval degeneration commonly occurred prior to development of third stage larvae. The mechanism of resistance in the resistant snapbean cultivar, WIS RRR 36, was apparently a hypersensitive reaction since necrotic cells were associated with degenerated larvae.

A335

POSSIBLE INVOLVEMENT OF HOPLOLAIMUS GALEATUS IN A DISEASE COMPLEX OF 'CENTENNIAL' SOYBEAN. McGawley, E. C., K. L. Winchell and G. T. Berggren. Dept. of Plant Path. and Crop Physiol., La. Agric. Exp. Sta., La. State Univ. Agr. Ctr., Baton Rouge, LA 70803.

Nematodes in the genus Hoplolaimus have been found associated with soybeans but pathogenicity has been demonstrated only with H. columbus. In 1983, lance nematode levels averaging 5,247 individuals per 500 cc soil were recovered from large patches of stunted 'Centennial' soybeans in a production field near Alexandria, La. Levels of Hoplolaimus in non-symptomatic (NS) areas averaged 640 individuals per 500 cc of soil. Randomly, 50 intact plants were collected from symptomatic (S) and NS areas and root systems dissected. All S and 15 NS plants contained active Hoplolaimus. Fungi in the genera Rhizoctonia, Fusarium, and Macrophomina were also isolated, from both S and NS plants. Subsequent greenhouse and laboratory work suggests that greater plant damage results from inoculation with both nematodes and these fungi than with either alone. On the basis of morphological and host range data, we have identified this nematode as Hoplolaimus galeatus.

A336

COMPARISON OF TWO EXTRACTION METHODS ON ESTIMATES OF SEASONAL FLUCTUATIONS OF GLOBODERA TABACUM SOLANACEARUM, A TOBACCO CYST NEMATODE. C. E. Grant and J. J. Reilly, Dept. of Plant Pathology, VPI & SU, Blacksburg, VA 24061 and Southern Piedmont Center, P.O. Box 448, Blackstone, VA 23824, respectively.

Field plots were maintained in the same location for two years. In the summer, the plots were planted to cultivars susceptible to Globodera tabacum solanacearum. A cover crop of winter wheat was planted in the fall and maintained until late spring. Soil samples were taken on a monthly basis and extracted by the centrifugation-sugar flotation (CSF) and sugar flotation (SF) methods. Cysts and larvae numbers peaked in September with the CSF method and in August or September with the SF method. Both methods detected a sharp increase in cysts populations in February and March 1982 that did not occur in that same period in 1983. The CSF method estimated higher numbers (by 25%) of cysts and larvae than the SF method during periods of maximum reproduction (summer), but estimated equal or lower numbers during the winter months.

A337

PHOTOSPOROGENESIS OF CERATOBASIDIUM SP. ON AGAR. J. Y. Uchida, M. Aragaki, and P. S. Yahata. Dept. of Plant Pathology, University of Hawaii, Honolulu, Hawaii 96822.

Several Ceratobasidium isolates (binucleate Rhizoctonia solani-like fungi), all in CAG-7 (Burpee's anastomosis group 7), developed basidiospores in axenic agar culture. There was wide variation in reproductive capacities among isolates. Under conditions favorable for sporulation, the most prolific isolate produced 500-2000 x 10^3 spores/60 mm petri dish culture in 12 days. Meager sporulation in cultures grown in continuous darkness and profuse sporulation in cultures grown under fluorescent illumination established that light is not required, although highly stimulatory. Sporulation was abundant on V-8 juice agar (5 - 20% V-8 juice), low on cornmeal agar, but did not occur on potato dextrose, Czapek's, or Mycophil agars. Optimal temperature for sporulation was 24 C. At 28 C, light stimulated formation of basidia, but was inhibitory to basidiospore formation.

A338

EFFECT OF DILUTION OF MEDIA CONSTITUENTS ON SPORULATION AND GROWTH OF BIPOLARIS MAYDIS RACE T IN VITRO. E. O. Bassey and M. O. Garraway, Dept. of Plant Pathology, OARDC and The Ohio State Univ., Columbus, OH 43210.

Bipolaris maydis race T (BMT) was incubated for 10 days in the

dark, at 28 C, on a medium containing 5 g glucose, 4 g L-asparagine, 1.5 g KH_2PO_4, 0.75 g $MgSO_4 \cdot 7H_2O$, and 20 g Difco agar per liter of distilled water. Sporulation and growth on this medium were compared with that on similar media in which all constituents other than agar were diluted. Sporulation on undiluted medium or media diluted 1:8, 1:16, 1:32, 1:64 or on water agar was 3,000, 1,900, 1,600, 4,200, 7,300 and 8,200 conidia/mg dry wt respectively. In contrast, growth on these media was 45, 13, 9, 5, 3 and 3 mg dry wt per thallus. Similar trends in sporulation and growth in response to dilution were seen when glucose or/and L-asparagine, but not mineral salts, were the only media constituents in agar. These data suggest that organic constituents in media suppress sporulation of BMT at concentrations which enhance growth. Our findings help to explain the effects of corn leaf leachates on sporulation of BMT in vivo.

A339

ROLE OF CYCLIC AMP IN DIFFERENTIATION OF UROMYCES PHASEOLI UREDOSPORE GERMLINGS. L. Epstein, R.C. Staples, and H.C. Hoch. Boyce Thompson Institute, Ithaca, NY 14853 and NY State Agricultural Experiment Station, Cornell University, Geneva NY 14456.

Nuclear division, an early event in the process of appressoria formation by bean rust uredospore germlings, can be induced by cyclic AMP (cAMP). To determine if changes in cAMP concentration are associated with nuclear division, we monitored extractable cAMP in germlings induced to differentiate chemically by 50 mM K^+ or physically by scratches on a polyethylene sheet. In both treatments and in a non-induced control, spore cAMP (ca. 1.5 picomoles cAMP/mg spore) decreased during spore hydration to ca. 0.7 picomoles cAMP/mg and then remained at a fairly constant level during germination, hyphal elongation, nuclear division and differentiation. As a more sensitive means to detect cAMP involvement in nuclear division, we are currently reacting cAMP photoaffinity probes with differentiation-induced proteins.

A340

THIGMODIFFERENTIATION AND CHEMODIFFERENTIATION MAY BE DIFFERENT PROCESSES IN THE RUST FUNGI. R. C. Staples, S. Hassouna, and H. C. Hoch. Boyce Thompson Institute, Ithaca, NY 14853, and NY State Agricultural Experiment Station, Cornell University, Geneva, NY 14456.

Bean rust uredospore [Uromyces phaseoli] germlings are induced to differentiate infection structures by several chemicals (e.g K^+, Ca^{+2}, sugars, cAMP) or by germ tube contact with an appropriate surface contour such as a scratch on a membrane. In a comparison, we found that both processes induced an increased rate of protein synthesis and the start of DNA replication. However, mitosis induced by chemicals begins about 6 h after the start of germination, while with contact stimuli it commences within 3 h. Further, while K^+ induced synthesis of both mitochondrial and nuclear DNA, contact stimuli induced synthesis only of nuclear DNA. While the data show that both processes start mitosis and formation of appressoria, thigmodifferentiation is a more specific physiological response with a faster reaction time. In nature, this may ensure accurate positioning of the appressorium over the stomatal opening.

A341

EFFECT OF PECAN PHYLLOPLANE EXUDATES ON GERMINATION OF CLADOSPORIUM CARYIGENUM. T. R. Gottwald and B. W. Wood, USDA-ARS, SE Fruit & Tree Nut Res. Lab., P.O. Box 87, Byron, GA 31008.

Leaf surface exudates were washed from immature and mature foliage of cv. 'Schley' and cv. 'Stuart' pecan with sterile distilled deionized water. Following washing, foliage was dipped for 10 sec in methylene chloride to partially remove the cuticle and entrapped exudates. All exudates were concentrated and fractionated via TLC. Fractions were adjusted to leaf surface concentrations per unit area, spotted on glass ring slides and dried. Laboratory grown Cladosporium caryigenum inoculum (conc. 2.5 x 10^5 spores/ml) of 'Schley' and 'Stuart' pathovars was spotted on the slides and allowed to germinate for 48 hr, 100% RH, 25°C in darkness. Fractions could be separated by their inhibitory or stimulatory effect on germination. Certain cv. 'Schley' fractions had inhibitory effect on 'Stuart' pathovar inoculum and vice versa indicating a chemical based physiological cultivar/pathovar specialization.

A342

CUTINASE INDUCTION IN GERMINATING SPORES OF Fusarium solani f. sp. pisi. C.P. Woloshuk, C.L. Soliday and P.E. Kolattukudy, Institute of Biological Chemistry, Washington State University, Pullman, WA 99164.

If cutinase is involved in the penetration of Fusarium solani f. sp. pisi into its host as previously suggested, this enzyme must be available to the germinating spores. The enzyme activity was, in fact, spectrophotometrically detectable 1 hr after addition of cutin to spore suspensions and increased rapidly over the next 5 hr. This activity was verified as cutinase using radiolabelled cutin as well as by inactivation with antibody. No cutinase activity was found in the absence of cutin or cutin hydrolysate in the medium. Hybridization with labeled cutinase cDNA showed that the cutinase mRNA level in the spores increased due to the presence of cutin or cutin hydrolysate in the medium. These changes occurred prior to germination of the spores indicating that cutinase induction by enhanced transcription of cutinase gene is an early event in the spore germination process.

A343

PHYSICAL AND CHEMICAL FACTORS AFFECTING VIABILITY AND GERMINATION OF OOSPORES OF PHYTOPHTHORA MEGASPERMA F. SP. MEDICAGINIS (PMM). Z. A. El-Hamalawi and D. C. Erwin. Department of Plant Pathology, University of California, Riverside, CA 92521.

Freezing (-15 C) PMM oospores in carrot broth for 0, 1, 2, 3 or 4 days reduced the % viability [measured by the tetrazolium bromide vital stain (Sutherland and Cohen, 1983. Phytopathology 73: 1532-1535)] from 98 to 95, 79, 55, and 17% respectively. Oospores became dormant, and germination was decreased. Viability and germinability of oospores frozen in water were reduced much more than oospores frozen in carrot broth. Lyophilization reduced viability and germinability to 0. Dessication of oospores in carrot broth reduced viability from 96% to 29% and germinability in water from 22% to 7%. Germination of oospores treated for 10 mins with 0, 0.025, 0.05, 0.10, 0.5, and 1% $KMnO_4$ was 25, 49, 77, 69, 7 and 0%, respectively, when incubated in water, and 58, 70, 87, 78, 12, and 0%, respectively, when incubated in alfalfa root extract. Viability and germinability were not affected by gluculase (5%).

A344

EFFECTS OF ALFALFA ROOT EXTRACT AND EXUDATE ON OOSPORE GERMINATION OF PHYTOPHTHORA MEGASPERMA F. SP. MEDICAGINIS (PMM). Z. A. El-Hamalawi and D. C. Erwin, Department of Plant Pathology, University of California, Riverside, CA 92521.

Incubation of PMM oospores either in alfalfa root extract or exudate increased oospore germination from 36% in distilled water to 86%. Sporangia arising from germinated oospores increased significantly in proportion to the concentration of extract but only up to a certain maximum concentration, after which sporulation but not germination declined. Lyophilization of extract enhanced sporangial formation on germinated oospores more than an untreated crude preparation, however, % germination was similar on both crude and lyophilized extracts. Compounds in root extract with a molecular weight greater than 6,000 induced the greatest increase in % oospore germination. Since autoclaved root extract or exudate also stimulated oospore germination, stimulation is not likely to be due to specific proteins. Oospore germination was greatest in the cationic, least in the anionic and intermediate with a neutral fraction of root extract or exudate.

A345

INFLUENCE OF TEMPERATURE, LIGHT AND pH ON GERMINATION OF TILLETIA INDICA TELIOSPORES. J. L. Smilanick and J. A. Hoffmann, USDA-ARS, Logan, Utah 84322.

Teliospores of karnal bunt fungus, Tilletia indica, obtained from Sonora, Mexico, were rinsed from infected wheat seeds with 0.01% Triton X-100 and surface sterilized by a 2 min rinse in 0.0025% sodium hypocholorite (pH 10.5). Percent germination of teliospores incubated on water agar (pH 7.0) or soil-extract agar (pH 6.7) in darkness for 5 wk at 5C, 10C, 15C, 20C, and 25C (±1C) was ca. 15, 35, 55, 45, and 35, respectively. The optimum temperature was ca. 15C. Percent teliospore germination (±S.D.) after 3 wk incubation on soil-extract agar at 5C, 10C, 15C, and 20C (±1C) with continuous light (30 $\mu E/m^2$, white fluorescent) was 11.3 (±4.6), 49.9 (±5.4), 56.2 (±2.9) and 39.4 (±6.6), respectively, whereas germination in darkness was 2.8 (±1.0), 33.3 (±3.3), 42.0 (±2.9) and 8.9 (±2.3), respectively. Light stimulated germination. Teliospores were incubated 2 wk on water agar buffered with 1.0 8/1 each of glycylglycine and sodium bicarbonate and adjusted to a range of hydrogen ion concentrations. Germination occurred equally over a pH range from 6.0 to 9.3 but was strongly inhibited below 4.5 or above pH 10.0.

A346

FUNGAL FIMBRIAE AND HOST INFECTION. Day, A. W., Svircev, A. M.,

Smith, R. and Gardiner, R. B. Dept. of Plant Sciences, University of Western Ontario, London, Ontario N6A 5B7

The presence on fungal cells of fibrillar protein polymers (fimbriae) varying in length from 0.5 μm to over 20 μm has obvious possible implications to the infection process. Fimbriae are narrow enough (7 nm) to penetrate plant cell wall plasmodesmata and therefore to allow intimate contact between the pathogen and host organelles, even in the case of fungi which are extracellular pathogens. Once in contact with such organelles fimbriae could serve to transmit signals, directing host genes or their products to meet the pathogen's need for nutrition. We report here initial results using protein A-gold immunocytochemical staining to identify and locate fimbrial proteins in infected host plants. Specific labelling has been detected both in and on the extracellular hyphae of Ustilago species and within the neighbouring host plant cells, indicating that extensive penetration of host cells by fungal fimbriae occurs.

A347

EFFECTS OF ANTIFIMBRIAL ANTISERA ON DEVELOPMENT OF INFECTION STRUCTURES IN THE BEAN RUST FUNGUS. Kaminskyj, S. and Day, A. W. Dept. of Plant Sciences, University of Western Ontario, London, Ontario N6A 5B7.

Germinating urediospores of Uromyces phaseoli var. typica develop infection structures (IS) in the absence of host plants in response to both physical and chemical stimuli. The hyphae produce a surface protein of similar M.W. and antigenicity to the fimbrial protein of Ustilago sp. Treatment with antifimbrial antisera and also other proteins markedly decreased IS frequency when the inducing signal was a chemical one, but not when it was a physical factor. IS formation on host leaf surfaces and the subsequent development of uredia were reduced by treatment with antifimbrial antisera. Although not conclusive these results suggest a possible involvement of surface fimbrial proteins in the response to chemical stimuli in the bean rust fungus.

A348

TOLERANCE OF THE PHYTOALEXIN, KIEVITONE, IS ASSOCIATED WITH VIRULENCE IN FUSARIUM SOLANI F. SP. PHASEOLI. Harry Wheeler and David Smith, Department of Plant Pathology, University of Kentucky, Lexington, Ky, 40546-0091.

Several variants of Fusarium solani f. sp. phaseoli (Fsp), isolate FB1-S, a virulent strain causing progressive hypocotyl lesions in Phaseolus vulgaris (French bean) (Phytopathology 72: 1319-1323), were obtained either following exposure of the fungus to the mutagenic chemical, N-methyl-N'-nitro-N-nitroso-guanidine, or by making transfers from naturally occurring sectors in agar cultures. Fusarial isolates were subsequently monitored for their pathogenicity to bean, their sensitivity to kievitone and their ability to cause clearing of an opaque, kievitone-amended, modified Fries agar - an assay taken as presumptive evidence of kievitone hydratase (KHase) activity. All isolates with apparently reduced KHase activity were more sensitive to kievitone and were less pathogenic to bean than FB1-S. No virulent isolate was found which appeared to lack KHase. Tolerance to the isoflavonoid phytoalexin, kievitone, by Fsp may be important to the pathogenicity of this fungus.

A349

POLAR LIPIDS FROM PEA REDUCE THE PISATIN SENSITIVITY OF APHANOMYCES EUTEICHES. J. A. Sweigard and H. D. VanEtten, Dept. of Plant Pathology, Cornell Univ., Ithaca, NY 14853-0331.

Aphanomyces euteiches readily invades pea tissue despite the presence of high pisatin concentrations in the infected tissue. In vitro, however, the fungus is very sensitive to this phytoalexin. When polar lipid extracts from pea epicotyls were added to growth media, the pisatin sensitivity of A. euteiches decreased. Similar extracts from other plants produced the same effect. Pea polar lipids did not decrease the pisatin sensitivity of Neurospora crassa or Fusarium solani f. sp. cucurbitae. The pea polar lipids also decreased the sensitivity of A. euteiches to the phytoalexins phaseollin and maackiain. Attempts to purify the polar lipids indicated that no single compound was responsible for the activity. The decrease in pisatin sensitivity could not be accounted for by physical sequestering of the pisatin by the lipids. Also, the lipids did not increase the rate of pisatin demethylation by the fungus.

A350

AMMONIUM PRODUCTION ON L-ASPARAGINE: ITS RELATIONSHIP TO THE PH IN CULTURES OF BIPOLARIS MAYDIS RACE T. T. W. Bischoff and M. O. Garraway, Dept. of Plant Pathology, OARDC and The Ohio State Univ., Columbus, OH 43210.

To explore the relationship between ammonium production and pH increases in cultures of Bipolaris maydis race T, we incubated the fungus with 1.0, 2.0 4.0 or 8.0 g/l of L-asparagine (L-asn) or L-aspartate (L-asp) on a glucose (10 g/l)-mineral salts agar medium (pH adjusted to 5.8 with NaOH). After 6 days of incubation in the dark at 28 C the pH rose, increasing with increasing concentration to 7.4 with 8 g/l. NH_4^+ was detected in media containing L-asn, but not in those with L-asp. When we titrated the medium (lacking either amino acid) with L-asn or L-asp, we found that addition of L-asn had no effect on pH while L-asp caused a pH change. When an unseeded basal medium containing L-asn was incubated with L-asparaginase from Escherichia coli, NH_4^+ was released but the pH did not change. Thus, when B. maydis race T is incubated on L-asn the increase in pH may be due to the utilization of L-asp which results in the accumulation of NH_4^+ in the culture medium.

A351

CELL-FREE TRANSMISSION OF HYPOVIRULENT PHENOTYPE OF ENDOTHIA PARASITICA. N. K. Van Alfen, D. R. Hansen, S. Miller, L. Barley. Department of Biology, Utah State University, Logan, UT 84322.

Hypovirulence of Endothia parasitica has been associated with the presence of dsRNA. To date, there has been no direct evidence by cell-free infection of the role of dsRNA in causing hypovirulence. The dsRNA is naturally packaged within membrane vesicles in the hypovirulent (H) strains EP-113. We have isolated these membrane vesicles and fused them with protoplasts of virulent (V) strains of the fungus. As an initial screen for cell-free transmission, we transferred regenerated protoplasts to media that distinguishes H strains from V strains on the basis of pigment formation. In typical experiments, we have found that from 1-5% of the regenerated protoplasts possessed the H phenotype. None of the controls had this phenotype. The H phenotype was not stable upon transfer, but experiments to remedy this are in progress.

A352

HIGH FREQUENCY MUTATION OF A VIRULENCE REGULATORY SITE IN ENDOTHIA PARASITICA. N. K. Van Alfen, D. R. Hansen, S. Miller, Department of Biology, Utah State University, Logan, UT 84322.

We have observed an unusual genetic phenomenon in which certain strains of Endothia parasitica can be mutated at high frequency to phenotypically mimic hypovirulent forms of this fungus. The mutation can be induced by growing the strains for a short period of time on high osmotic medium. The mutants lack pigmentation, and show reduced sporulation and virulence, yet lack detectable dsRNA which is present in naturally-occurring hypovirulent strains. This ability to be induced to the hypovirulent phenotype by osmotic shock (os) is recessive and most likely a cytoplasmic trait, as determined by heterokaryon tests. Genetic studies on the white or non-pigmented phenotype associated with os show that: 1)it is recessive; 2)it is nuclear; 3) it does not complement; and 4)it does not revert. We hypothesize that os is a regulatory locus that affects the control of certain phenotypic expressions, i.e. virulence, sporulation, and pigmentation.

A353

DISCOVERY OF PLASMIDS IN *CERATOCYSTIS ULMI*. Shozo Takai[1], Toshihiko Iizuka[2], and Wayne C. Richards[1]. [1]Great Lakes Forest Research Centre, Department of the Environment, Sault Ste. Marie, Ontario, Canada, P6A 5M7; [2]Faculty of Agriculture, Hokkaido University, Sapporo, Japan.

Five *C. ulmi* isolates, aggressive or non-aggressive in pathogenicity, were examined for the presence of plasmids. At least six different plasmids were obtained after gel electrophoresis. Approximate size of these plasmids are 22,18,3 (two species), 2 and 0.54 Kb. Plasmids were not identified in one isolate (X6) of the non-aggressive strains. A common plasmid species of about 22 Kb was found in four other isolates, irrespective of their aggressiveness and/or cerato-ulmin (CU) productivity. Two isolates of the aggressive strain (H323 and H220) and one non-sporulating isolate (WRB2-1), all of which are distinct in CU production, showed the presence of plasmids lower than 22 Kb. Neither of the two non-aggressive isolates neglibible in CU production (X6 and YU99) indicated the presence of any plasmids of these areas. Extended screening among strains and races of *C. ulmi* is also under way.

A354

IDENTIFICATION OF A GENE FOR BROWN STEM ROT RESISTANCE IN SOYBEANS. Scott A. Sebastian and Cecil D. Nickell, Dept. of

Agronomy, Turner Hall, 1102 S. Goodwin Ave., Urbana, IL 61801.

Brown stem rot (BSR), caused by Phialophora gregata (Pg) is a serious disease of soybean (Glycine max). Although sources of BSR resistance are available, little is known about the inheritance of BSR resistance. A cross between a susceptible and resistant soybean line was selected for a genetic study. An attempt was made to classify F3 families from this cross as either resistant, segregating, or susceptible for a classical Mendelian genetic study. Cluster analysis based on leaf symptom mean, stem symptom mean, and leaf symptom variance was used as the basis for classifying F3 families. A single dominant gene for resistance can account for the segregation for BSR resistance in this cross. This study exemplifies how the quantitative nature of certain traits can mask the expression of a simple inheritance pattern.

A355

ADDITIVE EFFECTS OF ALFALFA GENES CONDITIONING RESPONSE TO PERONOSPORA TRIFOLIORUM. D.Z. Skinner and D. L. Stuteville, Dept. of Plant Path. Kansas State University, Manhattan, 66506.

F_1 seedlings of diploid plants P5 and P6 were inoculated with conidia of isolate I-7. Infection types (ITs) were rated zero (no conidial production) to five (copious production). Four plants from high (H) ITs (greater than zero) were selected, as were six F_1 seedlings of plants P1 and P2. F_2 populations of each plant and F_1 populations from (P1 X P2) X (P5 X P6) crosses of plants of similar ITs were derived and inoculated. As all plants involved were from HIT interactions, no genes with complete dominance conditioning low IT were involved. ITs on F_2s indicated that plants of P5 and P6 parentage had more genes capable of combining to condition decreased IT than did plants of P1 and P2 parentage. The mean ITs of populations derived by crossing F_1s were intermediate between or exceeded the mean ITs of the S_1 populations of the plants crossed. There was no evidence of simply inherited genes with complete dominance conditioning a specific IT. Apparently many genes with additive effects were involved.

A356

GENETIC RELATIONSHIPS OF RESISTANCE IN TWO BROADLY RUST RESISTANT BEANS. J. R. Stavely, USDA, ARS, BARC-W, Beltsville, MD 20705.

Phaseolus vulgaris 'Compuesto Negro Chimaltenango' (CNC) has small pustule resistance (R) or immunity (I) to all 20 recently described races of Uromyces phaseoli (Plant Disease 68:95-99). Breeding line B-190 has similar R to many races, but is susceptible (S) to 3 races to which CNC has R. B-190 has a necrotic hypersensitive resistance (HR) to two races to which CNC has I. The resistance of B-190 is controlled by a linked series of monogenic dominant factors, one for each U. phaseoli race (Phytopathology 74:339-344). None of 468 F_2 plants from reciprocal crosses between CNC and B-190 was S to races to which the parents both had R, or to which CNC had I and B-190 R or HR. Hence these two parents contain the same or different alleles at single loci for reactions to each of the races to which both have R or CNC has I and B-190 R or HR. The R of CNC to the races to which B-190 is S is controlled by additional linked single dominant R genes, one per race.

A357

A RAPID, SENSITIVE, HIGH RESOLUTION ACTIVITY STAIN FOR DETECTING AND DIFFERENTIATING PECTIC ENZYMES IN GELS. Jeffrey L. Ried and Alan Collmer. Department of Botany, University of Maryland College Park, MD 20742.

A technique was developed for rapid identification of pectic enzymes resolved by isoelectric focusing (IEF) in ultrathin polyacrylamide gels. A 0.4 mm agarose gel containing 0.1% galacturonan and an appropriate buffer is cast on GelBond (FMC Corp.) and then overlayed on the IEF gel. After an incubation of 5 to 60 minutes or more, depending on the desired sensitivity, the agarose overlay is stained with 0.05% ruthenium red for 20 minutes and then rinsed with water. Clear bands in a red background appear where enzyme was present in the IEF gel. By altering the pH of the overlay the endopectic lyases and hydrolases produced by Erwinia chrysanthemi, Aspergillus niger and Sclerotium rolfsii can be differentially detected. The overlay technique is sufficiently sensitive to detect 0.001 activity units (μmoles product/min) per cm^2 in a 10 minute incubation and has permitted sharp resolution of seven bands of pectolytic activity in culture supernatants of E. chrysanthemi strain 1237.

A358

MUTANTS OF PSEUDOMONAS FLUORESCENS INCAPABLE OF GROWTH UNDER IRON LIMITING CONDITIONS. J. I. Stein, T. C. Currier, F. M. Solan, W. D. Gould. Allied Corporation, Syracuse Research Laboratory, Solvay, New York 13209

Pseudomonas fluorescens strain TR21 grows in minimal medium containing the iron chelator EDDA, suggesting the production of a siderophore. Mutants unable to grow in the presence of EDDA were obtained by transposon mutagenesis. The Tn5-containing plasmid pSUP1011, incapable of replicating in Pseudomonas species, was introduced into strain TR21 from E. coli strain SM10. Mutants were selected on minimal medium containing kanamycin. Colonies that either failed to grow or grew poorly on an EDDA-containing medium were further examined. Lack of vector DNA and presence of a single copy of Tn5 was confirmed by southern hybridization. The mobility of the hybridizing DNA from 49 mutants suggests that the affected genes are contained in at least 10 Eco RI fragments. Forty-three mutants failed to produce a yellow-green fluorescent pigment characteristic of wild-type TR21 growing under iron-limiting conditions. The growth of these mutants on EDDA-containing medium was restored either by adding exogenous iron or a compound partially purified from TR21 culture supernatants.

A359

A DEFINED MEDIUM FOR GROWTH AND SPORULATION OF PYRENOPHORA TRITICI-REPENTIS, CAUSAL AGENT OF TAN SPOT OF WHEAT. Robert M. Hunger, Plant Pathology Department, Oklahoma State University, Stillwater, OK 74078

The undefined medium V-8 juice agar (V8) supports growth and conidia formation of P. tritici-repentis (PTR) when colonies are grown at 21 C with a 12:12 photoperiod (Can J Bot 55:254-259). However, there is no defined medium reported that supports growth and sporulation of PTR comparable to that on V8. Thus, a defined medium was developed. Each liter of medium contained 1.52g KH_2PO_4, 1.3g K_2HPO_4, 5g glucose, 500mg L-asparagine, 1 ml of an aqueous salt solution (100μg $MnCl_2 \cdot 4H_2O$, 100μg $ZnSO_4 \cdot 7H_2O$, 100μg $FeCl_3 \cdot 6H_2O$, 22μg $CuSO_4 \cdot 5H_2O$, 20μg $BaCl_2 \cdot 2H_2O$, 10μg $CaCl_2 \cdot 2H_2O$), and 1.5% agar. Most PTR isolates tested had colonial diameters of approximately 65mm and 35mm after 6 days at 21 C on V8 and on the defined medium, respectively. Formation of conidia on the defined medium was 50-75% of that on V8. This defined medium should facilitate studying the genetics of PTR through mutagenesis and gene mapping.

A360

A DESCRIPTION OF MUTANTS OF AGROBACTERIUM TUMEFACIENS WHICH FAIL TO ATTACH TO SUSPENSION CULTURE CELLS. Ann G. Matthysse and Elaine Steele. Department of Biology, University of North Carolina, Chapel Hill, NC 27514

Transposon Tn5 mutants of A. tumefaciens were screened for their ability to bind to carrot suspension culture cells. Five mutants which failed to bind to carrot cells were isolated. All of these mutants were avirulent on Bryophyllum leaves and carrot discs. Revertants of the nonbinding mutants which recovered the ability to bind to carrot cells were isolated by incubating the plant cells with mutant bacteria and then collecting the plant cells with any associated bound bacteria. Bacteria which had recovered the ability to bind to carrot cells simultaneously recovered virulence. This result suggests that the ability of A. tumefaciens to bind to suspension culture cells is required for bacterial virulence. Surface properties of the parent strain, nonattaching mutants, and revertants were compared. No alterations in bacterial motility or in size of LPS were observed in any of the nonbinding mutants.

A361

HYPHAL INTERACTIONS AMONG SINGLE SCLEROTIAL ISOLATES OF SCLEROTINIA MINOR. C. L. Patterson and R. G. Grogan, Department of Plant Pathology, University of California, Davis, CA 95616

Fifty-seven single sclerotial isolates of S. minor from different locations were mated in all possible combinations. Three hyphal interactions resulted: 1) anastamosis between the paired isolates; 2) anastamosis followed by lysing of cells involved in anastamosis; and 3) anastamosis of hyphae followed by copious spermatia formation in the area of anastamosis. The individual isolates form 28 compatibility groups based on their ability to anastamose as in reaction 1. Members within a group have been obtained from several different and widely separated locations. Vegetative compatibility has been a criterion for differentiating S. minor, S. sclerotiorum, and S. trifoliorum. However, these results indicate there is considerable heterogeneity for vegetative compatibility within the species S. minor. Thus, we question the validity of anastamosis as a criterion for speciation.

A362

EVIDENCE FOR HETEROTHALLISM IN SOME ISOLATES OF SCLEROTINIA MINOR. C. L. Patterson and R. G. Grogan, Department of Plant Pathology, University of California, Davis, CA 95616.

Hyphal anastamosis among some single-sclerotial isolates of S. minor resulted in copious formation of spermatia in the area where anastamosis occurred. When sclerotia of such an isolate were spermatized by the other (and vice versa), apothecia were consistently produced in 6-8 wks. Non-spermatized sclerotia of each parent isolate did not produce apothecia. Single ascospore isolates from apothecia produced by spermatized sclerotia segregated in a 1:1 ratio for both parents; i.e. approximately 50% of the vegetative crosses resulted in the copious formation of spermatia whereas the others did not. Although some isolates of S. minor are homothallic the majority are heterothallic. Apothecial formation usually is not important in the epidemiology of S. minor in the field. This may be due in part to the fact that most isolates are heterothallic and sexually compatible strains usually are separated geographically.

A363

INDUCTION AND SELECTION OF COLLETOTRICHUM GLOEOSPORIOIDES MUTANTS DEFICIENT IN PRODUCTION OF CUTINASE. Martin B. Dickman and Suresh S. Patil, Department of Plant Pathology, University of Hawaii, Honolulu, Hawaii 96822.

C. gloeosporioides, the causal agent of papaya anthracnose, utilizes a cutinolytic enzyme to penetrate and infect papaya fruit. To further confirm the role of this enzyme, mutants of the fungus lacking in cutinase (Cut^-) were made. Following mutagenesis, spores were plated on minimal medium. Survivors were screened for Cut^- mutants by transferring them to a modified Czapek-Dox medium containing purified cutin as the sole carbon source and a pH indicator dye (phenol red). Wild type spores (Cut^+) grew and turned the medium yellow, presumably due to release of fatty acids generated by cutin hydrolysis. Spores failing to produce mycelia were unable to change the color of the dye, constituting Cut^- mutants. The Cut^- phenotype was confirmed by testing culture filtrates for cutinase activity and by using rabbit anticutinase in an indirect ELISA technique. In a laboratory fruit bioassay, Cut^- mutants did not infect papaya without wounding.

A364

ELECTROPHORETIC STUDY OF ISOZYMES IN Uromyces appenticulatus. Lu Tze Hong and J.V. Groth. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

About 100 mg of uredospores of 12 single uredial isolates of bean rust from different geographical areas and differing in virulence pattern were germinated, ground and placed on polyacrylamide tube and starch slab gels for separation of isozymes. Fourteen enzymes were detected. Of these, some 22 variant band patterns were detected among isolates (the band pattern shown by the majority of isolates was taken as the standard for each enzyme). According to band patterns, the isolates could be grouped into three categories, named for the most susceptible cultivars used to increase them respectively, 1) the Bush Blue Lake isolate, 2) the Topcrop isolate and 3) the Pinto isolates. Between categories the isozyme variation averaged 40% while in the pinto group, the variation ranged from 4-13%. Virulence pattern of isolates did not associate closely with isozyme pattern for most isolate comparisons.

A365

CHARACTERIZATION OF LEPTOSPHAERULINA SPP. FROM ALFALFA, CLOVER AND PEANUT. C. Lee Campbell and Wayne M. Thal, Dept. Plant Pathology, North Carolina State University, Raleigh 27695.

Seven isolates of Leptosphaerulina spp. from alfalfa, two from clover and one from peanut were morphologically characterized as L. briosiana (Lb), L. trifolii (Lt), and L. arachidicola, respectively. Isolates were grown on V-8 juice agar at 12-32 C. Maximum growth occurred at 20 or 24 C for all isolates. Pathogenicity was examined on detached leaves of four alfalfa cvs., four clover spp., peanut and soybean. Leaves were placed on moistened filter paper, inoculated with spores ejected from actively growing cultures and maintained in moist chambers. Lesions were often evident within 24 hr and assessments made 4-5 days after inoculation. Significant differences ($P=0.0001$) in disease severity were found among hosts and isolates. A significant ($P=0.0001$) host by isolate interaction was found. These results indicate the diversity present in spp. and isolates of Leptosphaerulina.

A366

SOIL SUPPRESSIVENESS TO A PATHOGENIC PYTHIUM SP. R. Lifshitz, B. Sneh and R. Baker, Botany and Plant Pathology Department, Colorado State University, Fort Collins, CO 80523.

When dried bean leaves were added at weekly intervals to Nunn sandy loam soil (pH 7.3 at 26 C) over a 6-wk period decrease in propagule density of a pathogenic species of Pythium was observed. Induction of suppressiveness in this soil was not observed, with the same treatments incubated at 19 C or in soil at pH 5.0 incubated at 26 C. The decline in propagule density of the pathogenic Pythium sp. was associated with an increase in population density of an unidentified species of Pythium. The latter was mycoparasitic to the pathogenic Pythium sp. in vitro and induced early lysis to germinating sporangia of this pathogen on membrane fillers laid on soil infested with the isolates. Addition of the antagonist to soil suppressed the competitive saprophytic ability of the pathogenic Pythium sp. and also its capacity to induce pre-emergence damping-off of cucumber. Therefore it induced both pathogen and disease suppressiveness.

A367

DYNAMICS OF PYTHIUM ULTIMUM INFECTION AND DAMPING-OFF OF SUGAR BEETS AND BIOCONTROL BY A STRAIN OF PSEUDOMONAS PUTIDA. R. M. Osburn and M. N. Schroth, Department of Plant Pathology, University of California, Berkeley, CA 94720.

Pythium ultimum infected sugar beet seeds within 4 hr after planting in loamy sand soil at a matric potential of -0.1 bar and day/night temperature of 21/16 C in growth chamber experiments. Within 24 hr, 75-100% seed coat infection occurred. Subsequent seed decay and damping-off usually resulted from the spread of infection from the initially infected seed coat. Pseudomonas putida strain R20, which was previously shown to reduce the incidence of P. ultimum damping-off in the greenhouse, reduced seed coat infection 45-79% by 24 hr. Of those R20-treated sugar beet seeds that became infected, most infections (44-71%) died out and could not be detected. The combined effects resulted in reductions in the incidence of seed decay and damping-off of 42-66%. After initial application of 10^7-10^8 colony forming units (cfu) per seed, external R20 seed coat populations initially doubled, then decreased to 10^5-10^6 cfu after 6 days while internal populations were 3×10^6 within the seed coat by 24 hr. It is possible that the internal colonization by R20 may be responsible for the dying out of P. ultimum infections.

A368

THE USE OF PYTHIUM OLIGANDRUM FOR THE BIOLOGICAL CONTROL OF PYTHIUM ULTIMUM. F. N. Martin and J. G. Hancock, Department of Plant Pathology, University of California, Berkeley, CA 94720.

The feasibility of using P. oligandrum as a biological control agent against P. ultimum was investigated in greenhouse studies. Pelleting of sugar beet seeds with P. oligandrum reduced the colonization of seed coats by P. ultimum from 96% to 16% and of embryo colonization from 61% to 0%. In soil with 13 propagules/g of P. ultimum, there was 60% emergence of non-treated seeds compared to 79% for pelleted seeds, and root colonization by P. ultimum was 60% and 6%, respectively. With pelleted seeds, the emerging radicle was colonized by P. oligandrum and was effectively protected from damping off. After 18 days, stand counts for pelleted seeds were 50% to 150% greater than non-treated controls and seedlings had greater shoot weight, root density and fewer hypocotyl infections by P. ultimum. The incidence of feeder root infections by P. ultimum was not reduced by seed pelleting. With the exception of feeder root infection, P. oligandrum pelleting gave almost as good control of P. ultimum as did fenaminosulf seed treatments.

A369

FURTHER INVESTIGATIONS ON THE USE OF LAETISARIA ARVALIS TO CONTROL RHIZOCTONIA SOLANI INFECTION OF POTATO. C.W. Murdoch, and S.S. Leach, N.E. Plant, Soil and Water Lab., Orono, ME 04469.

In preliminary tests, L. arvalis was shown to inhibit R. solani infection of potato (Amer.Pot.J. 60:813,1983). A 2 yr field study to evaluate the effect of liquid or dry L. arvalis seed treatments and soil ammendments on R. solani-caused damage was completed. Treatments and ammendments applied to plots were: L. arvalis; L. arvalis + R. solani; R. solani; PCNB + thiabendazole (50000 + 1500ppm); and none. Fungal populations (propagules/gm soil) were determined 8 and 12 wks after planting using a soil pellet-selective medium method. Emergence was determined at 4 and 6 wks, and disease incidence and severity were assessed 6 and 10 wks after planting. Total and marketable yield were determined at harvest. In 1983, dry L. arvalis preparations were comparable to the chemical treatments for disease control and marketable yield. Dry seed treatments alone resulted in significantly higher ($P=0.05$) L. arvalis soil populations. L. arvalis showed effective biological control of R. solani.

A370

ADDITION OF FOOD-BASE COMPOUNDS TO BIOLOGICAL SEED TREATMENTS FOR THE IMPROVEMENT OF TRICHODERMA BIOCONTROL ACTIVITY. E.B. Nelson, G.T. Nash, and G.E. Harman. Dept. of Horticultural Sciences, Cornell University, NYSAES, Geneva, New York 14456

Over 100 compounds, including organic acids, sugars, phenolics, and industrial by-products were evaluated for their ability to support the growth of Trichoderma spp. Compounds supporting growth were coated onto pea seeds along with conidia of the biocontrol agents: T. koningii or T. harzianum and planted in a Pythium-infested soil. Different seed treatments were evaluated after 6 days for their ability to improve seedling stands over seeds treated with Trichoderma spp. alone. Changes in microbial populations around seeds were also determined after 1 and 6 days. Treating seeds with some compounds together with Trichoderma resulted in increases in both plant stands and Trichoderma populations. This was generally accompanied by a decrease in Pythium populations. Application of other compounds to seeds together with Trichoderma resulted in increased plant stands although Trichoderma populations were unaffected. The possible mode of action of select compounds is discussed.

A371

TRICHODERMA SPP. AS BIOLOGICAL CONTROL AGENTS OF RHIZOCTONIA DAMPING-OFF OF RADISH IN OHIO MUCK SOILS. L. J. Mihuta and R. C. Rowe, Department of Plant Pathology, The Ohio State University, Ohio Agricultural Research and Development Center, Wooster 44691.

Eighty Trichoderma isolates were tested in the greenhouse as antagonists for biological control of Rhizoctonia solani on radish. Isolates were obtained from radish rhizosphere soil, by baiting techniques, or from other laboratories. From these, seven were chosen for use in two field studies. Antagonists were applied at seeding by three methods: 1) fluid drilling with seed and pregerminated conidia suspended in gel (Natrosol$^{(R)}$ - 250HHR Hydroxyethyl Cellulose), 2) application of pregerminated conidia in a wheat bran/sand mixture (1:1, v:v; 430.6 kg/ha) to the seed furrow, and 3) adherence of ungerminated conidia to seed (1.76×10^5 conidia/seed) 24 hr prior to planting. The fluid drilling technique was superior with all seven isolates. At one location, two isolates (both T. hamatum) resulted in 9 and 32% disease as compared with 89% in the control. At the second location, results were 52 and 51% (64% control), respectively.

A372

PROLIFERATION OF TRICHODERMA AND GLIOCLADIUM FROM ALGINATE PELLETS IN NATURAL SOIL AND REDUCTION OF RHIZOCTONIA SOLANI INOCULUM. J. A. Lewis and G. C. Papavizas. USDA, ARS, Beltsville, MD 20705

Alginate pellets containing preparations of Trichoderma or Gliocladium with inert or nutrient carriers were added to soil to stimulate increase in antagonist population (cfu) and to reduce R. solani inoculum. After 1 wk, there were 10^8 cfu/g of soil amended with 1% pellets composed of wet or dry fermentation biomass (FB) of T. viride (T-1-R4) and wheat bran. The population remained high for at least 6 wk. The inert carrier, kaolin, was less effective than bran in inducing antagonist proliferation in soil. Pellets with conidia were also less effective than those prepared with FB. Pellets with FB of seven out of 11 isolates of T. viride, T. hamatum, T. harzianum, and G. virens resulted in 10^8-10^9 cfu/g of soil after 1 wk of incubation. Three preparations reduced survival of R. solani embedded in beet seed by 70% in 1 wk and seven preparations completely prevented growth of R. solani from infested beet seed into soil.

A373

REDUCTION OF RHIZOCTONIA SOLANI IN SOIL WITH FERMENTOR PREPARATIONS OF TRICHODERMA AND GLIOCLADIUM. Jean Beagle-Ristaino and G. C. Papavizas. Dept. of Botany, Univ. of MD, College Park, MD 20742; and Soilborne Diseases Laboratory, USDA, ARS, Beltsville, MD 20705.

Fermentation biomass (FB) of T. viride (T-1-R9), T. harzianum (WT-6), T. hamatum (Tri-4) and G. virens (G1-21) consisting of conidia, mycelium, chlamydospores and residual nutrients (molasses and brewers yeast) in Pyrax, were evaluated for their effect on soilborne inoculum of R. solani that causes black scurf of potato. Significant reductions in number of propagules per g of soil (50-100%) were obtained by mixing FB of the antagonists with R. solani-infested soil. Within 4 wk, R. solani was not recovered by the multiple pellet soil sampler from soils treated with FB of isolate Tri-4. Inoculum levels of the antagonists increased 1000-fold within 4 wk. R. solani sclerotia retrieved from FB-amended soils after 3 wk were heavily colonized by the antagonists and viability was reduced. Results indicate that FB of Trichoderma and Gliocladium are effective in reducing soilborne inoculum levels of R. solani.

A374

BIOLOGICAL CONTROL OF RHIZOCTONIA ROOT AND CROWN ROT OF GREENHOUSE GROWN SNAPDRAGONS. J. C. Locke and R. D. Lumsden, USDA, ARS, Beltsville, MD 20705.

Cut flower snapdragon, Antirrhinum majus, is very susceptible to root and crown rot caused by Rhizoctonia solani. The plant may be attacked at any stage of growth, but is especially vulnerable immediately following transplanting. In a raised-bench system consisting of individual treatment blocks, a composted soil-peat-perlite medium was steam pasteurized before planting eight-week-old snapdragon seedlings (cv. Panama). Of several biocontrol candidate organisms evaluated, an isolate of Gliocladium virens (G1-21) was selected as the most effective. Initial evaluation showed that if G1-21 was incorporated along with Rhizoctonia into the surface of the freshly steamed medium, there was a reduction of at least 50% in crown rot from the Rhizoctonia control treatment. In subsequent studies, incorporation of G1-21 into the steamed medium, followed by 48 hrs of incubation to allow colonization prior to introduction of the Rhizoctonia, resulted in nearly 100% control of crown rot.

A375

BACTERIZATION OF RICE PLANTS FOR SHEATH BLIGHT CONTROL T.W. Mew and A.M. Rosales, The International Rice Research Institute, Los Baños, Laguna, Philippines

Bacteria with various colony types were isolated from *Rhizoctonia solani* sclerotia, rice field flood water, diseased rice plants and healthy plants. Both fluorescent *Pseudomonas* and non-fluorescent bacteria inhibited mycelial growth of the pathogen and affected sclerotial viability. Isolate In-b-17 killed the sclerotia, while In-b-24 reduced germination to 27% after 2 weeks. The effect of bacterization of rice plants with these isolates was evaluated in the greenhouse. Soaking seeds in a bacterial suspension (10^9 cells/ml) for 10 min before sowing in *R. solani* infested soil reduced disease and sclerotia production, and promoted the growth of IR36. Subsequent planting after the 1st crop in the same soil also reduced the disease severity.

A376

MYCOHERBICIDAL ACTIVITY OF GLIOCLADIUM VIRENS BY MEANS OF VIRIDIOL PRODUCTION. C. R. Howell, and R. D. Stipanovic. ARS, USDA, National Cotton Pathology Research Laboratory, P.O. Drawer JF, College Station, TX 77841

When the biological control agent Gliocladium virens was grown on an autoclaved rice medium, it produced a phytotoxin that caused necrosis of cotton seedling radicles. The phytotoxin was identified as viridiol, a compound previously reported from G. virens. Viridiol, a close relative of the antifungal compound viridin, had little antibiotic activity against a variety of fungi and bacteria, but it was herbicidal to germinating pigweed seed when used in vitro. Because of its instability, viridiol was not an effective herbicide when it was introduced into natural soil. However, when a dried and ground preparation of G. virens rice culture was worked into pigweed infested soil above planted cotton seed, viridiol apparently was produced in sufficient quantity and duration to prevent pigweed emergence without apparent harm to emerging cotton seedlings.

A377

WORLD DISTRIBUTION OF SPORIDESMIUM SCLEROTIVORUM AND TERATOSPERMA OLIGOCLADUM. P. B. Adams and W. A. Ayers, Soilborne Diseases Laboratory, USDA, ARS, Beltsville, MD 20705

Sporidesmium sclerotivorum, a mycoparasite of sclerotia of several pathogens is known to be widespread in the continental United States. Teratosperma oligocladum, a similar mycoparasite, has a more limited geographical distribution. Scientists in various countries were requested to send soil samples from fields with a history of diseases caused by Sclerotium cepivorum or Sclerotinia species so that they could be assayed for the presence of the two mycoparasites. To date 45 samples have been assayed from nine countries by adding 2 g of Sclerotinia minor sclerotia to 100 g of soil. The added sclerotia were retrieved from the soils at 4-week intervals and placed on moist filter paper to detect the presence of the mycoparasites in the soil

samples. Sporidesmium sclerotivorum has been detected in samples from Ontario, Quebec, Manitoba, and Saskatchewan, Canada; Hokkaido, Japan; Victoria and Tasmania, Australia; Vestfold, Norway; and Helsinki, Finland. Teratosperma oligocladum was detected in one sample from Hokkaido, Japan.

A378

BIOCONTROL OF PYTHIUM AND RHIZOCTONIA DAMPING-OFF OF ZINNIA IN SOILLESS MIX BY GLIOCLADIUM VIRENS. R. D. Lumsden, J. C. Locke, J. A. Lewis, and G. C. Papavizas, USDA, ARS, Beltsville, MD 20705 and J. J. Marois, Univ. California, Davis 95616.

Of 57 isolates of potential fungal and bacterial biocontrol agents only Gliocladium virens reduced pre- and post-emergence damping-off of zinnia caused by Pythium ultimum and Rhizoctonia solani under separate conditions in nonsterile soilless potting mixes. Control of P. ultimum was effective when the antagonist was added as a 3-day-old bran culture 1 wk prior to infesting the mix with the pathogen and planting zinnia seed. The disease control was equal to a non-inoculated check. Control of disease was lost during storage of the mix at 20 and 30 C for 4 wks, but was prolonged by storage at 4 C. Suppressiveness was retained when the mix was replanted up to three consecutive times. Control of R. solani was not as effective as with P. ultimum unless the antagonist was incubated with the pathogen prior to planting zinnia seed. Various formulations of G. virens and types of soilless media affected biocontrol of these two pathogens in this system.

A379

PLASMID TRANSFORMATION OF XANTHOMONAS CAMPESTRIS V. MALVACEARUM. D. W. Gabriel, Department of Plant Pathology, Oklahoma State University, Stillwater, OK 74078.

Xanthomonas campestris pv. malvacearum (Xm), bacterial blight pathogen of cotton, is the commercial source of three restriction endonucleases. Restriction enzymes are thought to be a major barrier to transformation. There are no reports of transformation of Xm, although X. campestris pv. phaseoli and oryzae have reportedly been transformed with their own linear DNA. I isolated a small (ca. 2.5 kb), high copy number, cryptic plasmid from X. campestris pv. vesicatoria and ligated the chloramphenicol antibiotic resistance (cat) gene from pBR325 into this plasmid. The resulting 3.7 kb plasmid lacks recognition sites for Xm restriction endonucleases. I transformed Xm with this construction using a freeze-thaw method. The plasmid appears to be stably maintained in both E. coli and Xm without loss of indiginous plasmids in the transformation hosts. This plasmid is being evaluated for potential use as a broad-host range DNA transformation vector.

A380

DESCRIBED 'RACES' OF XANTHOMONAS CAMPESTRIS PV. MALVACEARUM ARE MIXTURES. G. R. Lazo and D. W. Gabriel, Department of Plant Pathology, Oklahoma State University, Stillwater, OK 74078.

Xanthomonas campestris pv. malvacearum (Xm), a bacterial pathogen of cotton, follows the gene-for-gene pattern of host specificity recognition. Single colony isolations were made from different Xm 'races' collected in the United States and Africa. These clonal isolates were analyzed both for plasmid content and race specificities on differential host lines. Twenty-five different specificities from 11 nominal Xm races were found. Plasmid DNA was extracted by an alkaline lysis procedure and subjected to restriction endonuclease digestion and agarose gel electrophoresis. One or two 50-100 kb plasmids were found in all isolates. The plasmids differed between many Xm isolates, including ones from the same nominal race. Certain similarities occurred in the restriction digest patterns for the different Xm isolates, revealing apparently homologous regions among the plasmids. Conservation of these apparently homologous regions suggests a common function. We could not correlate plasmid similarities or differences with gene-for-gene race specificity.

A381

ANALYSIS OF PHASEOLOTOXIN PRODUCTION BY PSEUDOMONAS SYRINGAE PV. PHASEOLICOLA THROUGH Tn5 MUTAGENESIS. R. C. Peet, D. K. Willis, P. B. Lindgren, and N. J. Panopoulos, Department of Plant Pathology, University of California, Berkeley, CA 94720.

Tn5 mutagenesis of Pseudomonas syringae pv. phaseolicola was conducted using the suicide vector pUW964. Transconjugants were obtained at a frequency of 3.6 x 10^{-6}/donor cell plated. 1.4% of the Km^r survivors were auxotrophs with randomly distributed nutritional requirements and Tn5 insertion fragment sizes. Using a microbiological assay, 4604 presumptive Tn5 mutants have been screened for loss of toxin production. Five independent mutants have been isolated. One mutant (NPS4336) produces 10^4-fold less toxin than the wild type (NPS3121) when assayed with E. coli C600 as an indicator but gave no detectable toxin when assayed with S. typhimurium. The inhibition zone produced by NPS3121 and NPS4336 is OTCase specific and oligopeptide permease dependent. The rate of growth of NPS3121 and NPS4336 is equal both in vitro and in planta. Unlike NPS3121, NPS4336 does not cause systemic chlorosis. The Tn5 from NPS4336 is contained within an 8.0 kb EcoR1 fragment which has been cloned in pUC8.

A382

ANALYSIS OF Vir^- HR^- Tn-INSERTION MUTANTS OF PSEUDOMONAS SYRINGAE PV. PHASEOLICOLA. P. B. Lindgren and N. J. Panopoulous, D. K. Willis, and R. C. Peet, Dept. of Plant Pathology, Univ. of Calif., Berkeley, CA 94720.

The suicide plasmid vector pUW964 was used to introduce the transposon Tn5 into Pseudomonas syringae pv. phaseolicola. The frequency of kanamycin resistant transconjucants was 2 x 10^{-8}/donor cell plated, with 2.5% of these transconjugants being auxotrophs. Southern blot analysis showed that the insertion of Tn5 was random. One thousand four-hundred seventy of these transconjugants were tested for their ability to cause disease on bean (Phaseolus vulgaris) and their ability to produce a hypersensitive response on tobacco. Three prototrophic, phaseolotoxin producing mutants (NPS 4000, 4001, and 4002)) were obtained which were avirulent on bean and failed to induce the hypersensitive reaction on tobacco. All 3 mutants were also HR^- on other heterologous host plants including tomato, cowpea, and soybean. Southern blot analysis indicate that these mutants were the result of 3 independent insertions in the same EcoRI fragment suggesting that they were affected in the same gene or transcriptional unit.

A383

ISOLATION AND CHARACTERIZATION OF THE PSEUDOMONAS SYRINGAE PV. SYRINGAE recA GENE. D. K. Willis, M. J. Hickman*, C. S. Orser, and N. J. Panopoulos. Dept. of Plant Pathology, Univ. of Calif., Berkeley, CA 94720 and *Dept. of Biology, San Francisco State Univ., San Francisco, CA 94132.

Several recombinant plasmids have been identified within a genomic cosmid library of P. s. syringae which restore UV resistance and mitomycin resistance to an E. coli recA mutant strain. One of these plasmids, designated as pCUV8, has been characterized extensively and fully complements an E. coli recA deletion strain with respect to recombination proficiency and UV resistance by quantitative analysis. The recA-like gene from P. s. syringe (Pss recA) is contained within a 15kb EcoRI restriction fragment as determined by Tn5 mutagenesis of pCUV8 and the commonality of this fragment among the plasmids initially isolated. Although it is isofunctional, the Pss recA gene shows no apparent homology to the E. coli recA gene. Preliminary Southern blot experiments with a Pss recA probe reveal pathovar-specific restriction fragment conservation among phytopathogenic pseudomonads which may be of diagnostic value.

A384

TOX^- MUTANTS OF PSEUDOMONAS SYRINGAE PV. PHASEOLICOLA BY TRANSPOSON TN5 MUTAGENESIS. Charles J. Romeo, Department of Biochemistry and Biophysics and Suresh S. Patil, Department of Plant Pathology, University of Hawaii, Honolulu, Hawaii 96822

P.s. pv. phaseolicola PDDCC 4612 was mutagenized with Tn5 (Km^r) using a conjugative suicide plasmid, pJB4JI (pPH1JI::Mu::Tn5) in E. coli 1830. Of 1600 independent mutants, selected on minimal medium with Km and screened for phaseolotoxin in a microbial bioassay, three showed a Tox^- phenotype. Two mutants exhibited low levels of phaseolotoxin and a third mutant was completely Tox^- in the bioassay. Growth of Tox^- mutants and production of watersoaking in a susceptible host cultivar were similar to that of 4612 wild type. Chlorosis was caused by the mutants producing low levels of toxin, but not by the mutant that was completely Tox^-. Each mutant contained a single Tn5 insertion as revealed by Southern blot analysis of total genomic DNA digested with EcoR1. The sizes of the EcoR1 fragments containing Tn5 were approximately 40, 14, and 7 kilobases. In these mutants plasmid DNA did not contain Tn5 insertions, therefore, Tn5 was located chromosomally.

A385

THE PLASMID pRD1 INTEGRATES INTO THE CHROMOSOME OF PSEUDOMONAS SYRINGAE. J.R. Vincent and D.W. Fulbright, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

pRD1 is a 150 kb plasmid that was constructed from the IncP-1 antibiotic resistance plasmid RP4 and contains the genes for histidine biosynthesis and nitrogen fixation from Klebsiella pneumoniae. We previously determined in cultures of Pseudomonas

syringae strain PSSD220 the conditions under which pRD1 disappears as an extrachromosomal element, while maintaining the plasmid-determined phenotype of histidine prototrophy and kanamycin resistance. Total DNA from these cultures of PSSD220 was isolated and digested to completion with restriction endonucleases. After electrophoresis in 0.5% agarose gels, the DNA was transferred to nitrocellulose and hybridized to ^{32}P-pRD1 by the method of Southern. By comparing the restriction pattern of isolated pRD1 to the restriction pattern of pRD1 found within PSSD220 total DNA, it was determined that pRD1 was integrated into the chromosome of P. syringe PSSD220.

A386

CHARACTERIZATION OF INDIGENOUS PLASMIDS IN PSEUDOMONAS SYRINGAE PV. TOMATO. C. L. Bender and D. A. Cooksey, Department of Plant Pathology, University of California, Riverside, CA 92521.

Twenty strains of Pseudomonas syringae pv. tomato isolated from tomato plants in southern California and Mexico were screened for the presence of plasmid DNA. Four size classes of plasmids were detected: (A) 96-102 kb, (B) 73-81 kb, (C) 66 kb, and (D) 43-47 kb. Ten strains contained two plasmids in size classes A and B, nine strains had three plasmids in classes A, B, and D, and one strain contained plasmids in all four size classes. Plasmids in the same size classes had almost identical restriction patterns when digested with EcoRI, HindIII, and BamHI. These results suggest that different P. syringae pv. tomato strains contain at least two resident plasmids which are highly homologous. The large class A plasmid was cured from a strain with two plasmids with no loss of virulence or toxigenicity. Curing experiments are being conducted to determine the possible role of the remaining class B plasmid in pathogenicity.

A387

CLONING OF AN ENTIRE SMALL MOLECULAR WEIGHT PLASMID OF PSEUDOMONAS SYRINGAE PV. GLYCINEA IN pBR329 AND pRK404. Joe W. Willis and J. V. Leary, Department of Plant Pathology, University of California, Riverside, CA 92521.

A 7.3 kb plasmid (pPG1) was found in coronatine-producing strains but not in non-coronatine strains of Pseudomonas syringae pv glycinea, in addition to other larger plasmids. Purification of pPG1 by electroelution, followed by restriction enzyme analysis revealed that several enzymes had no recognition sites in pPG1 and others had only one or two restriction sites. Using EcoRI, for which there were no recognition sites, it was possible to purify large amounts of pPG1 on $CsCl_2$-ethidium bromide density gradients by pre-treatment. Use of HindIII, which has a single recognition site in pPG1, made it possible to clone the entire plasmid into pBR329 and pRK404. The hybrid plasmids were transformed into E. coli and conjugated into non-toxigenic P. glycinea strains. The transformants and transconjugates are being tested for coronatine production.

A388

CONSTRUCTION OF A GENOMIC LIBRARY OF PSEUDOMONAS SYRINGAE PV. GLYCINEA RACE 8 AND IDENTIFICATION OF A GENE CONFERRING RESISTANCE TO TRIMETHOPRIM. J. V. Leary and D. Trollinger, Department of Plant Pathology, University of California, Riverside, CA 92521.

Pseudomonas syringae pv. glycinea Race 8 strain PgB3 is naturally resistant to trimethoprim (Tm) at concentrations up to 500 µg/ml. A genomic library of total PgB3 DNA was constructed by ligating EcoRI-restricted DNA into EcoRI-restricted cosmid vector pLAFR1, packaging the religated DNA in vitro into bacteriophage lambda, and transfecting E. coli HB101 cells. Of 960 clones selected for resistance to tetracycline conferred by pLAFR1, six were resistant to trimethoprim at 500 µg/ml. An insert into pLAFR of about 8.5 kb was shown to be consistently present in the trimethoprim-resistant clones. The nature of the DNA-insert responsible for conferring Tm-resistance, whether chromosomal or plasmid, is being determined.

A389

FINE STRUCTURE MAPPING OF THE rsi LOCUS IN PSEUDOMONAS SYRINGAE PV. GLYCINEA. B. Staskawicz, D. Dahlbeck and C. Napoli, Dept. of Plant Pathology, University of California, Berkeley, CA 94720.

We have recently identified and cloned an avirulence gene from race 6 of P. s. glycinea that determines race specific incompatibility (rsi) on the appropriate soybean cultivars. The rsi locus was initially cloned on a 27.2kb fragment of DNA from race 6 of P s. glycinea into the cosmid vector pLAFR1. To begin a detailed molecular analysis of this locus we have subcloned a 3kb AccI fragment into the plasmid pRK404 that maintains avirulence activity. A detailed restriction enzyme map of this fragment has been generated and further subcloning has delineated the region important for activity. Saturation Tn5 mutagenesis of this locus is currently being carried out to determine the exact location of this gene. In addition, chimeric gene fusions with β-galactosidase have been constructed and these plasmids have been introduced into Escherichia coli X1488 minicells to determine the primary gene product.

A390

CLONING OF BACTERIOPHAGE pEa1(h) GENES IN E. COLI. J.S. Hartung, D.W. Fulbright and E.J. Klos, Department of Botany and Plant Pathology, Michigan State Univ., East Lansing, MI 48824-1312

When Erwinia amylovora is infected by phage pEa1(h) a capsular depolymerase (CDP) is produced which decapsulates the bacteria. These cells may be more sensitive to antibiotics than capsulated bacteria. In an attempt to clone the CDP gene of pEa1(h), the phage genome was partially restricted with endonuclease Sau3A and ligated into the BamHl site of plasmid pUC8. This plasmid allows expression of cloned DNA under lac operon control in E. coli strans JM83 (constitutive) and JM105 (inducible). Several putative CDP producing JM83 clones were selected by overlaying chloroform vapor killed replica plates with E.amylovora. These clones are surrounded by haloes when overlayed with capsule$^+$ E. amylovora but not when overlayed with a capsule$^-$ strain. The halo$^+$ phenotype is unstable in JM83 making a definitive CDP assay difficult. Cloning into strain JM105 is underway in order to improve the stability of the recombinant clones.

A391

AN INSERTION SEQUENCE FROM AGROBACTERIUM TUMEFACIENS STRAIN C58 WITH HOMOLOGY TO WILD-TYPE TI-PLASMID AND CHROMOSOMAL SEQUENCES. D. A. Cooksey, Department of Plant Pathology, University of California, Riverside, CA 92521.

A spontaneous agrocin-resistant mutant (C58R124) of the nopaline strain C58 of Agrobacterium tumefaciens was shown by restriction analysis to have a 1.2 kb insertion in the agrocin sensitivity region of the Ti-plasmid. A 3.0 kb HindIII fragment containing this insertion was cloned in the vector pRK404 and used to probe the Ti-plasmid and chromosome of the wild-type strain C58. The probe hybridized to a Ti-plasmid sequence outside of the agrocin sensitivity region of pTiC58 and to a C58 chromosomal sequence. The probe also hybridized to a sequence in the chromosome of A. tumefaciens strain K24. These data suggest that an insertion sequence with copies in the chromosome and Ti-plasmid of the nopaline strain C58 caused the mutation to agrocin resistance in C58R124. The relationship of this sequence to other known IS elements in octopine strains and to possible IS elements in other nopaline strains is being investigated.

A392

PROPERTIES OF ARSENIC-RESISTANCE PLASMIDS FROM CORYNEBACTERIUM FLACCUMFACIENS SSP. OORTII. **C. Hendrick and A.K. Vidaver, Dept. of Pl. Path., Univ. of Nebraska, Lincoln, NE 68583-0722.**

A 46 Mdal conjugative plasmid (pD8101), from Corynebacterium flaccumfaciens ssp. oortii (Cfo) strain C0101, codes for resistance to arsenate, arsenite, and antimony. This plasmid was transferred by conjugation to other members of the C. flaccumfaciens group and recently to members of the C. michiganense group. pD8101-mediated resistance to arsenate was inducible by subinhibitory levels of arsenate, arsenite, and antimony, while arsenite and antimony resistance were constitutive. Southern hybridization experiments showed no homology between pD8101 and six plasmids (three from Escherichia coli and three from Staphylococcus aureus) that also code for resistance to arsenate, arsenite, and antimony. A second conjugative plasmid (pD8499) from Cfo (strain C03499) also confers arsenite, arsenate, and antimony resistance. pD8499 is slightly smaller than pD8101 and has different restriction fragment patterns; however, Southern blots showed that the two plasmids have homologous sequences.

A393

INSERTIONAL INACTIVATION OF A CLONED ERWINIA CHRYSANTHEMI PECTATE LYASE GENE. David L. Roeder and Alan Collmer. Department of Botany, University of Maryland, College Park, MD 20742.

An Erwinia chrysanthemi pectate lyase isozyme (pI 8.3) gene that had been cloned into Escherichia coli on plasmid vector pBR322 was mutagenized by insertion of a kanamycin resistance gene derived from plasmid pUC-4K. The recombinant plasmid pCSR1, which contains a 6.7 kb E. chrysanthemi DNA insert, was partially digested with restriction endonuclease Sau3A. Linear

plasmid DNA fragments of approximately full length were then ligated with the 1.4 kb BamHl fragment of pUC-4K which encodes kanamycin resistance. Of 140 Amp^R Kan^R E. coli HB101 transformants examined, 25 failed to pit pectate semi-soft agar. Restriction analysis of the recombinant plasmids revealed the presence of deletions of various size. The deletion/insertions affecting pectate lyase actitity were mapped to a 3.9 kb EcoRV fragment of pCSRl. These pCSRl derivatives can now be used for site-directed mutagenesis of the pectate lyase isozyme structural gene in the E. chrysanthemi chromosome.

A394

THE INFLUENCE OF DENSITY AND PATCHINESS OF INOCULUM ON THE EPIDEMIOLOGY OF TOBACCO BLACK SHANK. D.M. Ferrin and D.J. Mitchell, Dept. of Plant Pathology, Univ. of Florida, Gainesville, FL 32611.

The influence of density and patchiness of inoculum of Phytophthora parasitica var. nicotianae on disease development in Hicks tobacco was assessed in 1983. Inoculum densities in the upper 10 cm of soil at planting ranged from 0.10 to 1.18 propagules/g and generally were fit best by the negative binomial distribution. Patchiness of inoculum was indicated by values of 1.49 to 7.54 for Lloyd's index of patchiness where values greater than 1.0 denote nonrandomness. A highly significant negative correlation (r=-0.997) between final disease proportion (Ymax) and the index of patchiness was observed. Transformation of disease proportion (Y) with ln[Y/(Ymax-Y)] as compared with ln[Y/(1-Y)] increased R^2 values when disease progress approached asymptotes of less than 1.0. Aggregation of inoculum imposed an upper limit on disease development, which may be mistaken for a reduction in the rate of disease increase if the appropriate transformation is not used.

A395

INFLUENCE OF TEMPERATURE AND WETNESS DURATION ON INFECTION OF STRAWBERRY FRUIT BY PHYTOPHTHORA CACTORUM. G. G. Grove, L. V. Madden, M. A. Ellis, and A. F. Schmitthenner, Department of Plant Pathology, The Ohio State University/OARDC, Wooster 44691.

Strawberry fruits (cv. Midway) were inoculated with a sporangial suspension (400 sporangia/ml) of Phytophthora cactorum to determine the effects of wetness duration and temperature on the level of infection. Infection increased with increased wetness duration (0-5 hr) at all temperatures tested (6-30 C). For each wetness duration, infection increased up to the optimum temperature and then declined. The optimum temperature for infection was 21 C. At temperatures between 17 and 25 C, 1-5 hr of wetness resulted in high (>80%) levels of infection. A multiple-regression logistic-model accurately described infection as a function of wetness duration and temperature. The model was validated under natural field conditions.

A396

COMPARISON OF ANNUAL PHYMATOTRICHUM ROOT ROT EPIDEMICS IN COTTON FOR THE YEARS 1965-1982. M. J. Jeger and S. D. Lyda, Department of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843.

Epidemics of Phymatotrichum root rot in cotton were monitored in a naturally infested field at the Blacklands Research Center, Temple, Texas, during the years 1965-1982. Cultivar, sowing date, and cultural practice varied according to season. Epidemics were readily classified according to timing, level and pattern of disease development. In most years disease development was smoothly sigmoid, and there was a direct relationship between the Julian day of epidemic commencement and the asymptotic level of disease (range 4-100%). In two years, however, there were atypical increases of disease late in the growing season. The severity of root rot epidemics was related directly to cumulative precipitation (range 36-101 cm) during the season, and inversely to mean maximum temperatures (range 34-39 C) during successive 10-day periods of the epidemic.

A397

RELATIONSHIP BETWEEN SCLEROTIAL POPULATIONS OF SCLEROTINIA MINOR AND THE INCIDENCE OF LETTUCE DROP. H. R. Dillard and R. G. Grogan, Dept. of Plant Pathology, Univ. of California, Davis, CA 95616.

At planting time, inoculum densities of sclerotia of S. minor ranged from 1.66 to 11.35 sclerotia/100-cc soil in 15 naturally infested fields near Salinas, California. In all but 4 fields, variance-to-mean ratios were significantly greater than one, indicating a clustering of inoculum. The position of each obviously diseased plant was mapped at weekly intervals in 5 of the 15 plots. At harvest, there was a random distribution of healthy and infected plants, and no significant plant-to-plant spread had occurred. Disease progress curves constructed for all 15 plots showed nil disease for the first 30 days after planting and a rapid increase during the last 10 days prior to harvest. Percent disease incidence at harvest was significantly correlated (R^2 = 0.81) with the mean number of sclerotia per 100 cc of soil at planting, and more so with the percentage of soil samples with a density of $\geq$7 sclerotia/100 cc soil (R^2 = 0.88).

A398

EFFECT OF TEMPERATURE ON SURVIVAL OF THE ALFALFA WILT PATHOGEN VERTICILLIUM ALBO-ATRUM IN SOIL. P. K. Basu, Agriculture Canada, Research Station, Ottawa, Ontario, K1A OC6.

An isolate of Verticillium albo-atrum from alfalfa was tested for its viability on nylon threads and in alfalfa stems buried in sterile and non-sterile soil plates for 6 and 12 months (mo), respectively, at temperatures (temp) ranging from -5 to 35 C. On threads the pathogen survived 6 mo in sterile soil at -5 to 25 C, and 5 mo in non-sterile soil at -5 to 5 C. In alfalfa stems it survived 12 mo at low soil temp (-5 to 5 C) only; but at higher temp the longevity of the pathogen was reduced to 11, 8, and 7 mo at 15, 30 and 35 C, respectively. However, in alfalfa stems placed in plates without soil the pathogen survived at least 12 mo at all temp tested (-5 to 35 C). Dark resting mycelia of the fungus were found consistently in alfalfa tissues, indicating strongly that these served as survival structures.

A399

COMPARATIVE EPIDEMIOLOGY AND CONTROL OF LETTUCE DROP CAUSED BY SCLEROTINIA MINOR and S. SCLEROTIORUM. C. L. Patterson and R. G. Grogan, Department of Plant Pathology University of California, Davis, CA 95616.

The epidemiology of S. minor and S. sclerotiorum on lettuce is distinctly different. Sclerotial inoculum was decreased and the disease eliminated after 3 consecutive years of removing lettuce plants infected by S. minor from the field. Significant reduction in drop caused by S. minor was achieved by one application of DCNA, Iprodione, or Vinclozolin immediately after thinning. In contrast, S. sclerotiorum was controlled only by fungicides applied during the rosette stage of growth prior to senescence of lower leaves. Thus, procedures that controlled S. minor failed to control S. sclerotiorum and vice versa. Apothecia of S. sclerotiorum were found in late fall, winter, and spring in the San Joaquin Valley during prolonged wet periods; ascospores were trapped on acidified potato dextrose agar in lettuce fields when apothecia were not observed. Ascospores of S. minor were never trapped even in fields where it was causing disease.

A400

FREQUENCY DISTRIBUTION ANALYSIS OF LETTUCE DROP, CAUSED BY SCLEROTINIA MINOR, AS A FUNCTION OF QUADRAT SIZE. J. J. Marois and P. B. Adams, Department of Plant Pathology, Univ. of California, Davis, CA 95616, and Soilborne Diseases Laboratory, USDA, ARS, Beltsville, MD 20705.

The location of plants affected by lettuce drop was mapped in six lettuce fields. Frequency distributions of diseased plants were developed for each field using five quadrat sizes. The derived distributions were analyzed by the Chi Square test for their goodness of fit to eight standard frequency distribution formulas. Quadrat size affected the type of distribution best fit, the goodness of fit, and the parameters of formulas for the frequency distributions. For example, in the same field, data derived from quadrat dimensions of 0.9 X 0.9 m fit the Poisson distribution best (P = 0.88), quadrat dimensions of 0.3 X 3.0 m fit the Poisson Binomial distribution best (P = 0.90), quadrat dimensions of 0.6 X 3.0 m fit the Neyman Type A distribution best (P = 0.97), and quadrat dimensions of 0.9 X 3.0 m fit the Positive Binomial and Poisson With Zeros distributions equally (P = 0.69).

A401

THE POTENTIAL USE OF AN AERIAL REMOTELY PILOTED VEHICLE (DRONE) FOR SPORE COLLECTION AT VARIOUS HEIGHTS ABOVE THE CROP CANOPY. T. R. Gottwald and W. L. Tedders, USDA-ARS, SE Fruit & Tree Nut Res. Lab., P. O. Box 87, Byron, GA 31008.

A radio-controlled, remotely-piloted vehicle was constructed for several agricultural applications. The drone is a bi-wing

aircraft (2.44 m wingspan x 2.04 m fuselage length) which has been fitted with two wing-pod spore traps moved midway between fuselage and wingtip on the top of the lower wing. Traps are constructed of a servo modified to rotate 180° which holds a trapping drum (5.5 cm dia x 1.4 cm height). The edge of the drum is affixed with a cellophane tape coated with a mixture of polyvinyl alcohol, petroleum jelly, and paraffin softened with toluene. The drum can be rotated to multiple positions in front of a 1 mm wide slit orifice. Numerous collections at various heights above a field crop or orchard canopy can be achieved in a relatively short time.

A402

PATTERNS OF ASCOSPORE DISCHARGE BY VENTURIA INAEQUALIS DURING RAIN-INITIATED WET PERIODS. W. E. MacHardy and D. M. Gadoury. Dept. of Botany and Plant Pathology, University of New Hampshire, Durham, 03824.

Discharge of ascospores of the apple scab pathogen, Venturia inaequalis, was monitored with a Burkard volumetric spore trap in an orchard with a large overwintering population of the pathogen. Weather recording instruments in the orchard provided hourly records of temperature, amount and intensity of rainfall, relative humidity, and leaf wetness. Nearly all spores were trapped between 0800 and 2000 hrs: 97% in 1981, 98% in 1982, and 99% in 1983. Of all spores trapped, 31%, 47%, and 20% were trapped between 0800-1200, 1200-1600, and 1600-2000 hrs, respectively. Spores were not trapped during intervals when leaves were wetted only by dew. When rain began during the night, few spores were discharged before sunrise. The extremely low density of spores in the air between 2000 and 0800 hrs has significant implications for determining Mills' infection periods and for scheduling post-infection fungicide sprays.

A403

METEOROLOGICAL ASPECTS OF THE SPREAD AND DEVELOPMENT OF BLUE MOLD ON TOBACCO IN NORTH CAROLINA. J. M. Davis and C. E. Main, Department of Plant Pathology, North Carolina State University, Box 7616, Raleigh, NC 27695-7616.

Blue mold, a highly weather sensitive disease caused by Peronospora tabacina, occurred in the major tobacco production areas of North Carolina in 1980. Analysis of blue mold first occurrence dates and temporal and spatial properties of temperature and precipitation indicated that the epidemic continued to spread and develop despite temperatures outside the range previously considered favorable for the disease. Availability of moisture on the tobacco leaves for spore germination appeared to be a predominant factor in disease development in all parts of the state. Meteorological trajectory analysis indicated many days in April, May and June 1980 when conditions were favorable for spore transport over North Carolina tobacco fields from infected fields to the south which acted as possible inoculum source areas. Taking latent periods into account, certain trajectory dates were selected as representing the most probable periods of spore transport.

A404

ASSOCIATION OF PHYTOPHTHORA SYRINGAE WITH PRUNING WOUND CANKERS IN ALMOND. R. M. Bostock and M. A. Doster. Department of Plant Pathology, University of California, Davis, CA 95616

Cankers with profuse gumming associated with pruning wounds in almond trees were frequently observed in California orchards during the winters and springs of 1983 and 1984. Phytophthora syringae was isolated from these cankers. Pruning wounds inoculated in February with mycelial disks from cultures of an almond isolate of P. syringae produced large gumming cankers resembling those of naturally affected trees. Likewise, excised branch pieces inoculated in the laboratory indicated that P. syringae is highly virulent between 2° and 20°C. In an initial survey, greater than 99% of the cankers in an orchard were associated with pruning wounds or injuries created during pruning in late fall and winter months. Although at this time the involvement of other organisms in addition to P. syringae cannot be excluded, the similar symptoms observed in other orchards, the absence of other known pathogens in diseased tissue, and the seasonal occurrence of this disease strongly suggest that P. syringae is a common causal agent of pruning wound cankers in almond.

A405

VEGETATIVE COMPATIBILITY GROUPS AND THE EFFECT OF WATER POTENTIAL ON THE GROWTH OF CYTOSPORA KUNZEI. Tyre J. Proffer and John H. Hart, Dept. of Botany and Plant Pathology, Michigan State University, E. Lansing, MI 48824-1312

Cytospora kunzei isolates were collected from 46 resinous branch cankers on 32 spruce (Picea spp.) located in six separate locations within Michigan. Vegetative compatibility (vc) pairings of 108 isolates on potato dextrose agar indicated that there are at least 24 vc-groups of C. kunzei in Michigan. Isolates from a single canker were always contained within the same vc-group. Separate cankers on a single tree sometimes contained different vc-groups. The effect of water potential on the in vitro growth of C. kunzei was examined by varying the water potential of the medium by the addition of various solutes; KCl, NaCl, sucrose, mannitol, and polyethylene glycol. Fungal growth was measured as increases in colony diameter or weight. At 26°C the maximum growth of C. kunzei occurred at water potentials ranging between -2.6 bars for NaCl to -15 bars for sucrose. The growth response to water potential was dependent on the osmoticum utilized.

A406

CRYPTOSPHAERIA POPULINA ASSOCIATED WITH MORTALITY IN NEW YORK STATE ASPEN PLANTATIONS. C. M. Catranis and P. D. Manion, State University of New York, College of Environmental Science and Forestry, Syracuse, NY 13210

Cryptosphaeria populina has recently been associated with extensive mortality of 16 to 22-year old Populus tremuloides in plantation plantings at the Tully Genetics Field Station. Three intensive surveys of 2330 trees, representing seven replicated experiments, show up to 22% mortality associated with C. populina and Hypoxylon mammatum. C. populina accounted for up to 13.7% mortality. C. populina and H. mammatum occurred together on 2 to 3% of the trees and H. mammatum occurred alone on the remaining dead trees. The 7.5 cm dbh class trees had the highest incidence of C. populina. Up to a 3% increase in mortality occurred over a 6-month period. Mortality associated with C. populina was higher in the plantations than in naturally occurring wild clones. Variations in disease incidence occurred among families of trees demonstrating a potential for genetic selection for resistance.

A407

CHARACTERIZATION OF SEPTORIA MUSIVA ISOLATES FROM ONTARIO AND THE UNITED STATES. Linda J. Spielman and M. Hubbes, Faculty of Forestry, University of Toronto, Toronto, Ont., Canada M5S 1A1

Septoria canker of hybrid poplars, caused by S. musiva, is less severe in Ontario than in the United States. We have investigated the hypothesis that the lower disease level in Ontario is due to genetic differences among the pathogen populations. Isolates from Mississippi, Iowa, Wisconsin, and Michigan did not differ from Ontario isolates in morphology on natural substrates or in cultural characteristics. Optimal growth temperatures ranged from 17° to 29° C, but did not correlate with region of origin. Isozyme patterns for esterase, hexokinase, and other enzymes were specific to locality, and isolates from the same region were more similar than those from different regions. We conclude that, although geographically separated populations of S. musiva are similar physiologically, they do differ genetically, and that this may explain the observed differences in disease severity.

A408

VARIATION OF POPULUS TREMULOIDES REACTION TO TOXIC CULTURE FILTRATES OF HYPOXYLON MAMMATUM. D. H. Griffin and P. D. Manion. College of Environmental Science and Forestry, Syracuse, NY 13210.

Nine clones of aspen were tested for sensitivity to toxin preparations from 5 single-spore isolates of H. mammatum with a leaf puncture bioassay. Toxic filtrates from submerged, shake cultures in defined medium were compared to extracts from oat grain cultures. The responses of the clones to these were similar. Regression analysis of lesion diameter with dilution of the toxins classified the clones into three sensitivity groups. Two groups with the greatest sensitivity were more resistant to cankering and the group with the lowest sensitivity contained both susceptible and resistant clones. PCA analysis of the clones clearly identified the susceptible clones from the resistant. Toxin reactions showed negative correlations to canker lengths and positive correlations to callus formation when the isolates were inoculated into branches of the clones. The results suggest that the toxic reactions were more related to a hypersensitivity response than to a host-killing, fungal invasion response.

A409

HYPOXYLON CANKER OF Salix spp. D.W. French and J. Juzwik. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

Large, diffuse cankers with stromata and perithecia of Hypoxylon mammatum were found on several stems of Salix daphnoides (S.d.) in two locations and a diamond-shaped canker with stromata and peritheca was observed on an unidentified Salix sp. (S.u.) in St. Paul, MN. Ascospores from (S.u.) averaged 21.0 x 8.9 um comparable to previously reported Salix collections; colony characteristics also were similar on malt agar. Ascospores from isolates on (S.d). averaged 28.2 x 11.1 um, and those from H. mammatum on Populus tremuloides were 26.2 x 10.8 um; also colonies were similar on malt agar. Inoculation of P. tremuloides with mycelium from the isolate obtained from (S.d.) resulted in typical cankers with synnemata within 2 years. This is the first report that isolates of H. mammatum from Salix can incite cankers on P. tremuloides similar to those caused by isolates from aspen. The host genotype may account for the susceptibility of S. daphnoides.

A410

Similarities in physiological characters between Endothia Eugeniae and Cryphonectria cubensis, causal agents of cankers in clove and eucalyptus, respectively. A.C. Alfenas[1], C.S. Hodges[2], and R. Jeng[3]. [1]Universidade Federal de Viçosa-Brasil, [2]U.S.D.A. Forest Service - Hawaii and [3]University of Toronto.

In 1981, C.S. Hodges observed a fungus on clove (Caryophyllus aromaticus L.) in Indonesia. Recently the same author collected the same fungus on clove in Brazil. This fungus has been identified as Endothia eugeniae. However, the morphological characters of this fungus resembled those of Cryphonectria cubensis, the causal agent of the eucalyptus canker. Cultures isolated from clove and eucalyptus were equally pathogenic to Eucalyptus grandis. The electrophorectic analysis of soluble proteins and the isoenzymes esterase (EST), glutamate-oxalo-acetate-transaminase (GOT), malate dehydrogenase (MDH), and peroxidase (PO) confirmed the similarities of Endothia eugeniae to Cryphonectria cubensis.

A411

DISSEMINATIVE FITNESS OF ENDOTHIA PARASITICA CONTAINING DIFFERENT AGENTS FOR CYTOPLASMIC HYPOVIRULENCE. J. S. Russin and L. Shain, Dept. of Plant Pathology, Univ. of Kentucky, Lexington 40546.

Three different agents for cytoplasmic hypovirulence (CH) were transferred individually to a virulent (V) isolate (EP 155) of Endothia parasitica. These isolates were obtained from J. E. Elliston, Conn. Agr. Exp. Sta. The CH agent HI_2 obtained from an Italian isolate of E. parasitica had little effect on the ability of EP 155 to sporulate asexually or to cause cankers on American chestnut. CH agent HM_2 from a Michigan isolate reduced these parameters by 60% and 80%, respectively whereas CH agent HT_2 from a Tennessee isolate reduced both parameters almost totally. Agent HT_2 was markedly less efficient than HI_2 or HM_2 at conversion of mycelium and stromata in EP 155 cankers. Agent HT_2 also was transmitted through a much lower percentage of conidia than were HI_2 or HM_2. This may not be a barrier to spread of hypovirulence, however, as very few CH conidia in droplets of conidial suspensions were required for conversion of resultant colonies to CH.

A412

RELATIVE SUSCEPTIBILITY OF FOUR HONEYLOCUST CULTIVARS TO THYRONECTRIA AUSTRO-AMERICANA. W. R. Jacobi, Department of Botany and Plant Pathology, Colorado State University, Fort Collins, CO 80523.

Management of Thyronectria canker of honeylocusts (Gleditisia triacanthos L.) induced by Thyronectria austro-americana (Spegz.) Seeler apparently requires manipulation of various environmental factors and utilizing proper tree maintenance. Since the predisposing environmental stresses are not easily identified or corrected, host resistance could play a role in disease prevention. Four honeylocust cultivars, Imperial, Skyline, Sunburst and Thornless were assessed for resistance. Fifty, 3 yr old trees of each cultivar were inoculated Oct. 15, 1983 and fifty on April 15, 1984 to assess seasonal response. Half of the trees were stressed by deep girdling 75% of the tree base two weeks before inoculation. Two diameter measurements along with observations on symptoms, signs, and indications of wound healing were recorded monthly. The results of this study will be reported and implications discussed.

A413

EFFECT OF STEM WOUND AGE ON INFECTION AND CANKER DEVELOPMENT IN HONEYLOCUST SEEDLINGS BY THYRONECTRIA AUSTRO-AMERICANA. Jerry W. Riffle and Glenn W. Peterson. Rocky Mountain Forest and Range Expt. St., Forestry Sciences Laboratory, Univ. of Nebr., Lincoln, 68583.

Gleditsia triacanthos seedlings (20-mo-old) were wounded to the sapwood at 21,14,7, and 0 days before inoculation. Seedlings with wounds of each age were inoculated simultaneously with mycelial discs from one isolate of Thyronectria austro-americana; wounded control seedlings received discs of PDA. Mean stem diameter at inoculation sites was 5.5mm. Seedlings were kept for 4 weeks in growth chambers at 28 C with 16-hr photoperiod. At 4 weeks, 6,10,62, and 100% of the fungus inoculated seedlings with wounds of age 21,14,7, and 0 days were infected, respectively. Mean lengths of stem cankers were 22,25,16, and 44mm, and mean percent of circumference of stems girdled was 58,64,52, and 91 on infected seedlings with wounds of age 21,14,7, and 0 days, respectively. Thus, T. austro-americana readily infects fresh bark wounds on honeylocust seedlings.

A414

EFFECT OF TEMPERATURE ON INFECTION AND CANKER DEVELOPMENT IN HONEYLOCUST SEEDLINGS BY THYRONECTRIA AUSTRO-AMERICANA. Jerry W. Riffle and Glenn W. Peterson. Rocky Mountain Forest and Range Expt. Stn., Forestry Sciences Laboratory, Univ. Nebr., Lincoln, 68583.

Gleditsia triacanthos seedlings (20-week-old) were wounded to the sapwood and inoculated with mycelial discs of three isolates of Thyronectria austro-americana (200 seedlings per isolate). Fifty seedlings inoculated with each isolate and 50 control seedlings were kept in each of four growth chambers at 16,20,24, and 28 C for 6 weeks with 16-hr photoperiod. Mean stem diameter at inoculation sites was 2.9mm. All inoculated seedlings became infected, and mean number of days for their foliage to wilt was 40,23,19, and 11 at 16,20,24, and 28 C, respectively. All infected seedlings developed cankers; their mean lengths below inoculation sites were 17,24,27, and 26mm at 16,20,24, and 28 C, respectively. These results reveal that disease development is more rapid at higher temperatures (24,28 C).

A415

OBSERVATIONS ON FIELD GROWN PINES INOCULATED WITH THE PINE WOOD NEMATODE. M.J. Wingfield, P.J. Bedker, and R.A. Blanchette. Dept. of Plant Pathology, Univ. of Minn., St. Paul, MN 55108.

Branches on 20 Pinus sylvestris, 20 P. resinosa, and 13 P. banksiana, approximately 11-yr-old trees, were inoculated with 10,000 Bursaphelenchus xylophilus in June 1983. Branch wounds that exposed the cambium were inoculated with nematodes grown on cultures of Botrytis cinerea. Controls were treated similarly but were inoculated only with a slurry from nematode free cultures of B. cinerea. Fourteen wks after inoculation 25% and 35% of the branches were dead distal to the inoculation point on P. banksiana and P. sylvestris, respectively. No branches had died on P. resinosa. Although relatively few nematodes (5 to 6 per gm of wood) were found in wood samples taken from branches inoculated with B. xylophilus, branch death was attributed to girdling as a result of nematode inoculation. No nematodes were found in the main stems of inoculated trees or any of the controls. Rapid death of pines after nematode inoculation, as observed in Japan, did not occur.

A416

POPULATIONS OF BURSAPHELENCHUS XYLOPHILUS IN SCOTS PINES INOCULATED WITH B. XYLOPHILUS AND CERATOCYSTIS IPS. G. J. Hunt, J. R. Bloom, and D. D. Davis, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

In recent years, the pine wood nematode, Bursaphelenchus xylophilus, frequently has been isolated from wilted Scots pines (Pinus sylvestris) in the United States. A blue-stain fungus, Ceratocystis ips, also has been associated with the pine wilt syndrome. The terminal shoots of 2-year-old Scots pines were inoculated on 2-4 July 1983 with 500 or 5000 B. xylophilus. In addition, some plants were inoculated with C. ips on 21-22 July. Thirty-two to 35 days after nematode inoculations were performed, nematodes were extracted by incubating cut stem sections for 24 hr at 25 C in 0.75% NaCl solution. Seedlings inoculated with C. ips contained significantly higher ($p<0.01$) densities of nematodes/g dry stem than seedlings which were not inoculated with C. ips. Nematode inoculum level and stem diameter were significant factors ($p<0.01$) influencing final nematode density.

A416a

INDUCED VIRULENT MUTATION IN *MELAMPSORA MEDUSAE*. C.S.Prakash and W.A.Heather, Dept. of Forestry, The Aust. Nation. Univ., GPO Box 4, ACT 2601, Australia.

Five single uredia were selected from some 36 developed when

uredospores of Race 5A of *M. medusae* were gamma irradiated (10, 20 or 69 kR) and placed separately on *Populus deltoides* cv.T-173 on which Race 5A is avirulent. The infection type (scale 0-4) produced by uredospores of the mutants on replicate leaf disks of twelve cultivars was higher than (seven cultivars), equal to (four cultivars) or pronouncedly less than (type 4 to type 1 or 2 on cv.7-2) that of Race 5A. The latter case suggests the loss in the mutants of a gene for virulence on cv.7-2. Although the five mutants produced uniform infection type within a particular cultivar, uredial numbers varied significantly between mutants on a particular cult-ivar and such differences in aggressiveness were not related to the dosage levels of irradiation applied.

A417

DYNAMICS OF CATION RELEASE FROM DELAWARE SOILS SUBJECTED TO SIMULATED ACID RAIN. D. L. Sparks and C. R. Curtis, Department of Plant Science, University of Delaware, Newark, DE 19716 and Department of Plant Pathology, The Ohio State University, Columbus, OH 43210.

Cations may accumulate in plant tissues to phytotoxic levels. This study was initiated to determine the effect of simulated acid rain (SAR) on the release of selected soil cations. Cation release was investigated using three Delaware soil types having low buffering capacity. The release kinetics were evaluated by leaching the soils with SAR at pH 2.5, 3.4, 4.8 and 5.6. For all soils and pH's, the cations were released in the following order: $Ca^{2+} > K^{+} > Mg^{2+} > Na^{+}$. As the SAR pH decreased, the total quantity of each element increased. A rapid initial release rate was followed by lower rates for each element. The total Al^{3+} released from the soils ranged from 0.74 to 25 μM at pH 2.5 and 5.6, respectively. The amount of Si^{4+} and heavy metal release was low.

A418

YIELD AND QUALITY OF FIELD GROWN POTATO PLANTS EXPOSED TO ACID RAIN. E. J. Pell, C. J. Arny and N. S. Pearson, Dept. of Plant Pathology and Center for Air Environment Studies, The Pennsylvania State University, University Park, PA 16802.

Solanum tuberosum L. cv. Norchip were grown according to standard field practices from June 1-September 16, 1983. Two 27 x 9 m greenhouses, mounted on rails, served as structures from which simulated rain was delivered to plants and as exclusion shelters to protect against ambient rain. Three times each wk 14.19 l of simulated rain of pH 2.8, 3.8, 4.6 or 5.6 were delivered to 1.2 x 1.8 m plots, respectively. Rain was delivered with whirl plate square stainless steel nozzles at 5 psi from cannisters under pressure with nitrogen. Each treatment was replicated 4 times per greenhouse. Approximately 0.5 cm of rain was delivered in 20 minutes. Shelters were only over plots during a rain treatment or an ambient rain event. At harvest, tuber yield by grade, number and weight revealed no acid rain effects. Subsamples of tubers per plot were analyzed for total solids, α- and β-glucose, fructose and sucrose and total glycoalkaloids. Quality parameters were not affected by acidic rain.

A419

EFFECT OF SIMULATED ACID RAIN TREATMENTS ON ETHYLENE PRODUCTION OF POTATO, SOYBEAN AND RADISH. C. J. Arny and E. J. Pell, Dept. of Plant Pathology and Center for Air Environment Studies, The Pennsylvania State University, University Park, PA 16802.

Solanum tuberosum cv. 'Norland', Raphanus sativus cv. 'Cherry Belle' and Glycine max cv. 'Amsoy 71' were exposed to simulated acid rain treatments of pH 2.8, 3.8, 4.6 and 5.6. Rain duration was 1-hr or 1-hr on 3 consecutive days. Petioles of excised leaves were immersed in 2 ml 0.5mM ACC and 200mM $NaHCO_3$ and incubated in sealed 50 ml flasks for 24 hr in the dark. Air space was sampled and ethylene was quantified using a gas chromatograph fitted with an activated alumina column and a flame ionization detector. Regardless of treatment pH or duration, radish produced more ethylene than potato or soybean. No species exhibited increased ethylene production in response to 1-hr rain treatments. After multiple rain treatments, plants exposed to pH 2.8 rain generally exhibited increased ethylene production. When data were subjected to regression analysis, a weak relationship between treatment acidity and ethylene production was found.

A420

INFLUENCE OF SITE AND CLIMATIC FACTORS ON TREE GROWTH IN PENNSYLVANIA. R. Long and D. D. Davis, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Tree growth in relation to site and climatic factors is being investigated to determine if significant declines in productivity have occurred during the last 50 years. Approximately 540 codominant and dominant trees ≥ 50 years old growing at three locations in Pennsylvania were sampled using a common protocol. Ring widths of two increment cores per tree were measured to the nearest 0.01 mm and crossdated. Ring widths were standardized using a cubic spline function and individual tree chronologies were merged into stand chronologies. Substantial intra-species growth synchronicity was apparent in Quercus alba, Q. prinus, and Liriodendron tulipifera on some sites based on analysis of variance. On more mesic sites, inter-species differences between Q. alba and Q. prinus were evident and intra-species synchronicity was reduced.

A421

THE ROLE OF FOLIAR EPICUTICULAR WAXES IN THE TOLERANCE OF PINES TO DE-ICING SALT SPRAY. M. Simini and I. A. Leone. Plant Pathology Department, Cook College, New Jersey Agricultural Experiment Station, Rutgers University, New Brunswick, NJ 08903.

Five replicates of 3-year-old seedlings of Pinus thunbergii and Pinus strobus were grown at ambient conditions and at three temperature (10C,21C,32C) and photoperiod (8h,12h,16h) regimes. Foliar epicuticular waxes were extracted and analyzed for alkane content using temperature-programmed gas chromatography. Foliar epicuticular wax from the salt-tolerant Pinus thunbergii seedlings had significantly more alkanes than did the wax from the salt-sensitive Pinus strobus seedlings. The alkane content increased in both species as the temperature and photoperiod were increased. Greater alkane concentrations in the waxes of the tolerant Pinus thunbergii and in the waxes of both species at the higher temperatures and photoperiods make the waxes less permeable to polar solutions. These higher alkane waxes may be an important mechanism of de-icing salt spray tolerance in pines.

A422

A COMPUTERIZED SYSTEM FOR EXPOSING PLANTS TO GASEOUS POLLUTANTS. Y. S. Yang, E. J. Pell, and D. D. Davis, Dept. of Plant Pathology and Center for Air Environment Studies, The Pennsylvania State University, University Park, PA 16802.

A computerized system has been developed for air pollutant dispension and data acquisition. The concentrations of pollutants delivered to chambers are regulated by microsolenoid valves. A Digital PDP 11 computer controls degree and time span of valve opening within a 2 sec. interval, thereby controlling pollutant dispersion rate and concentration into exposure chambers. We are applying this technique to control O_3 and SO_2 exposures. Ambient O_3 concentration is monitored; the computer multiplies the ambient value by 1.33, 1.66, or 1.99 and then sets the microsolenoid valves to achieve the desired concentration in the chamber. SO_2 exposures are being simulated based on field data. Field chambers are monitored to establish actual pollutant concentrations and a feed back loop allows the computer to adjust pollutant concentrations where predicted and actual levels deviate. The system can control up to 180 exposure chambers within error of less than 1 pphm.

A423

EFFECTS OF SULFUR DIOXIDE EXPOSURE ON THE DEVELOPMENT OF BACTERIAL COMMON BLIGHT CAUSED BY XANTHOMONAS CAMPESTRIS PV PHASEOLI IN FIELD-GROWN KIDNEY BEANS. K. L. Reynolds, M. L. Zanelli and J. A. Laurence. Boyce Thompson Institute, Ithaca, NY 14853.

Field-grown kidney beans (Phaseolus vulgaris L., cv. 'California Light Red') were exposed to SO_2 using an open air fumigation system for 3 hr, 2 or 3 times per week between July 10 and Sept. 6. Plants were exposed to 10 different SO_2 concentrations that ranged from 0 to 1.00 ppm. Plants were inoculated at the beginning of the exposure period by spray application of a suspension of the bacterium in sterile water. Lesions were counted at 2- to 3-day intervals throughout the exposure period. Increasing cumulative SO_2 dose resulted in a significant decrease in the rate of lesion appearance. The rate of this response remained constant over the course of the exposure period. Increasing cumulative SO_2 dose also led to a reduction in yield of non-infected plants, however there was no apparent reduction in yield of infected plants. Exposure to SO_2 effectively inhibited disease development but reduced yield.

A424

JOINT ACTION OF HF AND SO_2 ON DEVELOPMENT OF COMMON BLIGHT OF BEAN CAUSED BY XANTHOMONAS CAMPESTRIS PV PHASEOLI. J. A. Laurence and K. L. Reynolds. Boyce Thompson Institute, Tower Road, Ithaca, NY 14853.

Three week old Phaseolus vulgaris cv 'California Light Red Kidney' plants were exposed to HF at 0, 1, or 3 ugF m^{-3},

SO_2 at 0, 262, or 786 ug m^{-3}, or all possible combinations of the two gases. Exposures were conducted continuously (HF) or for 6 hours daily (SO_2) for 5 days before, after, or both before and after inoculation with X. campestris pv phaseoli. Diameters of lesions were measured when first visible (after the latent period) and again 5 days later. Pre- and post-inoculation SO_2 caused significantly smaller lesions, and longer latent periods. Post-inoculation exposures to HF significantly increased latent periods. Interactions between HF and SO_2 only occurred when the gases were supplied in combination. In general, the interactions resulted in longer latent periods and smaller lesions.

A425

EFFECTS OF OZONE AND FUSARIUM OXYSPORUM ALONE AND IN COMBINATION ON GROWTH OF EARLY MATURING SOYBEAN LINES. John P. Damicone, William J. Manning, Dept. of Plant Pathology, UMass, Amherst, 01003, and William A. Feder, Suburban Experiment Station, Waltham, MA, 02154.

Early maturing soybean P.I. 180.499 (susceptible) and P.I. 161.989 (resistant) were selected for their reactions to ambient and controlled foliar ozone injury. A factorial-split plot experiment was conducted to determine the effects of ozone exposure (6 pphm, 6 hrs/day, 5 days/wk), soil infestation with Fusarium oxysporum (500 cfu/g soil dw.) and cultivar susceptibility to ozone in plastic greenhouses. Plant dry weights were reduced by ozone 24% with the susceptible line and 11% with the resistant line. Ozone injury was increased by infesting soil with Fusarium, but ozone had no apparent effect on root rot. Ozone and Fusarium reduced relative growth rate of the susceptible line, and this was due to a lower unit leaf rate and not a lower leaf area ratio.

A426

DIFFERENCES IN ALFALFA CULTIVAR SENSITIVITY TO OZONE. Daniel R. Cooley, William J. Manning and William A. Feder. Dept. of Plant Pathology, University of Mass., Amherst, MA, 01003 and Suburban Experiment Station, Univ. of Mass., Waltham, MA, 02154.

Fourteen alfalfa cultivars were screened for susceptibility to ozone (O_3). Four-week-old seedlings were exposed to 6-8 pphm O_3 in a greenhouse for 4 weeks, and compared to seedlings grown in a carbon filtered air greenhouse for the same period. O_3 caused a significant depression for both fresh and dry weights for Buffalo, Iroquois, Oneida, Team and Vernal. Apollo II, Honeyoye, Saranac AR and Vangard were less affected. Dry wt reductions ranged from 0.5% to 69%, and fresh weights were reduced from 8% to 77%. In most cases, a cultivar's visual injury rating correlated with the mean weight reduction. These results suggest that O_3 may decrease alfalfa growth in the Northeast.

A427

THE PHYSIOLOGICAL RESPONSE OF SOYBEAN TO OZONE FUMIGATION AT VARIOUS GROWTH STAGES. G. Smith and E. Brennan, Department of Plant Pathology, Rutgers University, New Brunswick, NJ 08903.

Surface sterilized soybean (Glycine max (L.) Merr. var. Cutler) seed was inoculated with a commercial preparation of Rhizobium japonicum and planted (2 seeds/pot) in 5 L plastic pots containing an autoclaved mixture (1:1:2) of perlite, vermiculite, and sand. Each pot was supplied daily with 500 ml of an N-free solution until emergence of the first trifoliate after which all plants also received a daily supplement of 10 mM KNO_3. At V5, R1, R3, R5, and R6 plants were exposed to a 12-hour O_3 fumigation starting at 0.02 ppm at 0800 hr, increasing gradually to 0.20 ppm at 1500 hr and then decreasing again to 0.02 ppm at 2000 hr. At each growth stage O_3 increased stomatal resistance, decreased leaf chlorophyll, and temporarily inhibited nitrate reductase activity in the leaves (in vivo assay). Root nodule nitrogenase activity (C_2H_2 reduction) was not affected by O_3 except at R3 when a significant decrease was noted in treated plants. A second group of plants grown to maturity under the same conditions and exposed to O_3 at the same growth stages showed no O_3 effect on dry matter production or seed yield.

A428

ULTRAVIOLET-B IRRADIANCE EFFECTS ON SOYBEAN PRODUCTIVITY AND INCIDENCE OF FUNGI IN SEED. P.G. Webb and R.H. Biggs. Fruit Crops Department, University of Florida, Gainesville, FL 32611

There is a 2X amplification factor between ozone concentration in the stratosphere and ultraviolet-B radiation (280-320 nm) in the biosphere of the subtropics. Perturbation in natural or man-made stratospheric gases could lead to a decrease in ozone. Investigations of the possible influences of increased UV-B radiation (0, 16, 23 and 32% enhancement) on soybean (Glycine max) under field conditions has been concluded for the third season. During the first and second seasons, twelve plots per treatment were sampled for plant height, leaf dry weight, pod dry weight, and number of pods at 21, 35, 49, 63, 91 and 111 days. On harvest day 113 during the third season, plant and dry weights were recorded. No significant differences in plant biomass were found among treatments. Analysis of fungal incidence was conducted during the third season and Phomopsis and Cercospora spp. were found in greatest abundance. Significantly higher numbers of seeds were infected by Phomopsis, Cercospora and other fungi among the UV-B treatments in contrast to the control seeds.

A429

EDU: A TOOL FOR ASSESSING CROP LOSS DUE TO AMBIENT OXIDANTS. B. B. Clarke, E. Brennan, and J. Rebbeck. N.J.A.E.S., Rutgers, Univ., New Brunswick, N.J. 08903 #K-11410-1-84.

For years EDU has been used by researchers to protect plants from foliar ozone injury. Recently, applications of the antioxidant have been employed to examine the impact of oxidants on tuber yield and quality of field grown potato plants. Since EDU-treated plants often experience elevated yields, information was needed to determine whether EDU stimulates tuber production. In 1981, Bel Rus, Norchip and Superior potato cultivars were grown under commercial field conditions in the presence or absence of EDU. Foliar symptom developement and accumulated oxidant dose were relatively low during the growing season. Tuber yield, specific gravity and size distribution within cultivars were similar for EDU-treated and untreated plants. Similar results were obtained in 1977 & 1979 when EDU reduced foliar injury by 12%. When oxidant dose and symptom expression increased significantly in 1978 & 1980, untreated plants sustained up to a 31% yield reduction in comparison to EDU-treated plots. These results support the theory that EDU has no effect on yield.

A430

THE OCCURRENCE OF DOUBLE-STRANDED RNA IN SPAWN STRAINS OF AGARICUS BISPORUS. K. L. Deahl, J. P. San Antonio, and E. L. Civerolo. USDA, AR, HSI, BARC-West, Beltsville, MD 20705.

An earlier detailed comparison by electron microscopy indicated that the detection of dsRNA by polyacrylamide gel electrophoresis (PAGE) was a very reliable diagnostic technique for virus infection in mushrooms. Therefore, we initiated a study to determine if the PAGE method could be used to detect double-stranded RNA (dsRNA) in stock strains used to generate commercial spawns (seed). Sporophores were derived directly from test spawns by covering the spawn with a layer of casing material. Sporophores were grown in environmentally controlled isolation chambers. Based on an extensive analysis of spawns from 8 separate national and international stock programs, 41% of the strains tested contained dsRNA. None of the isolates showed aberrant growth, but several strains had abnormal growth rates. Although fruiting initiation by several of the strains was difficult, there was no correlation between fruiting ability and dsRNA content.

A431

DETECTABILITY OF BARLEY YELLOW DWARF VIRUS IN CEREAL LEAF SURVEY SAMPLES. D. L. Clement, M. Skaria, J.A. McFatridge and R.M. Lister, Purdue University, W. Lafayette, IN 47907.

Assessment of the results of enzyme-linked immunosorbent assay (ELISA) of samples received in diagnostic laboratories is increasingly important in surveys for barley yellow dwarf virus (BYDV). Tests of virus detectability by ELISA in cereal leaf samples mailed world-wide and returned to our laboratory showed that it survived much longer in air-dried leaf than in fresh leaf. Survival was excellent with dried leaf pieces after transit periods of 1 month or more, but with fresh leaf virus detectability was drastically reduced after only one week. Virus extraction efficiency was similar for leaf dried over $CaCl_2$ at 4 C or simply exposed to air at 25 C. Detectability of the RPV and MAV isolates of Rochow (Phytopathology 59:1580-1589) in phosphate-buffered leaf extracts was improved by mixing them with chloroform, especially with dried leaf and especially with the RPV isolate.

A432

ASSOCIATION OF THE Yd_2 GENE WITH REDUCED BARLEY YELLOW DWARF VIRUS PRODUCTION IN BARLEY. M. Skaria, R.M. Lister, and J.E. Foster, Purdue University & USDA, SEA, W. Lafayette, IN 47907.

Yd_2, a gene derived from Ethiopian barleys, is reported to confer symptomatic resistance to barley yellow dwarf virus

(BYDV) infection in barley. To investigate its effect on virus synthesis we inoculated 1-wk. old plants of California Mariout (Yd_2-) barley and the near-isogenic CM67 (Yd_2+) with PAV, MAV, or RPV isolates of BYDV (i.e. transmitted by Rhopalosiphum padi L. and Sitobion avenae Fabr.; by S. avenae; or by R. padi, respectively). Plants were grown at 20 ± 1C with 14 hr. light. The virus content of shoots and roots was assessed at six day intervals for one month by enzyme linked immunosorbent assay. With PAV, significantly less virus was detected in CM67 than in California Mariout overall, but with MAV and RPV there were no such differences. Fresh weight reduction was similarly isolate-specific. In other experiments PAV production also behaved similarly in Prato (Yd_2+) barley and the near-isogenic Briggs (Yd_2-), and in Atlas 68 (Yd_2+) barley and the near-isogenic Atlas 57 (Yd_2-).

A433

Detection of tobacco ringspot virus infecting yellow summer squash (Cucurbita pepo) in SC. Bernard Sammons and O. W. Barnett. Department of Plant Pathology & Physiology, Clemson Univ., Clemson, SC 29631.

Commercial fields of yellow summer squash (Cucurbita pepo 'Dixie Hybrid') located in Greenville County were monitored weekly for watermelon mosaic virus-1, watermelon mosaic virus-2 (WMV-II), tobacco ringspot virus (TRSV), cucumber mosaic virus, and squash mosaic virus. Disease incidence was determined by counting plants with or without symptoms. Young leaves with virus-like symptoms were collected from selected fields and virus(es) present identified by enzyme-linked immunosorbent assay (ELISA) and Ouchterlony double gel diffusion. WMV-II and TRSV were found in several fields, although in one field only TRSV was detected. During 4 weeks of observation, TRSV spread in a plant-to-plant fashion, resulting in clusters of infected plants. Infection in this field was less than 1% throughout the study. TRSV was transmitted to Cucumis sativus when planted in field soil that contained Xiphinema americanum; thus nematodes in the field were viruliferous. This is the first report of TRSV in commercial squash fields in South Carolina.

A434

A POTYVIRUS FROM CLOVER THAT INFECTS PEANUT. D. C. Bays and J. W. Demski, Dept. of Plant Pathology, Georgia Experiment Station, Experiment, GA 30212, and A. M. Schubert, TAMU, TAES, Plant Disease Research Station, Yoakum, TX 77995.

A virus was recovered from arrowleaf clover (Trifolium vesiculosum) that infected peanut (Arachis hypogaea). In peanut, initial symptoms were chlorotic rings and spots. After 2-3 weeks these symptoms faded and were no longer evident. Crystalline inclusions were observed in cytoplasm and nuclei of infected peanut plants. No serological relationship was found to the common peanut viruses, peanut mottle and peanut stripe, or to the potyviruses, tobacco etch, blackeye cowpea mosaic, soybean mosaic, watermelon mosaic-2, pepper veinal mottle and potato virus-Y using direct ELISA. A strong serological reaction was obtained in direct ELISA tests against clover yellow vein and bean yellow mosaic viruses.

A435

CARDAMOM MOSAIC A MEMBER OF THE POTYVIRUS GROUP IN GUATEMALA. J. E. Dimitman, Anthony Flores and Jon A. Nickoloff. Dept. of Bio. Sci., Cal State Poly. Univ., Pomona, CA 91768.

Thin sections of young leaf tissue of cardamom, Elettaria cardamom (L) Maton, with typical symptoms of Cardamom Mosaic (CaMV) were found to show flexuous rods with a helical symmetry. Pinwheels similar to those associated wtih Potato virus Y (PVY) were seen in leaf sections. Serological studies with enzyme-linked immunosorbent assay (ELISA) using PVY were not conclusive, but did indicate some relationship. CaMV has not been mechanically transmitted but has been transmitted by two species of aphids.

A436

THE ROLE OF WHEAT SPINDLE STREAK AND WHEAT SOILBORNE MOSAIC VIRUSES IN AN EPIPHYTOTIC OF RESISTANT WHEAT IN KANSAS. S. A. Lommel and W. G. Willis, Dept. of Plant Pathology, Kansas State University, Manhattan, KS 66506.

Severe mosaic and yellowing symptoms in patterns typical of the Polymyxa graminis vectored wheat soilborne mosaic virus (WSBMV) appeared in early March 1984 in south central Kansas. Many of the winter wheat cultivars showing symptoms, particularly Newton, are considered resistant to WSBMV. Electron microscopy revealed WSBMV and wheat spindle streak mosaic virus (WSSMV) particles in a ratio of approximately 20:1, respectively, in all symptomed samples collected. Newton plantings in northeastern Kansas grown in WSBMV infested soil were essentially symptomless and no WSBMV or WSSM virus could be detected. Environmental conditions since fall 1983 planting were apparently optimal for WSSMV and may have reduced resistance to WSBMV, or possibly a new strain of WSBMV has arisen, in south central Kansas.

A437

GEMINATE PARTICLES ASSOCIATED WITH THE LEAF CURL OR 'CHINO' DISEASE OF TOMATOES IN COASTAL AREAS OF WESTERN MEXICO. J.K. Brown and R.B. Hine. Dept. of Plant Pathology, University of Arizona, Tucson, AZ 85721

Leaf curl or 'Chino', a disease incited by a whitefly-transmitted virus-like pathogen of tomato (Lycopersicon esculentum Mill.), was first observed in Mexico in the state of Sinaloa in 1976 but now occurs annually in all coastal tomato growing areas of Western Mexico. The most dramatic foliar symptoms and yield reductions result from early season (Sept-Oct) infection of greenhouse-grown and field transplanted seedlings. All commercially grown tomato varieties and hybrids are susceptible. Leaf curl was transmitted from tomato to tomato by Bemisia tabaci Genn. in greenhouse tests but not by mechanical means. Paired (18x30nm) and single (18nm) virus-like particles were observed by electron microscopy in partially purified preparations of greenhouse-inoculated tomato plants. The dimeric and monomeric particle morphologies are characteristic of members of the geminivirus group of plant viruses and suggest that leaf curl or 'Chino' may be incited by a geminivirus.

A438

SOLANUM X BERTHAULTII: A NECROTIC HOST FOR VIROIDS FROM CITRUS, CHRYSANTHEMUM, POTATO AND TOMATO. R.P. Singh, Agriculture Canada, Research Station, P.O. Box 20280, Fredericton, N.B., E3B 4Z7, Canada.

A clone of Solanum x berthaultii (USDA P.I. 265857) developed necrotic symptoms when infected with either mild or severe strains of potato spindle tuber viroid. It was sensitive enough to allow for the direct indexing of individual tubers using cut pieces or using nucleic acid extract for composite samples. S. berthaultii also developed necrotic symptoms when inoculated with citrus exocortis, Chrysanthemum stunt, and tomato apical stunt viroids. However, S. berthaultii did not become infected and consequently it did not develop any symptoms when inoculated with Chrysanthemum chlorotic mottle viroid either mechanically or by grafting. The symptoms in this clone for all viroids consisted of necrotic spotting of petioles and stems, along with leaf collapse. The necrotic, rolled leaves eventually dry out but they remain attached to the stem. Any new leaves were reduced in size and the entire plant was severely stunted.

A439

SUPPRESSION OF A POTATO SPINDLE TUBER VIROID INHIBITOR IN TRUE POTATO SEEDLINGS. M. E. Grasmick and S. A. Slack, Dept. of Plant Pathology, University of Wisconsin-Madison, WI 53706.

A preliminary study showed the presence of a potato spindle tuber (PSTV) inhibitor in plant sap of true potato seedlings generated from PSTV-infected parents (Phytopathology 71:877). For seedlots with high but not low rates of PSTV-infection, symptoms developed on the bioassay host Rutgers tomato upon serial dilutions of sap inocula. Inocula amended with 30 mg/ml bentonite or 20 units/350 µl placental RNasin but not with 1% sodium lauryl sulfate or 1% β-mercaptoethanol suppressed inhibition. In a comparison of bentonite-amended inocula with non-amended inocula, polyacrylamide gel electrophoresis assays indicated that infection efficiency and PSTV concentration in bioassay hosts were enhanced by bentonite amendments.

A440

CALCIUM INTERACTS WITH IAA AND KINETIN ON THE FORMATION OF LOCAL LESIONS BY ALFALFA MOSAIC VIRUS (AMV) IN BEAN (PHASEOLUS VULGARIS). J. C. Tu, Research Station, Agriculture Canada, Harrow, Ontario NOR 1GO.

The local lesion (LL) production by AMV on bean leaves was reduced if IAA (10^{-3}, 10^{-4} and 10^{-5} M) were applied as post-inoculation spray instead of water. Addition of 0.03 M $CaCl_2$ to 10^{-4} and 10^{-5} M IAA not only nullified the effect of IAA on LL production but stimulated it. However, addition of calcium to 10^{-3} M IAA had little effect on LL production. Kinetin at concentrations of 10^{-3} and 10^{-5} M, had little effect on the LL production and addition of 0.03 $CaCl_2$ to kinetin in these solu-

tions did not alter significantly the LL production as compared to 0.03 M $CaCl_2$ used alone. However, kinetin at 10^{-4} M with or without calcium significantly increased the LL production over distilled water or 0.03 M $CaCl_2$, respectively. The reduction in size of LL on leaves sprayed with IAA or kinetin at 10^{-3}, 10^{-4} and 10^{-5} M was directly related to the concentration. Addition of 0.03 M $CaCl_2$ to IAA and kinetin lessened the effect of IAA and kinetin on the size of LL.

A441

RELATIVE VIRUS CONCENTRATION AND YIELD OF INFECTED SORGHUM HYBRIDS HAVING RED-LEAF OR MOSAIC REACTIONS TO MAIZE DWARF MOSAIC VIRUS STRAIN B. D. L. Seifers and H. L. Hackerott. Fort Hays Branch, Kansas Agricultural Experiment Station, Hays, Kansas 67601.

Infection of red-leaf reactors resulted in high virus concentration, 14 to 18 percent decreases in plant height, and 33 to 66 percent yield reductions. Infection of sorghum hybrids having a mosaic reaction resulted in low virus levels, 8 to 12 percent decreases in plant height and 0 to 8 percent yield reductions.

A442

EFFECT OF LIGHT ON INFECTION OF CORN WITH MAIZE DWARF MOSAIC VIRUS. Eugen Rosenkranz. USDA-ARS, Dept. of Plant Pathology & Weed Science, Mississippi State Univ., Mississippi State 39762.

Infection rate varies yearly when corn is inoculated mechanically with maize dwarf mosaic virus strain A (MDMV-A) in the field. On the premise that light influences infection, I studied the effect of time of day of inoculation on disease development. Corn seedlings in the 3-4 leaf stage inoculated at 18:30-19:00 had a higher disease incidence than those inoculated at 8:30-9:00 or at 13:30-14:00 on the same sunny day. To determine if postinoculation light conditions were responsible for the difference in disease incidence, corn seedlings were inoculated in the greenhouse and one half of them received 8 hr natural light, the other half 8 hr darkness. The latter group simulated inoculation in the field at sunset. In all 8 hybrids tested, the dark treatment resulted initially in a higher disease incidence than the light treatment. The differences were greatest early in che disease development and declined gradually. A small difference in disease severity seemed to persist. Work is in progress to explore the effect of preinoculation light conditions on MDMV-A infection of corn.

A443

POLLEN TRANSMISSION OF AVOCADO SUNBLOTCH VIROID AND THE FATE OF THE POLLEN RECIPIENT TREE. P.R.Desjardins, R.J.Drake,P.J.Sasaki, E.L.Atkins and B.O.Bergh, University of California,Riverside,CA 92521.

A five year study using bee pollination has provided evidence that avocado sunblotch viroid is pollen transmitted in field trees. Under the experimental conditions used, the rate was found to be somewhat low, ranging from 1 to 4%. Both symptomatic and symptomless carrier trees can serve as pollen donors. Most of the infected progeny seedlings exhibited symptoms on germination; however, one seedling did not display symptoms as a seedling, and was only found to be infected when a viroid-free scion was grafted to it. The scion eventually exhibited symptoms indicating that the rootstock had become infected via pollen transmission. This single seedling must be classified as a symptomless carrier type. Extensive tests on the two Zutano variety avocado field trees used as pollen recipients indicate that they did not become infected during the course of pollen transmission.One of the two has been tested annually for six years while the other has been tested for three years.

A444

AN INDIRECT ELISA FOR QUANTITATIVE ESTIMATION OF VERTICILLIUM ALBO-ATRUM IN HOPS. J. E. Leach and T. R. Swinburne. Department of Plant Pathology, Kansas State University, Manhattan, KS 66506 and East Malling Research Station, Maidstone, Kent ME19 6BJ U.K.

An indirect enzyme-linked immunosorbent assay (ELISA) has been adapted for estimating the amounts of Verticillium albo-atrum in field and greenhouse infected hop (Humulus lupulus) roots, leaves and bines. The assay, which uses an antiserum prepared to a water-soluble extract of washed, macerated V. albo-atrum mycelium, can detect less than 100 ng of freeze-dried V. albo-atrum. Specificity for V. albo-atrum was enhanced by cross absorption of the antiserum with proteins extracted from V. dahliae. Representatives of seven different fungal genera were assayed using this antiserum; all except a V. dahliae isolated from hops had reactivities less than 0.1% that of V. albo-atrum. Artificially produced infection levels representing 0.5% infected tissue are easily distinguished from healthy tissue extracts by the assay.

A445

A BAITING TECHNIQUE FOR SELECTIVE ISOLATION OF RHIZOCTONIA ZEAE FROM SOIL. A. S. Windham and L. T. Lucas, Department of Plant Pathology, North Carolina State University, Raleigh 27695.

A baiting technique was developed for isolating R. zeae from soil using fungicide-treated cotton stem segments and a selective medium consisting of water agar 2%, benomyl 10 µg/ml, metalaxyl 10 µg/ml, penicillin g 50 µg/ml, and streptomycin sulfate 50 µg/ml. Cotton stem segments 10 mm long were soaked in sterile distilled water containing 500 µg/ml benomyl and 100 µg/ml metalaxyl for 2 hr. The stems were placed into a naturally infested soil and incubated for 72 hr at ca 24 C. The stems were removed, washed under tap water for 20 min, blotted dry and placed onto the selective medium. The plates were observed after 24 and 48 hr. Rhizoctonia zeae was isolated from 74.6% of the fungicide treated stem segments, but from only 10.6% of the untreated stem segments. Pythium spp., R. solani, and binucleate Rhizoctonia-like fungi colonized untreated stem segments but did not colonize fungicide-treated stem segments.

A446

Sugar maple cotyledons used to detect Phytophthora citricola and P. cactorum in soils. M.J. Drilias, J.E. Kuntz, and G.L. Worf, Dept. Plant Pathology, Univ. Wisconsin, Madison, WI 53706

Collar rot induced by P. citricola (Pci) is a cause of sugar maple decline in Wisconsin. An assay to detect Pci was developed to determine the distribution of the pathogen in urban, nursery, and woodland soils. An 8 cc subsample of air-dried and sieved (3.5 mm screen) soil was spread in a 90 X 12 mm petri plate and moistened with 5 ml sterile distilled water (SDW). Five days after the subsample had been moistened, it was flooded with 25 ml SDW and six half-cotyledons from 3- to 5-week-old sugar maple seedlings were floated on the water. Sporangia of Pci developed on colonized cotyledons 2-11 days after soils had been flooded. Incubation at 20 C was superior to incubation at 16 C and 24 C. Phytophthora cactorum (Pca) also was detected by this method and occasionally colonized the same cotyledons as did Pci. Both fungi have been detected near the base and under the crown of infected and non-infected ornamental maples. However, Pca has not been isolated from maples in Wisconsin.

A447

SYNERGISTIC INTERACTIONS BETWEEN VERTICILLIUM DAHLIAE AND PRATYLENCHUS SPECIES IN POTATO EARLY DYING DISEASE. Randall C. Rowe and Richard M. Riedel. Dept. of Plant Pathology, Ohio Agric. Res. and Devel. Center and The Ohio State University, Wooster 44691 and Columbus 43210.

Field microplot studies over the past 6 years have conclusively demonstrated a synergistic interaction of the lesion nematode Pratylenchus penetrans with Verticillium dahliae in potato early dying. Tests with three populations of P. penetrans and two of V. dahliae, alone and in all combinations, have shown that disease occurs when both pathogens interact synergistically at populations that individually have little or no effect. Yield loss is not always associated with foliar symptom development, but occurs with the addition of high temperature stress during tuberization. Comparative studies with two other species of Pratylenchus commonly found in Ohio potato soils have shown that P. crenatus does not interact with V. dahliae and P. scribneri interacts only slightly. All three nematode species feed and reproduce well on potato, thus, negating the theory that Verticillium interactions are primarily due to wounding.

A448

PHYTOPHTHORA SPECIES ASSOCIATED WITH NURSERY-GROWN APPLE ROOTSTOCKS AND TREES. S. N. Jeffers and H. S. Aldwinckle, Dept. Plant Pathology, Cornell Univ., NYSAES, Geneva, NY 14456.

In 1983, root washings from individual bundles of clonal apple rootstock liners (CRL), apple seedling rootstocks (SR), and 2-yr-old, commercially available apple trees were assayed for contamination with Phytophthora species using a baiting bioassay. CRL (MM.106, MM.111, M.7a, M.26, and M.9) had P. cactorum and Phytophthora spp. (typified by nonpapillate, internally

proliferating sporangia) on 67/70 and 47/70 bundles, respectively. Eleven of 14 SR bundles yielded only P. cactorum. Trees had P. cactorum and Phytophthora spp. on 85/92 and 6/92 bundles, respectively. Data from 1984 were similar. Contaminated MM.106 CRL planted in vermiculite and subjected to periodic flooding became severely diseased, whereas those not flooded remained healthy. The association of Phytophthora species, especially P. cactorum, with roots of nursery-grown apple plants appears to be a widespread phenomenon and could be important in the incidence and spread of Phytophthora crown rot of apple in New York and elsewhere.

A449

THE PATHOGENICITY AND RELATIVE VIRULENCE ON MAHALEB AND MAZZARD CHERRY OF 7 PHYTOPHTHORA SPP. W. F. Wilcox and S. M. Mircetich, Dept. Plant Pathology, NY State Agr. Expt. Station, Geneva, NY 14456, & USDA, ARS, Dept. Plant Pathology, UCD, Davis, CA 95616.

Isolates of Phytophthora cryptogea, P. cambivora, and P. megasperma from cherry, isolates of P. cinnamomi and P. citricola from walnut, and an isolate of P. cryptogea from safflower caused 88-100% root rot and caused crown rot on 15, 11, 6, 12, 11, and 4 out of 15 Mahaleb cherry seedlings, respectively, when plants were grown for 15 wk in artificially infested UC mix and periodically flooded. In contrast, cherry isolates of P. drechsleri and an unidentified Phytophthora sp. caused 62% and 41% root rot, respectively, and caused no crown rot. Mazzard cherry appeared significantly more resistant than Mahaleb to both root and crown rot caused by P. cambivora, P. megasperma, and the safflower isolate of P. cryptogea, and to crown rot caused by P. cinnamomi and P. citricola. However, Mazzard appeared nearly as susceptible as Mahaleb to root rot caused by P. cinnamomi, P. citricola, P. drechsleri, and Phytophthora sp. and to root and crown rots by the cherry isolate of P. cryptogea.

A450

POPULATIONS OF PRATYLENCHUS, PYTHIUM, RHIZOCTONIA, AND THIELAVIOPSIS OF 20 SNAP BEAN FIELDS IN NEW YORK. G. S. Abawi and A. C. Cobb, Dept. of Plant Pathology, NYSAES, Cornell Univ., Geneva, NY 14456.

Densities of Pratylenchus spp., low-temperature Pythium spp. (mostly P. ultimum), R. solani, and T. basicola in 20 central and western New York fields were determined at planting and after harvest. Fields were sampled whenever beans were grown for 3 consecutive years. Densities of all pathogens varied greatly within and among fields, and those determined after harvest generally were higher. The densities of the pathogens as an average for all fields, range of field averages, and range of total samples were respectively as follows: Low-temperature Pythium spp. - 599, 344-1066, and 37-2426 propagules (pg)/g soil; R. solani - 5.2, 1.1-9.1, and 0-45 pg/100 g; T. basicola - 223, 39-516, and 0-1213 pg/g; and Pratylenchus spp. - 59, 3-168, and 0-420 nematodes/100 g. Statistical analysis of the data showed variance-to-mean ratios were significantly greater than one, which suggested clumped inocula distributions in the field.

A451

YIELD REDUCTION IN LETTUCE RESULTING FROM SUBCLINICAL INFECTION OF FEEDER-ROOTLETS BY PYTHIUM DISSOTOCUM. M.E. Stanghellini, Department of Plant Pathology, University of Arizona, Tucson, 85721.

Isolations from asymptomatic rootlets of healthy-appearing bibb lettuce plants, obtained from commercial hydroponic facilities in Arizona, California, and Illinois, consistently yielded Pythium dissotocum. Pathogenicity tests, conducted under hydroponic conditions, showed that P. dissotocum, in the absence of any root rot symptoms, was responsible for a 54% reduction in yield. Pythium dissotocum, in addition to P. uncinulatum (a known seedling pathogen of lettuce not previously reported in the United States), was also consistently isolated from healthy-appearing feeder rootlets collected from field-grown head lettuce plants.

A452

FUSARIUM ROOT ROT OF GUAYULE (PARTHENIUM ARGENTATUM). R. E. Ykema and J.C. Stutz, Division of Agriculture, Arizona State University, Tempe, AZ 85287.

Isolates of Fusarium oxysporum, F. solani, and F. roseum from diseased guayule roots with root rot symptoms were screened for pathogenicity. Guayule plants were grown using the slant board nutrient solution technique (Kendall and Leath, 1974). Twelve week old plants were inoculated by placing a mycelium-covered polyester/cotton cloth (1cm^2) directly over the root tip. Results showed that not all of the Fusarium species isolated from diseased guayule roots were pathogenic. Isolates of F. oxysporum did cause necrosis of inoculated roots. Inoculated roots were stained and examined for hyphal colonization of the root cortex by F. oxysporum. Guayule plants were also grown for two months in pots containing pasteurized U.C. mix and inoculated with individual isolates of the three Fusarium species. Two months after inoculations the plants were assessed for root rot symptoms, and results were similar to those obtained using the slant board technique.

A453

A SUCCESSION OF FUNGAL COMMUNITIES ASSOCIATED WITH ROOTS AND CROWNS OF WINTER WHEAT IN NEW YORK. R. T. Kane and R. W. Smiley, Dep. of Plant Pathology, Cornell Univ., Ithaca, NY 14853.

Studies were conducted during two growing seasons to determine the fungal species associated with root and crown rots of winter wheat. Fungi were isolated from healthy as well as discolored tissue for six sampling dates. Changing patterns of colonization indicated that a succession of fungal communities may occur. Early colonists of seedlings included members of the Mucorales, *Alternaria*, and *Fusarium oxysporum*. Following winter dormancy, *Microdochium bolleyi*, *Trichoderma*, *Penicillium*, and a number of nonsporulating types predominated. *M. bolleyi* was isolated at high frequencies until after anthesis, when numbers declined. *Periconia*, *Botryotrichum*, *F. oxysporum*, and *Colletotrichum graminicola* increased in frequency as the crop matured. *F. graminearum*, *F. avenaceum*, and *F. tricinctum* occurred at moderate levels throughout both seasons. *Bipolaris sorokiniana* was notably absent from all samples. Pathogenicity of predominant isolates will be briefly discussed.

A454

INTACT CORES FOR SIMULATED PASTURE MANAGEMENT STUDIES ON ROOT DISEASE COMPLEXES OF TRIFOLIUM SPP. Richard W. Smiley, P. A. Taylor, F. C. Greenhalgh, and R. G. Clarke. Dept. of Agriculture, Plant Research Inst., Burnley, Victoria, Australia 3121.

Subterranean clover seedlings were grown in pasture soils collected as intact cores (8-cm diam by 11-cm deep) and incubated in growth chambers. Influences of simulated pasture management procedures were evaluated for effects on root rot complexes. Root rot severity and incidence were reduced by using resistant cultivars, fungicidal soil drenches, and seed treatments, and were increased by simulated grazing and by alternate soil drying and wetting. Root rot was least at 15 C. Simulated cultivation reduced disease only in soils from pastures where native inocula were near the soil surface. Intact cores provided root disease responses similar to those observed in the field. Cores are more useful than shovel-collected soils for development of integrated soil and plant management procedures to control root disease complexes.

A455

PATHOGENICITY OF PHYTOPHTHORA CITRICOLA FROM AVOCADO TO PERSEA INDICA. Yigal Cohen and Michael D. Coffey, Department of Plant Pathology, University of California, Riverside, CA 92521.

Seven isolates of P. citricola collected from avocado groves in southern California were compared for pathogenicity on P. indica in a rapid laboratory technique. Inoculum droplets containing zoospores were placed on the intact stem and/or main root of P. indica plants (35-45 days old) laying in moist aluminum trays at ~22 C. Dark brown lesions were initially seen on stems and roots at 20-44 hr after inoculation, depending on inoculum concentration and isolate. Lesions expanded at a rate of ~0.1-1.0 cm/day, according to isolate. Rates of lesion expansion on stem and root were positively correlated for six out of 7 isolates. In roots, tips were most susceptible to the pathogen, while in stems, lower part was least susceptible. Although stem lesions readily expanded into leaves no symptoms developed in leaves when surface inoculated with zoospores, unless wounded. P. citricola did not sporulate on the surface of infected tissues of P. indica.

A456

DIFFERENTIAL SELECTIVITY OF METALAXYL IN ALTERING PSEUDOMONAS POPULATION COMPOSITION IN THE RHIZOSPHERE. T.V. Suslow. Advanced Genetic Sciences, Inc., 6701 San Pablo Ave., Oakland, CA 94608

Metalaxyl, applied as a soil drench, altered the species composition among fluorescent Pseudomonas spp.(FP) colonizing cotton rhizospheres. Metalaxyl 2E-G applied to soil in pot

trials at 1.0 g per 1.0 Kg soil selectively enhanced the colonization and subsequent population density of P. fluorescens strain TS011 relative to P. putida strain TS013. TS013 was the predominant FP on cotton roots in both Yolo Clay Loam (YC) and Hersperia Fine Sandy Loam (HFL) in the absence of metalaxyl. Colonies morphologically identical to strain TS011 were present on roots, without metalaxyl, at frequencies of approx. 1.0 colony forming unit per 10^5 total FP, in field isolations from YC soil. Addition of metalaxyl favored the growth of TS011 when co-inoculated on seed at densities of 1:1, 1:100, and 1:1000 relative to strain TS013. Metalaxyl added to agar media was neither inhibitory nor utilized by either strain.

A457

DOT-IMMUNOBINDING ASSAY FOR VIRUS DETECTION. D. J. Gumpf, Wichai Kositratana, and Guang-Yu Zheng, Department of Plant Pathology, University of California, Riverside 92521.

A rapid, sensitive method has been developed to detect viral antigens applied to nitrocellulose membranes as spots of crude sap. Remaining sites of attachment were blocked when the nitrocellulose was washed with polyoxyethylenesorbitan monolaurate (Tween 20). The spots were then reacted with polyclonal or monoclonal antibodies followed by treatment with anti-antibody conjugated with alkaline phosphatase. The alkaline phosphatase was detected by the reduction of the tetrazolium salt to deformazan by the hydrogen ions released in the formation of indigo by the reaction of the phosphatase on the indoxyl phosphate. This reaction forms an insoluble precipitate which stains the nitrocellulose membrane.

A458

DETECTION OF TOBACCO MOSAIC AND TOBACCO RINGSPOT VIRUSES BY ELISA IN HERBACEOUS AND WOODY HOSTS IN CENTRAL NEW YORK. P. Shiel and J. D. Castello. State University of New York, College of Environmental Science and Forestry, Syracuse, NY 13210.

Leaf, root, flower, and seed tissue from 45 herbaceous and woody plant species in three central New York fields were collected monthly from May through September 1983. The samples were frozen at -20°C until indexed for tobacco mosaic virus (TMV) and tobacco ringspot virus (TbRSV) by enzyme-linked immunosorbent assay (ELISA). Tobacco mosaic virus was detected consistently from either leaves and/or roots of: black raspberry (Rubus occidentalis L.), red osier and red-panicle dogwood (Cornus stolonifera Michx. and C. racemosa Lam.), goldenrod (Solidago canadensis L.), plaintain (Plantago lanceolata L.), burnet (Sanguisorba minor L.), cinquefoil (Potentilla sp.), and cultivated oats. Tobacco ringspot virus was detected consistently in either root, leaf, flower or seed tissue of 13 of 45 plant species tested. Compounds that interfered with the ability to detect virus by ELISA were present in many of the herbaceous species and some of the woody species tested.

A459

HIGHLY SENSITIVE SEROLOGICAL DETECTION OF POTATO VIRUS Y. P. H. Berger, D. W. Thornbury, and T. P. Pirone, Dept. of Plant Pathology, University of Kentucky, Lexington, KY. 40546.

As little as 1-5 pg potato virus Y could be detected, using a dot-blot immunobinding assay. A 1 or 2 µl sample of purified virus was applied to washed, dry nitrocellulose (NC) paper, allowed to dry and washed 3x with tris buffered saline (TBS - 0.05 M, pH 7.4, 200 mM NaCl). Unbound sites in the NC were blocked with 10% horse serum and 0.3% BSA in TBS. Blots were incubated overnight with the primary (rabbit) anti-virus antibody. After removing unreacted IgG, NC was incubated 3-4 h with the secondary antibody, goat anti-rabbit-alkaline phosphatase conjugate. Phosphatase was detected by incubating blots with 5-bromo-4-chloro-3-indolyl phosphate as substrate and nitro blue tetrazolium as reaction product stain. Protein A and avidin or biotin conjugates and complexes, as well as horseradish peroxidase and β-D-galactosidase enzymes allowed similar levels of detection, and can be used when alternate detection systems are desired.

A460

DOT-ELISA ON NITROCELLULOSE MEMBRANES FOR DETECTION OF POTATO LEAFROLL VIRUS. F. Davis Smith and E.E. Banttari, Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

Potato leafroll virus (PLRV) was detected in potato foliage, tubers and sprouts using double antibody sandwich enzyme-linked immunosorbent assay (dot-ELISA) on 0.45 µm nitrocellulose membranes (NCM). The assay utilized alkaline phosphase γ-globulin conjugate, ASMX phosphate substrate with fast red TR stain that produced red dots on the NCM for PLRV infected samples. The dots were visually detectable at a 1:2048 dilution PLRV-infected:healthy leaf sap. Quantitative data on reaction intensities were obtained with a MacBeth densitometer. The addition of 0.01 M NaDIECA and 0.01 M NaEDTA in a Tris-saline buffer to each sap dilution (1:1) resulted in an enhancement of positive and a reduction of nonspecific reactions. The dot-ELISA assay for PLRV in potato leaves was four times more sensitive than double antibody sandwich ELISA in polystyrene cuvette packs.

A461

DETECTION OF THREE PLANT VIRUSES BY AN IMMUNO-BLOT ASSAY. Charles A. Powell. Pennsylvania Department of Agriculture, 2301 N. Cameron Street, Harrisburg, PA 17110-9408

An immuno-blot assay (IBA) was evaluated for detection of tobacco mosaic virus (TMV), tobacco ringspot virus (TbRSV), and tomato ringspot virus (TmRSV). Antigen was first adsorbed to a nitrocellulose membrane. Next, the membrane was incubated in antibody to the adsorbed antigen, and then in enzyme-conjugated antibody to the first antibody. Finally, the membrane was exposed to substrate and examined for a colored precipitate. IBA detected 30 pg (30 ng / ml in a 1uL sample) of purified TMV or TbRSV and 100 pg (100 ng / ml in a 1uL sample) of purified TmRSV. Similar detection levels were achieved when purified virus was diluted in healthy tobacco or Chenopodium quinoa sap. Cross - adsorption of the first antibody with healthy plant sap and incubation of the membrane and adsorbed antigen in 2% Triton X-100 were necessary to eliminate healthy sap background. IBA is a sensitive indirect immunoassay for plant viruses which eliminates the use of microtiter plates.

A462

AUTORADIOGRAPHIC DETECTION OF TOBACCO ETCH VIRUS IN APHIDS. P. H. Berger, D. W. Thornbury, and T. P. Pirone, Department of Plant Pathology, University of Kentucky, Lexington, KY 40546

Green peach aphids (Myzus persicae Sulzer) were fed through a parafilm membrane on suspensions containing 100 µg/ml ^{125}I-labeled tobacco etch virus (TEV) and purified helper component protein. After a 10 min acquisition access period, insects were counted in a gamma counter, chilled immediately to approx. 5 C, anesthetized, and sectioned serially in a cryostat at -20C. Sections were glutaraldehyde fixed and post-coated with 5% gelatin, followed by at least 14 days autoradiographic exposure. Virus was detected by autoradiography in 24 of 32 aphids. In 21 of these 24 aphids, virus was located near the tip of proboscis and frequently in 2 or 3 other distinct areas anterior to the cibarium. In the remaining 3, virus was found only in the gut. The maximum amount of virus obtained by an aphid was about 5 pg (60,000 particles), based on virus specific activity and gamma counter readings.

A463

SEROLOGICAL DETECTION AND EVIDENCE FOR MULTIPLICATION OF MAIZE MOSAIC VIRUS IN THE PLANTHOPPER, PEREGRINUS MAIDIS, B.W. Falk and J.H. Tsai, University of Florida, EREC, Belle Glade, 33430; and Fort Lauderdale Research and Education Center, 33314.

Maize mosaic virus (MMV) was detected in individual Peregrinus maidis using the enzyme-linked immunosorbent assay. MMV was detected in P. maidis both after acquisition access period (AAP) on MMV-infected maize and after injection of MMV into P. maidis. MMV was not detected in similarly treated Dalbulus maidis, a non-vector of MMV. 58% and 76% of individual P. maidis reacted positively for MMV 20 days after AAP and ten days after injection, yet in experiments on individual P. maidis only about 30% of the MMV-positive P. maidis transmitted MMV to maize. MMV always was detected much earlier (5-6 days) in P. maidis that acquired MMV by injection as compared to P. maidis after AAP (11-14 days). Both the latent period (time between injection and detection of MMV) and the percentage of individuals infected were dose dependent. P. maidis injected with higher concentrations of MMV had shorter latent periods and more individuals infected.

A464

MAIZE DWARF MOSAIC VIRUS (MDMV) TITER IN SORGHUM TISSUE **M. K. Palomar, S. G. Jensen and E. M. Ball, Dept. of Plant Pathology, University of Nebraska, and Agricultural Research Service, U.S. Department of Agriculture, Lincoln, Nebraska 68583.**

The double antibody sandwich method of the enzyme-linked immunosorbent assay was used to measure the concentration of strains A and B of MDMV in various sorghum tissues under different envi-

tonmental conditions. We could distinguish 50% differences in virus titer. Environmental factors had the following influence: (a) Virus titer was higher at 25C than at 15C or 35C. (b) Light quality and intensity had no effect on virus titer, yet plants grown in the controlled environment chamber had more virus than those grown in the greenhouse. In different plant parts virus titer increased from the youngest down to about the 4-5th leaf from the top and then declined only slightly from there on down. During grain fill there was considerable virus in every plant part, even the head, but the virus disappeared from the grain and declined, in the older leaves, by physiological maturity.

A465

COMPARATIVE ANALYSIS OF CELL-FREE TRANSLATIONAL PRODUCTS OF SOME STRAINS OF CUCUMBER MOSAIC VIRUS. D. K. Lakshman and D. Gonsalves, Department of Plant Pathology, Cornell University, New York State Agricultural Experiment Station, Geneva 14456.

The unfractionated RNAs of five strains of cucumber mosaic virus (CMV) had been translated in rabbit reticulocyte lysate system and their products were compared by SDS-polyacrylamide gel electrophoresis. All five strains translated principal polypeptides with molecular weights of about 110-, 105 to 97-, 30.5 to 29-, and 25 x 10^6, the last being identified as coat protein by immunoprecipitation tests. On the basis of these results the five CMV strains examined could be divided in two groups -- B, C, and N Group and WL and L-2 group. Further serological studies by immunodiffusion tests indicated that CMV strains B, C, and N belong to DTL serotype (Devergne and Cardin, 1973, Ann. Phytopathol. 5:409-430) whereas WL and L-2 belong to ToRS serotype. This grouping is in general agreement with that based on coat protein peptide mapping (Edwards and Gonsalves, 1983, Phytopathology 73:1117-1120).

A466

SEPARATION OF CITRUS TRISTEZA VIRUS STRAINS AND STRAIN VARIANTS BY STEM-SLASH INOCULATION OF CITRUS RECEPTORS. S. M. Garnsey, U.S. Department of Agriculture, Agricultural Research Service, 2120 Camden Road, Orlando, FL 32803.

Several biologically different isolates of citrus tristeza virus (CTV) were recovered when partially purified preparations of a severe (seedling yellows) isolate of CTV were stem-slash inoculated to etrog citron. Most infected receptor plants showed typical, severe symptoms, but several showed atypical mild symptoms. Symptom differences were confirmed by graft-inoculation to other indicators. Further segregants were obtained from the original variants by subsequent stem-slash inoculation. Stem-slash inoculations with partially purified CTV from plants dually infected (experimentally) by a mild and a severe strain of CTV also yielded infected plants with differing symptoms. The stem-slash procedure is effective for separating in vitro mixtures because the systemic infections obtained apparently often result from a single locus of infection.

A467

COMPARISON OF THE PROTEIN AND NUCLEIC ACIDS OF TWO HORDEIVIRUSES. Brenda Hunter, Louis A. Heaton, Robert M. Hanau and A. O. Jackson, Department of Botany & Plant Pathology, Purdue University, West Lafayette, IN 47907.

Poa semilatent virus (PSLV) is a candidate for inclusion in the hordeivirus group based on its reported serological relationship to barley stripe mosaic virus (BSMV) and its particle morphology. Further studies are needed to compare the physicochemical properties of PSLV and BSMV before their relationship can be firmly established. Our polyacrylamide gel electrophoretic data show that both BSMV and PSLV contain a single polypeptide, but the coat protein of BSMV has a molecular weight of 25,000 compared to 23,000 for PSLV. The genomes of both viruses separate into two or more RNA species during electrophoresis in agarose gels. However, the two PSLV RNAs are slightly larger than the corresponding BSMV RNAs, and they have little nucleotide sequence homology because they fail to cross hybridize. These results suggest that BSMV and PSLV have only a distant relationship.

A468

SEROLOGICAL RELATIONSHIPS AND HOST RANGE PROPERTIES OF ZUCCHINI YELLOW MOSAIC VIRUS AND WATERMELON MOSAIC VIRUS ISOLATES. Robert F. Davis and M. A. Yilmaz, Plant Pathology Dept., Rutgers Univ., N.J.A.E.S., New Brunswick, NJ 08903 and Plant Protection Dept., Cukurova Univ., Adana, Turkey. Pub. # K-11191-4-84.

Selected isolates of zucchini yellow mosaic virus (ZYMV) and watermelon mosaic virus (WMV) strains 1 and 2 were compared. Differences were observed in the host ranges of WMV-1, WMV-2, and ZYMV, but not among isolates of each individual virus. In Ouchterlony tests, reactions with some antisera (AS) indicated that ZYMV and WMV-2 were related but distinct, whereas relationships were not detected with other AS. Using enzyme-linked immunosorbent assay (ELISA), moderately strong reactions occurred with ZYMV and AS to WMV-2, and vice versa. The degree of cross-reaction varied with AS and with each test. WMV-1 reacted weakly with AS to ZYMV and vice versa in ELISA but not Ouchterlony tests. WMV-1 and WMV-2 did not cross react in either test. Precise identification and determination of serological relationships among these viruses appears to depend partly on the AS used. However, ZYMV appears to be related to WMV-2 and WMV-1.

A469

UNSUITABILITY OF GLYCEROL FOR THE PRESERVATION OF POTATO VIRUS Y. R.P. Singh, Agriculture Canada, Research Station, P.O. Box 20280, Fredericton, N.B. E3B 4Z7, Canada.

Glycerol has been used for the long-term preservation of purified stable plant viruses, but no information is available of its effect on labile viruses like potato viruses A and Y. Potato virus Y was purified by two cycles of differential centrifugation and two of equilibrium centrifugation in CsCl. The purified virus (2 mg/ml) was mixed with equal volumes of either glycerol or buffer and stored at 25, 4, -20, and at -70 C. Samples were removed periodically from each temperature-storage regime and tested for infectivity based on local lesion and for serological reaction with the ELISA test. Both infectivity and ELISA readings were reduced in virus samples containing glycerol compared to buffer only samples. The diminished infectivity and ELISA readings occurred more rapidly at high temperature than at low. The ultraviolet spectra of samples did not show significant variation.

A470

ON THE IDENTIFICATION OF COMMON COTYLEDONOTROPHIC TRANSLOCATORY DENOMINATOR IN THE HOSTS SUSCEPTIBLE TO GROUNDNUT (= PEANUT) CHLOROTIC SPOT VIRUS. Vedam Chandrasekharam, Research Centre in Biology, Department of Zoology, S.G.S.Arts College, Tirupati, India 517501

In the leaves of the hosts susceptible to the groundnut (= peanut) chlorotic spot virus (GCSV), gamma-methylene glutamic acid (gamma-MGA), the omega-deamidation product of gamma-methylene glutamine (gamma-MG), was identified as the principal constituent of the free amino acid pool. Dedicated to Dr. L. FOWDEN as a mark of appreciation of his contribution to the understanding of the physiological biochemistry of groundnut plant.

A471

IDENTIFICATION OF A GROUP 4 STRAIN OF POTATO VIRUS X INFECTING THE CULTIVAR KING EDWARD. M. J. Foxe, Plant Pathology Dept., University of Florida, Gainesville, FL 32611.

An isolate of potato virus X (PVX), was found in samples of foliage from the cultivar King Edward. This isolate was transmitted to a range of tobacco plants and was routinely propagated in Nicotiana glutinosa or N. tabacum White Burley. It systemically invaded potato cultivars with the PVX hypersensitivity genes Nx or Nb, or Nx and Nb in combination. These reactions are similar to those of the group four strain of PVX. This isolate did not systemically infect PVX-immune cultivars. The isolate had slightly flexuous filamentous particles with a normal length of 504nm. Sap from infected N. glutinosa was infective after dilution of 10^{-7} but not 10^{-8}, after 10 min at 75°C but not 80°C and after 6 months at 20°C. The isolaTE IS NAMED X_{KE}.

A472

EXPRESSION OF RESISTANCE TO STRAINS OF POTATO VIRUS X IN ISOLATED POTATO PROTOPLASTS. M. J. Foxe and J. Prakash, Plant Pathology Dept., University of Florida, Gainesville, FL 32611.

Resistance to Potato Virus X (PVX) in potato cultivars is conferred by the genes Nx and /OR Nb. The Nx gene is activated by group 3 strains of PVX. When protoplasts from the cultivars King Edward containing the Nx gene and Pentland Crown containing neither resistance gene were inoculated with a group 3 strain of PVX resistance was expressed in the King Edward protoplasts.

Replication of PVX occurred in King Edward protoplasts but only at a very low level as compared with the level observed in the susceptible Pentland Crown protoplasts. Similar results were obtained when protoplasts obtained from leaf discs mechanically inoculated with PVX were used. Virus levels were assayed by immunofluorescence and ELISA. Maintenance of resistance in protoplasts of King Edward following inoculation with PVX RNA was confirmed suggesting that resistance probably operates at the transcription stage in replication. These results clearly demonstrate single gene resistance in potato protoplasts inoculated with PVX.

A473

METABOLISM OF FUNGITOXIC TERPENOIDS IN RESISTANT AND SUSCEPTIBLE COTTON STELE SUBSEQUENT TO INOCULATION WITH VERTICILLIUM DAHLIAE. N. A. Garas, M. S. Lee and A. C. Waiss, Jr., USDA, ARS, WRRC, 800 Buchanan St., Berkeley, CA 94710.

The fungitoxic sesquiterpenoids and sesquiterpenoid aldehydes which accumulate in the stele of cotton plants inoculated with V. dahliae was studied in 4 cotton varieties. A new HPLC method was developed for this purpose. The major antifungal compounds detected in the resistant variety Seabrook Sea Island (SBSI-Gossypium barbadense L.) were; hemigossypol, desoxyhemigossypol and the methylated derivative of both compounds. Only trace amounts of the methylated derivatives were found in the 3 varieties of Gossypium hirsutum L. and none was detected in extracts from non-inoculated plants. The accumulation of these terpenoids was twice as high in SBSI as it was in the most tolerant variety of G. hirsutum (Acala SJC-1). In the G. hirsutum varieties, the total terpenoids which accumulate during the first 4 days showed quantitative correlation with the varietal tolerance to wilt, but was not so in the fifth day. The fungitoxicity of total stele extracts and each purified compound will be presented.

A474

HISTOCHEMICAL LOCALIZATION OF THE PHYTOALEXIN DESOXYHEMIGOSSYPOL IN VERTICILLIUM DAHLIAE-INFECTED COTTON STEM. M. E. Mace, R. D. Stipanovic, and A. A. Bell. USDA, ARS, National Cotton Pathology Research Laboratory, P. O. Drawer JF, College Station, TX 77841.

The terpenoid desoxyhemigossypol (dHG) is a precursor of the phytoalexin hemigossypol in Verticillium-wilt resistant Seabrook Sea Island cotton (SBSI). The $SbCl_3/HClO_4$ reagent at room temperature (23-25°C) gave a green-colored product that was specific for dHG in stem stele sections of Verticillium-infected SBSI plants. The green Sb-dHG product was localized in paravascular parenchyma cells of fresh stele sections at 2 to 10 days after stem-puncture inoculation with V. dahliae conidia. No green Sb-dHG product was detected in the stem steles of water-injected plants.

A475

TOXICITY OF TERPENOID PHYTOALEXINS FROM COTTON TO VERTICILLIUM DAHLIAE. M. E. Mace, R. D. Stipanovic, and A. A. Bell. USDA, ARS, National Cotton Pathology Research Laboratory, P. O. Drawer JF, College Station, TX 77841.

Hemigossypol (HG), methoxyhemigossypol (MHG), desoxyhemigossypol (dHG) and desoxymethoxyhemigossypol (dMHG), from Verticillium dahliae-infected, wilt-resistant Seabrook Sea Island (SBSI) cotton were tested at pH 6.3-7.5 in liquid nutrient media for toxicity to V. dahliae. The terpenoids dHG, HG, dMHG, and MHG at 25 C killed conidia after 18-40 hr at 10, 45, 25, and 60 μg/ml, respectively; and mycelia after 48 hr at 15, 35, 25, and 45 μg/ml, respectively. Inhibition of conidia germination also occurred at concentrations well below the fungicidal concentrations. Dimethylsulfoxide at 2 to 5% and/or increase of the pH above that of infected stem xylem (pH 6.3) was required to solubilize HG, MHG, and dMHG at inhibitory concentrations. Only dHG was sufficiently water soluble at pH 6.3 to account for death of V. dahliae conidia and mycelia at most sites in the stem stele of SBSI cotton 10 days after inoculation. The dHG in the stem stele was well in excess of fungicidal concentrations.

A476

ELICITATION OF SESQUITERPENOID STRESS METABOLITES IN POTATO TUBER SLICES BY HELMINTHOSPORIUM CARBONUM. M. Zook and J. Kuć. Dept. of Plant Pathology, University of Kentucky, Lexington, KY 40546.

Living spores and mycelium from incompatible races of the potato pathogen Phytophthora infestans and Helminthosporium carbonum, a pathogen of corn, elicit accumulation of sesquiterpenoid stress metabolites (SSM) in potato tuber slices. Heat or ethanol-killed spores and mycelium of P. infestans, but not H. carbonum, also elicit accumulation of SSM. A 10 fold increase in heat or ethanol-killed spores and mycelium of H. carbonum added to slices did not elicit accumulation of SSM. Eicosapentaenoic and arachidonic acids, isolated from P. infestans, elicit SSM accumulation. It appears that the elicitor of SSM accumulation from H. carbonum is neither a cell wall polysaccharide nor eicosapentaenoic or arachidonic acids.

A477

TIME-COURSE FOR INHIBITION OF ARACHIDONIC ACID-ELICITED SESQUITERPENE ACCUMULATION IN POTATO BY SALICYLHYDROXAMIC ACID. Carol L. Preisig and Joseph A. Kuć. Dept. Plant Pathology, Univ. of Kentucky, Lexington, KY 40546.

The most rapid accumulation of fungitoxic sesquiteprenes by potato slices in response to arachidonic acid (AA) occurred 48 to 72 hr after treatment. The effect of salicylhydroxamic acid (SHAM), an inhibitor of this accumulation (Stelzig et al., 1983. Pl. Physiol.(72:746-749), applied at various time intervals after treatment with AA, was tested. SHAM inhibited sesquiterpene accumulation when added to potato slices at the same time as AA and up to about 6 hr afterward. This suggests that a process inhibited by SHAM and necessary for AA-elicited sesquiterpene accumulation occurs shortly after AA is applied to potato. Although SHAM is an inhibitor of several metabolic processes, simultaneous addition with AA was found to have no effect on the recovery of AA 6 hr later; recovery was ∿ 50% from control and SHAM-treated tissues.

A478

RESPONSIVENESS OF CUCUMBER LEAVES TO INDUCED SYSTEMIC RESISTANCE AS A FUNCTION OF THE STAGE OF LEAF DEVELOPMENT. X. L. Xuei and J. Kuć, Dept. of Plant Pathology, Univ. of Kentucky, Lexington, KY 40546.

Cucumber plants cv. SMR-58 were inoculated on the first true leaf with Colletotrichum lagenarium. At the time of inoculation on leaf 1, leaf 2 was fully expanded and leaves 3 and 4 were 2/3 and 1/4 expanded, respectively. Seven days after inoculation of leaf 1, leaves 2,3 and 4 were challenged with the fungus. At that time, leaves 2 and 3 were fully expanded and leaf 4 was 2/3 expanded. Resistance was induced in leaves 2, 3 and 4, and the effectiveness of protection was leaf 4>3>2. Excising the apical bud above leaf 4 at the time of inoculation of leaf 1 increased protection of leaves 2,3 and 4, and excising the apical bud above leaf 2 at the time of inoculation of leaf 1 increased protection of leaf 2 when compared to plants with the apical bud intact. The stage of leaf development influences the effectiveness of induced resistance. This may be explained by a sink effect on signal translocation due to rapid leaf expansion and/or to the physiological state of the leaves.

A479

DIFFERENTIAL PRODUCTION OF GLYCEOLLIN ISOMERS IN THE SOYBEAN-PHYTOPHTHORA MEGASPERMA F.SP. GLYCINEA INTERACTION. M.K.Bhattacharyya and E.W.B.Ward, Department of Plant Sciences, University of Western Ontario and Agriculture Canada, Research Centre, London, Ontario, N6A 5B7.

Glyceollin isomers I, II and III were quantitated by HPLC in soybean tissues (cvs. Harosoy, Harosoy 63) with and without challenge by Phytophthora megasperma f.sp. glycinea (race 1) or $AgNO_3$ (10^{-3}M). Small quantities of II and lesser amounts of I were detected in etiolated hypocotyls. Following challenge of both cultivars with race 1 or $AgNO_3$ major increases occurred only in I which reached 5-10 times the levels of II and III by 48 hr. Increases in I occurred much earlier in the incompatible (10 hr) than in the compatible (18 hr) interaction. Similar proportions of the three isomers were found also in green hypocotyls and in roots. However, in green cotyledons relative amounts of II and III were much higher.

A480

PARTIAL STRUCTURAL CHARACTERIZATION OF GLUCANS FROM FIVE RACES OF PHYTOPHTHORA INFESTANS. Jeffrey S. Rush and Joseph Kuć. Dept. of Plant Pathology, University of Kentucky, Lexington, KY 40546.

Glucans from the mycelium and spore germination fluids of Phytophthora infestans are reported to mediate race-specific suppression of the hypersensitive response of potato to P. infestans. These glucans are predominantly β1→3 linked with β1→6 branches and contain 17-23 (Doke, et al., Physiol. Plant Pathol. 15, 117-126) or 14-5 (Ozeretskovskaya, et al., Chem Abstracts, 98 50594) glucose units. Glucans from races 0,1,4, 1.4, and 1.2.3.4 of P. infestans were prepared and their structures partially characterized. Widely different quantities of glucan were obtained from each of the races (race 1, 1 mg/gfwt;

race 4, 18 mg/gfwt). The glucans ranged in size from 19 to 21 glucose units and were predominantly β1→3 linked. Smaller quantities of 3,6 and 2,3 disubstituted β-glucose units were also detected. The number of branched residues was similar from each glucan suggesting that the relative positions of the branches may play a key role in the suppression of hypersensitivity.

A481

PRODUCTION OF VICTORIN BY VARIOUS ISOLATES OF HELMINTHOSPORIUM VICTORIAE AND ITS PHYTOALEXIN ELICITOR ACTIVITY. S. Mayama and N. T. Keen, Lab. of Plant Path., Kagawa Univ., Japan and Dept. of Plant Path., Univ. of California, Riverside 92521.

Victorin occurs as a mixture of related toxins in culture fluids of H. victoriae. Isolates Hv-1, 013 and Rosa produced toxin-active HPLC peaks B, C and D, but filtrates of isolates 1146 and 033 contained little or no peak D. The latter produced another major peak in addition to B and C, but root growth bioassays showed that these culture fluids were ca. 100x less active than fluids from isolates producing peak D. Production of peak D may therefore be characteristic of highly toxigenic isolates. Toxin D purified by HPLC was an efficient elicitor of the oat avenalumins in lines carrying the Pc-2 allele for crown rust resistance and toxin sensitivity. Avenalumin production was elicited in primary Pc-2 oat leaves by the D compound at 5-10 pg ml^{-1}, but concentrations of 10 ng ml^{-1} gave no effect in pc-2 leaves. If Pc-2 leaves were treated with toxin at 1 ng ml^{-1} or more, considerable cell necrosis but relatively little avenalumin accumulation resulted.

A482

WITHDRAWN

A483

WITHDRAWN

A484

PURIFICATION OF FUNGAL ELICITORS. Craig S. Tepper and Anne J. Anderson, Dept. of Biology, Utah State University, Logan, UT 84322.

Cell wall β glucans from Colletotrichum lindemuthianum are potent elicitors, yet they cannot explain race-cultivar specificity. However, on Dark Red Kidney beans extracellular compatible β race culture filtrate components lack elicitor activity but extracellular incompatible α race components are strong elicitors. Therefore, purification and characterization of extracellular elicitor components may aid in our understanding of race-cultivar specificity. Purification on DEAE Sephadex of elicitors from α race culture filtrates has revealed that two distinct types of elicitor structures exist, each exhibiting a different carbohydrate to protein ratio. Elicitors were active at less than 10^{-6}M protein and carbohydrate. The majority of the DEAE Sephadex fractions were inactive as elicitors. Further gel filtration chromatography and polyacrylamide SDS gel electrophoresis of both active fractions suggest that the elicitor not absorbed by DEAE Sephadex was approximately 30,000 daltons and the absorbed elicitor was between 30,000 and 60,000 daltons.

A485

THE DISTRIBUTION AND MECHANISM OF ACTIVATION OF POLYPHENOLOXIDASE IN HEALTHY AND PHYTOPHTHORA INFECTED SOYBEAN HYPOCOTYLS. George Lazarovits and Betty Singh. Agr. Canada, Res. Centre., Univ. Sub P.O., London, Ont., Canada, N6A 5B7.

Polyphenoloxidase (PPO) activity is an early response of etiolated soybean hypocotyls to infection by incompatible races of Phytophthora megasperma f. sp. glycinea. The basis for this reaction was studied by histochemical localization of PPO in healthy and infected tissues. Tissues treated with Dopa after fixation contained PPO in thylakoids and membrane bound inclusions of chloroplasts in epidermal and adjoining cell layers. Those treated with Dopa prior to fixation had reaction product only in necrotic cells, indicating that healthy cells are impermeable to Dopa. Inhibitors of PPO prevented reaction product formation. Penetration by the incompatible race resulted in the death of one or more epidermal and mesophyll cells at 90% of the sites examined 4 hours after inoculation. The compatible race rarely caused cell damage. Activation of PPO thus results from disruption of cellular compartmentalization.

A486

WITHIN-LEAF VARIATION IN RECEPTIVITY OF FOUR WINTER WHEAT CULTIVARS TO ERYSIPHE GRAMINIS F. SP. TRITICI. J. R. Pelletier and R. D. Schein, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Primary leaves of 16-day-old seedlings of Chancellor, Hart, Knox and Titan winter wheats were inoculated with isolate 13 of Erysiphe graminis f. sp. tritici with the aid of a settling tower. Ten days after inoculation, colony numbers were assessed within leaf segments which were 0.00-1.75, 1.75-3.50 and 3.50-5.25 cm from the ligule. Receptivity (colonies/conidium applied) was greatest near the ligule in all four cultivars. Cultivar differences in receptivity (based on whole leaves) were best represented by the segment furthest from the ligule. The decline in receptivity with distance was cultivar-dependent. These data indicate the need to use either whole leaves or identical distal segments for receptivity comparison tests.

A487

THE EFFECT OF HEAT-INDUCED FUNGAL DEATH ON COMPATIBLE AND INCOMPATIBLE INTERACTIONS INVOLVING THE BEAN RUST FUNGUS. Michele C. Heath, Botany Dept., University of Toronto, Toronto, Ontario M5S 1A1, Canada.

Post-inoculation heat treatment of rust-infected, susceptible, French bean leaves resulted in the inhibition of fungal growth, and the encasement of haustoria in callose-like material. Browning of host cells was rare. Heat treatment of rust-infected, incompatible, bean leaves inhibited the normal browning of plant cells if applied soon after the formation of the first haustorium. Later heat treatment had no effect on the subsequent frequency or extent of browning even though fungal growth was curtailed sooner than in unheated tissue. It is suggested that this necrosis is the result of some activity of the living fungus rather than the release of constitutive, toxic components during fungal death.

A488

HOST-PARASITE INTERACTIONS OF UROMYCES PHASEOLI VAR. TYPICA AND U. PHASEOLI VAR. VIGNAE WITH SPECIES OF THE PHASEOLUS - VIGNA PLANT COMPLEX. Janice F. Elmhirst and Michele C Heath. Botany dept., University of Toronto, Toronto, Ontario M5S 1A1, Canada.

Fungal growth and the expression of pre-haustorial defenses to infection were compared in African and American species of the Phaseolus - Vigna plant complex infected with Uromyces phaseoli var. typica (bean rust fungus) and U. phaseoli var. vignae (cowpea rust fungus). The bean rust fungus triggered effective pre-haustorial defenses at relatively few infection sites in Phaseolus vulgaris , P. filiformis and P. acutifolius . Such defenses were seen at 30-50% of infection sites in Vigna unguiculata , V. luteola , V. caracalla and V. vexillata , and at about 70% of infection sites in V. lasiocarpa . Effective pre-haustorial defenses occurred at over 70% of infection sites

of the cowpea rust fungus in all species except V. unguiculata and V. vexillata ; in the latter species, the fungus was inhibited before haustorium formation at less than 15% of infection sites.

A489

THE ROLE OF EPICUTICULAR WAX IN CANOLA IN RESISTANCE TO ALTERNARIA BRASSICAE. K. L. Conn, J. P. Tewari, and D. Hadziyev, Depts. of Plant Science and Food Science, University of Alberta, Edmonton, Alberta, Canada T6G 2P5

As shown by SEM and wax extraction experiments, Brassica napus cultivars (Westar and Altex) have more foliar epicuticular wax than B. campestris cultivars (Candle and Tobin). The wax as analysed by TLC and IR spectroscopy consists of nine classes of compounds. The conidia of A. brassicae germinate more slowly on the leaves of B. napus than on those of B. campestris. Wiping of the leaves of B. napus results in marked acceleration of conidial germination, whereas little such effect is seen in the case of B. campestris. SEM and TEM studies indicate that the wax is crystalline and forms a fluffy layer on the leaf surface. It is postulated that one of the ways wax could offer resistance to A. brassicae is by impeding the diffusion of foliar exudates.

A490

VASCULAR COATING MATERIAL: A RESISTANCE MECHANISM IN VERTICILLIUM WILT OF TOMATO. Jane Robb and Peter F.S. Street. Dept. of Botany and Genetics, University of Guelph, Guelph, Ont. N1G 2W1.

Petioles of near-isolines of Craigella tomatoes that were wilt-susceptible or resistant were precision infused with living or boiled conidial suspensions of Verticillium albo-atrum. The levels of fungal colonization and host response were assayed quantitatively by light and electron microscopy at set time intervals from 3 hours to 5 days post-inoculation. In resistant tomatoes the primary factor resulting in the lateral restriction of colonization was the formation of vascular coating material which prevents fungal penetration of pit membranes and vessel walls. The response, which is non-specific, is suppressed by V. albo-atrum to a limited extent in resistant plants but strongly in the suscept.

A491

PATTERNS OF BARRIER ZONE FORMATION IN PYRUS WOOD TISSUES INFECTED WITH ERWINIA AMYLOVORA. A. L. Shigo, USDA Forest Service, Durham, NH 03824, and T. van der Zwet, USDA, ARS, Appalachian Fruit Research Station, Kearneysville, WV 25430.

Longitudinal dissections of 30 fire blight cankers (E. amylovora), representing scores 1 to 9 on the USDA fire blight scoring system, were collected from 100 6-year-old pear trees (P. communis). Following bark removal from 25 stems, the internal cankers showed that patterns of blight infected wood followed the CODIT model for compartmentalization of decay in trees. Most cankers were associated with branch crotches. When infected leader stems died, the boundary between living and dead tissues farther down on the stem usually occurred above a healthy lateral branch. Further spread of the infection below a healthy branch was limited to wood connected to the leader stem and not to the healthy branch. After the cankers were walled-off, first wood cells formed a barrier zone that separated the infected from the healthy wood. Development of an effective barrier zone appeared correlated with the onset of growth after the dormant period.

A492

HISTOPATHOLOGY OF CUCUMBER CALLUS INFECTED BY COLLETOTRICHUM LAGENARIUM. K.A. Rosenberg, P.A. Morgan, and F.L. Caruso, Dept. of Botany & Plant Path., University of Maine, Orono, 04469

Callus derived from cucumber cultivars SMR-18 (susceptible), GY-14 (resistant), and Poinsett (resistant) was inoculated with a 10 ul drop of a conidial suspension (10^3/ml) of C. lagenarium race 1 and incubated at 22, 28, or 32 C. Callus pieces were sampled at 3-8 days after inoculation, fixed, embedded, sectioned, and stained with safranin-fast green or Conant's multiple stain. At 28 C infection in all cultivars progressed from 1-2 cells to 11-14 cells in depth at 3 and 8 days respectively. Hyphae grew inter- and intracelluarly and acervuli were formed on the callus surface at 6 days. Callus cells in contact with the pathogen lost their integrity and formed loose clusters. Cells adjacent to infected cells had thickened cell walls containing bead-like areas which filled the intercellular spaces and formed a barrier that prevented further invasion by the fungus. There were no apparent differences among the responses of the 3 cultivars to C. lagenarium.

A493

GROWTH AND MICROSCLEROTIAL FORMATION OF VERTICILLIUM DAHLIAE ON STEM SEGMENTS FROM RESISTANT AND SUSCEPTIBLE COTTON VARIETIES. N. A. Garas, S. Wilhem*, and J. E. Sagen*, USDA, ARS, WRRC, 800 Buchanan St., Berkeley, CA 94710 and Dept. of Plant Pathology, University of California, Berkeley, CA 94720.*

A bioassay was undertaken in vitro using excised basal, middle, and apical cross-sections of seedling stem segments of 4 cotton varieties; one Gossypium barbadense (resistant), and 3 G. hirsutum (different levels of wilt tolerance). Individual germinated conidia of the mild strain SS-4 were transferred to stem segments previously placed on water agar in multi-well tissue culture plates. Growth and microsclerotial formation were recorded weekly for 3 weeks. Initital growth of conidia on apical and basal segments from G. barbadense was significantly less than on even the most tolerant G. hirsutum varieties. Transfer to PDA of germinated conidia showing zero growth for 3 weeks showed that they were still viable and capable of rapid growth. Microsclerotial formation on segments from G. barbadense was delayed by at least one week and numbers of microsclerotia were reduced by a factor of 4 to 8 times.

A494

ARE PAPILLAE IN BARLEY COLEOPTILES IMPERMEABLE TO SMALL MOLECULES? M.G. Smart, J.R. Aist, and H.W. Israel. Department of Plant Pathology, Cornell University, Ithaca, NY, 14853.

Impermeability of papillae to ions and small molecules, owing to the presence of a callosic component, has been cited as a possible basis of resistance to fungal penetration. Non-porous papillae could prevent nutrient transfer or pathogen-host recognition. In this first reported study of papilla porosity, living barley coleoptiles, inoculated with Erysiphe graminis hordei were treated with lanthanum ions (139 daltons), fluorescein (332 daltons) and two of its derivatives (386 and 416 daltons), chlortetracycline (CTC 515 daltons) and fluorescently labelled protein A (42,000 daltons). Detection of markers by light and electron microscopy showed papillae to be permeable to lanthanum, to fluorescein and its isothiocyanate and diacetate derivatives and slightly permeable to CTC. Protein A does not penetrate coleoptile cell walls. These papillae are pervious to ions and molecules up to 416 daltons but impervious to molecules larger than 500 daltons.

A495

EFFECTS OF ENVIRONMENT ON RESISTANCE OF INBRED LINES OF CORN TO ISOLATES OF BIPOLARIS MAYDIS AND COLLETOTRICHUM GRAMINICOLA. Anne E. Jenns, Department of Plant Pathology, North Carolina State University, Raleigh 27695.

Six inbred lines of corn were grown under three temperature or three illuminance regimes for one week before and after inoculation in all combinations with four isolates of race 0 of Bipolaris maydis. Lesion length, infection efficiency and sporulation per lesion were measured. A significant interaction between isolate and line was revealed by the analysis of sporulation per lesion for plants grown at the lowest illuminance. Six inbred lines of corn were grown under three illuminance regimes for one week before and after inoculation in all combinations with four isolates of Colletotrichum graminicola. Lesion length, lesions per cm^2, sporulation per lesion and sporulation per cm^2 were measured. No significant ixolate x line interaction effects were observed for any variable under any illuminance condition.

A496

INHERITANCE OF TMV RESISTANCE IN BACKCROSS PROGENY OF SOMATIC HYBRIDS OF NICOTIANA TABACUM AND N. NESOPHILA. S. A. Miller, C. E. Flick, and D. A. Evans. DNA Plant Technology Corporation, 2611 Branch Pike, Cinnaminson, NJ 08077.

Nicotiana nesophila (Nn) is one of a number of wild relatives of N. tabacum (Nt) resistant to tobacco mosaic virus (TMV). However, attempts to introduce TMV resistance from Nn into Nt by conventional sexual hybridization have been unsuccessful. In recent years, the incompatibility barrier between these two species has been overcome by somatic hybridization (Science 213:907-909). Hybrids resulting from the fusion of protoplasts of Nt and Nn were resistant to TMV and at least partially fertile. Backcross populations have been generated for two of the somatic hybrids, using Nt as the pollen parent, and screened for resistance to TMV. Resistance (local lesion type) was expressed in the majority of the BC_1 progenies tested; it appears to behave as a dominant trait, similar to the N gene from

N. glutinosa, the source of TMV resistance in cultivated tobacco. A BC_2 population has been generated and is currently being evaluated for resistance to TMV.

A497

EFFECT OF LOW CONCENTRATIONS OF IMAZALIL ON INFECTION EFFICIENCY AND SPORULATION CAPACITY OF COCHLIOBOLUS SATIVUS ON WHEAT SEEDLINGS. J.P. Hill and C.L. Biles, Dept. of Botany and Plant Path., Colorado State University, Fort Collins, CO 80523.

Four week old seedlings of wheat cultivars Baca, Newton, Scout and Vona were sprayed with various concentrations of imazalil (1-[2-(2,4-dichlorophenyl)-2-(2-prophenyloxy)ethyl]-1 H-imidazole) at the rate of 1 ml per plant. Two days later the plants were inoculated with a Cochliobolus sativus conidial suspension. Six days later lesions were counted, excised, and induced to sporulate in petri dishes lined with moist filter paper. After 6 days, sporulation capacity (SC) was determined as number of conidia per produced lesion and infection efficiency (IE) as number of lesions per plant. Decreasing concentrations of imazalil were used until IE was significantly decreased but not completely suppressed (0.009% AI). SC was not significantly affected at this rate. There was no significant variation in IE or SC among wheat cultivars. The effect of imazalil on the SC reducing capability of Trichoderma harzianum is currently under investigation.

A498

EVALUATION OF SELECTED CORN HYBRIDS FOR REACTION TO CERCOSPORA ZEAE-MAYDIS IN VIRGINIA. E. L. Stromberg, Dept. of Plant Pathol., Physiol. & Weed Sci., VPI&SU, Blacksburg, VA 24061.

Gray leaf spot of corn, caused by Cercospora zeae-maydis, has increased in prevalance and severity in the last 10 yr within the mid-Atlantic and Southeastern United States. This increase is associated with continuous no-till corn production. In 1982, six public and 24 commercial hybrids were evaluated for their reaction to C. zeae-maydis at one farm location. In 1983, seven public and 22 commercial hybrids were tested at two locations. Plots were scored on a 0 (resistant) to 5 (susceptible) scale three times each season. Grain yield and stalk lodging were recorded at harvest. Public hybrids possessing any one of the inbreds Va14, Va59, or B68 had higher levels of resistance than the mean of all hybrids. In 1982, six of 24 commercial hybrids were more resistant ($P \leq 0.05$) while, in 1983, five of 22 were more resistant.

A499

AN UNIDENTIFIED SPECIES OF CERCOSPORA PATHOGENIC TO CORN. F. M. Latterell and A. E. Rossi, USDA-ARS Plant Disease Research Laboratory, Fort Detrick, Frederick, Maryland 21701.

A species of Cercospora isolated from corn leaf debris from a Franklin Co. Pennsylvania field caused a distinctive leaf spot in greenhouse inoculations of several commercial hybrids at 5- to 8-leaf stages. Typical lesions are pale tan in color, and, although limited in lateral spread by major veins, they tend to be more irregular in shape than the rectangular lesions characteristic of gray leaf spot disease caused by Cercospora zeae-maydis. In culture on V-8 Juice agar the fungus produces velvety gray densely sporulating colonies that grow radially faster than most species of Cercospora. Upon exposure to high humidity in a moist chamber, lesions sporulate abundantly both on fascicles of conidiophores from stomata and from free standing conidiophores. Conidia are hyaline, long obclavate, or short cylindrical when catenulate; narrowly truncate at the base, 16-180 um long, 2.4-4.7 um wide, with 1-16 septa. Conidiophores are smoky to light brown, short, sturdy, and geniculate.

A500

SIGNIFICANCE OF INFESTED CORN RESIDUES AS A SOURCE OF INOCULUM FOR ANTHRACNOSE LEAF BLIGHT AND STALK ROT OF CORN. P. E. Lipps, Dept. of Plant Pathology, The Ohio State University/OARDC, Wooster 44691.

Experiments were designed to determine the relative importance of buried and surface residues as sources of inoculum for leaf blight and stalk rot of corn caused by Colletotrichum graminicola. No increase in the incidence of stalk rot was detected when corn seed was placed in the furrow with either infested corn stalks or infested oat kernels, although roots penetrated these residues. Inoculating corn plants at the early leaf whorl stage with conidial suspensions (10^6 ml) or placing infested corn residues on the soil surface in plots consistently increased the incidence of leaf blight and stalk rot. Regression analysis indicated that the the incidence of leaf blight and stalk rot decreased with distance from residue inoculum sources. Results indicate that surface residues and not buried residues, are an important source of inoculum for anthracnose leaf blight and stalk rot.

A501

CHLOROTIC SPOTS ON ZEA MAYS SEEDLINGS INFECTED WITH Sphacelotheca reliana (Head Smut) AND THEIR USE IN SCREENING FOR RESISTANCE. C. A. Matyac and T. Kommedahl. University of Minnesota, St. Paul, MN 55108.

Zea mays hybrids and inbreds were grown in the greenhouse in soil containing 1.8×10^6 head smut teliospores/g soil. After 8 weeks, seedlings were transplanted to the field and those with spotted leaves were tagged. At maturity, each plant was examined for smut. A three-way contingency table indicated the data fit a model with interactions between spot formation and sorus formation ($X^2 = 37.5$, 23 df); 91.5% of the seedlings with chlorotic spots on leaves subsequently produced sori at maturity. Also, the percentage of seedlings with leaf symptoms grown in the greenhouse were compared to the percentage of plants with sori planted in inoculated field plots. Regression analysis of 20 entries (15 hybrids and 5 inbreds) indicate that if 40-50 plants are used, seedling symptoms can be used to predict the degree of resistance in the field (with 1, 2 and 3 replicates, r = .66, .734 and .74, 18 df, respectively).

A502

METHODS FOR STORAGE OF PERONOSPORA TABACINA SPORANGIOSPORES. R. C. Rufty, E. A. Wernsman and G. V. Gooding, Jr., Depts. of Crop Science and Plant Pathology, North Carolina State University, Raleigh 27695.

Peronospora tabacina, causal agent of tobacco blue mold, cannot be grown in culture because it is an obligate parasite. Several treatments were investigated for their efficacy in preserving sporangiospore viability. Sporangiospore concentrations were adjusted to 1×10^6 spores/ml in all treatments and viability, measured as percent germination, was determined prior to storage and at monthly intervals. Sporangiospores stored at -18C in a 50% (w/v) sucrose solution retained viability near the original level (30-40%) for up to 4 months. Germination declined to 10% by the fifth month and to 5% after 8 months. Similar results were obtained when sporangiospores were collected by vacuum onto filters, dessicated over silica gel, and stored at -18C. Sporangiospores stored in distilled water or in 15% DMSO at either -18C or -78C were viable for only 4 months. Viability was lost when sporangiospores were lyophilized in milk, 20% dextrose or water.

A503

VIRULENCE OF CORN STALK ROT FUSARIA FROM COLORADO. R. L. Gilbertson, W. M. Brown, and E. G. Ruppel. Dept. of Botany and Plant Pathology, Colorado State University and USDA, ARS, Crops Res. Lab., Fort Collins, CO 80523.

Corn stalk rot in Colorado can be caused by Fusarium graminearum, F. moniliforme, and F. moniliforme var. subglutinans. A study was conducted in 1983 to assess the virulence of isolates of these three fungi obtained from various sources, including seed, soil, corn stalks, corn rootworm adults (Diabrotica virgifera), and corn stubble. The toothpick inoculation method was used, and virulence evaluated based on percent rot of internode above inoculation point. F. graminearum isolates were most virulent and all caused severe stalk rot. F. moniliforme and F. moniliforme var. subglutinans isolates were less virulent than F. graminearum, and these two fungi caused almost equal levels of stalk rot. There was no correlation between isolate source and virulence. Stalk rot fusaria overwintered in stubble from the previous cropping season, and isolates recovered from this stubble caused severe stalk rot.

A504

RACES OF Phytophthora megasperma f. sp. glycinea, PHYTOPHTHORA ROOT ROT OF SOYBEANS IN NORTHERN ALABAMA. D. A. Collins and R. P. Pacumbaba. Dept. of Natural Resource and Environmental Studies, Alabama A & M University, Normal 35762.

Phytophthora megasperma f. sp. glycinea (Pmg) was isolated for the first time in northern counties of Alabama (Morgan, Madison, and Limestone). Two predominant physiologic races of Pmg, R_2 and $R_{1,1}$, from Morgan and Madison counties, respectfively, were identified by their virulent and avirulent disease reactions on standard soybean differential cultivars (Horosoy, Horosoy 63, Sanga, Mack, Altona, PI 171442,

and PI 103091). Screening and selecting improved soybean germplasm will be for resistance against the identified Pmg races. Other soilborne pathogens isolated and identified from soil and suspected Pmg-infected root samples were Pythium spp., Fusarium spp., Sclerotium rolfsii, and Rhizoctonia solani.

A505

SOYBEAN STEM CANKER IN FLORIDA. F. M. Shokes, AREC, Rt. 3, Box 638, Quincy, FL 32351, T. A. Kucharek, University of Florida, Gainesville, FL 32611, and R. K. Sprenkel, AREC, Quincy.

Soybean stem canker caused by Diaporthe phaseolorum (Cke, & Ell.) Sacc. var. caulivora Athow & Caldwell (DPC) was detected for the first time in Florida in 1983. A survey was made between September 13-23 of 483 fields representing 4,362 ha in 11 north Florida counties. Of the counties surveyed, three had >60% of the hectares with canker, two had >50% and six had none to 18%. In the 1,560 ha of soybeans with canker, 66% had 10% or less of the plants affected, 24% had 11 to 50% affected and 10% had >50% affected. Tests of Koch's postulates using toothpick inoculation of 'Hutton' soybeans revealed 21 DPC isolates which caused cankers and/or death of plants. Ten of these isolates also killed Tracy-M soybeans in the greenhouse.

A506

FACTORS AFFECTING SUGARCANE RUST SEVERITY. Jack C. Comstock and Stephen A. Ferreira, Dept. of Genetics & Pathology, Hawaiian Sugar Planters' Association, 99-193 Aiea Heights Dr., Aiea, Hawaii 96701.

During 1983, the incidence and severity of sugarcane rust were variable on susceptible and intermediate reacting varieties. Using 6 varieties with rust reactions ranging from resistant to susceptible, the effect of plant age and time of year on incidence and severity was determined by planting sugarcane monthly and estimating monthly the rust severity (% top visible dewlap leaf infected) beginning at 2 months of age. Severity increased until plants were 4-5 months old, then decreased with age. For example, susceptible variety H54-775 had 1, 6, 10, 7, and 4% TVD leaves infected on 2-, 3-, 4-, 5- and 6-month-old plants in September. Severity also was seasonal, being greater during those months with a higher percentage of days when the minimum temperature was below 20°C. Four-month-old plants of susceptible variety H54-775 had 19, 1, 9, and 16% TVD leaves infected during March (91% days 15-20°C), June (0 days), September (14% days), and December (73% days), respectively. All other varieties tested had similar trends.

A507

Effect of soybeans double cropped with wheat on subsequent wheat take-all incidence. **F. J. Crowe and W. W. Bockus, Dept. of Plant Pathology, Kansas State University, Manhattan, KS 66506.**

Field tests were established to determine if double crop soybeans would increase wheat take-all. Treatments were cropping regimes (conventional tillage) initiated in the fall of 1979 and included continuous wheat, wheat-double crop soybeans-wheat, and fallow-double crop soybeans-wheat. During the first seeding of each sequence subplots were either non-amended or artifically-infested once with virulent G. graminis var. tritici (GGT) isolates grown on autoclaved oats. Plots were kept free of wheat and weed grasses during the summer. In both wheat seasons artifically-infested plots averaged over 40% whiteheads and non-amended plots averaged below 3%. The presence of soybeans in 1980 did not increase take-all in 1980-81 wheat. Repeated attempts to isolate or indirectly trap GGT from any tissues collected from soybeans grown in infested field soil or lab potting mix failed.

A508

SOIL-BORNE FUNGI ASSOCIATED WITH SORGHUM IN TROPICAL ALUMINUM SOILS IN COLOMBIA. Trevathan, L. E., J. A. Cuarezma-Teran, and L. M. Gourley. Department of Plant Pathology and Weed Science and Agronomy, respectively. Mississippi State University, Mississippi State, MS 39762

A field experiment was conducted on clay soils with different aluminum (Al) concentrations (1.3 and 3.1 milliequivalent/100 g soil) in Santander de Quilichao, Colombia, to determine the occurrence of soil-borne fungi in roots of grain sorghum. Fungi isolated were: Curvularia lunata var. aeria, Phoma macrostoma, Fusarium moniliforme, Trichoderma viridae, Fusarium spp., Penicillium sp., Nigrospora sphaerica, F. lateritium, Absidia sp., Aspergillus niger, F. roseum, Drechslera sorokiniana, F. acuminatum, Papularia sp., and Chaetomium sp. Fungi isolated most frequently from low and high Al soils were: C. lunata var. aeria (47 and 40%), P. macrostoma (33 and 52%), and F. moniliforme (20 and 16%, respectively).

A509

DISPERSAL OF VERTICILLIUM ALBO-ATRUM BY FUNGUS GNATS (BRADYSIA). D. W. Kalb and R. L. Millar, Dept. of Plant Pathology, Cornell University, Ithaca, NY 14853-0331.

Alfalfa plants serving as noninoculated controls frequently developed symptoms of Verticillium wilt in controlled environment chambers. Because fungus gnats (Bradysia spp.) were observed to fly from diseased to healthy plants, individual flies were checked for contamination with V. albo-atrum (Vaa). Two lots of 100 flies each were collected by aspiration into test tubes; the two collections were made 3 mo apart and from different experiments. Each fly was rinsed with 1 ml sterile distilled water, which was then spread over ethanol-streptomycin agar in a petri plate. Vaa was recovered from 33% and 51% of the flies, respectively; colonies per plate were 0 to > 100. Newly hatched adults from flies reared on Vaa grown on Brewers' yeast-prune agar were transferred first to Vaa cultures for 16-24 hr and then in lots of 10-12 to individual, caged alfalfa plants. Vaa was isolated from 20 of 64 plants. Control plants did not become infected.

A510

ETIOLOGY OF FUSARIUM WILT AND ROOT ROT OF CHICKPEAS IN SOUTHERN SPAIN. R. Jimenez-Diaz and A. Trapero Casas, Depto. de Patologia Vegetal, ETSIA, Univ. de Cordoba, Cordoba, Spain.

Disease surveys in 1979-81 indicated that chickpeas in Southern Spain were severely affected by Fusarium wilt and root rot. An annual average yield loss of about 12% was estimated. F. oxysporum (Fo) induced vascular wilt, vascular yellowing, or nonvascular yellowing and cortical collar-root rot. F. solani (Fs) induced nonvascular yellowing, and severe black root rot. Vascular wilt and yellowing were the most prevalent symptoms in the field. Isolates of Fo that induced vascular wilt or yellowing were pathogenic to chickpeas but not to alfalfa, bean, broadbean, lentil, Lupinus albus, L. angustifolius, L., luteus, L. mutabilis, pea and soybean. Isolates of Fs were pathogenic to the above legumes except alfalfa and soybean, which, however, became infected. Those most severely affected were chickpea > broadbean > pea. Isolates of Fo that induced vascular yellowing in a local chickpea cultivar were not pathogenic to cv JG-62 which was highly susceptible to the vascular wilt isolates.

A511

COTTON SEEDLING SURVIVAL AS AFFECTED BY TILLAGE AND COVER CROP. D. H. Rickerl, J. T. Touchton, and W. B. Gordon, Agronomy and Soils, Auburn University, AL 36849.

Field and greenhouse studies were conducted to determine the effects of tillage and legume cover crop on cotton stands and yield. In the greenhouse test, seedling survival was 83, 88, and 91% with mean dry weights of .11, .12, and .12 g/seedling for clover, vetch, and fallow soils, respectively. Field stand reductions were 26, 4, and 0% in the no-till compared to till plots for clover, vetch, and fallow areas. Although no-tillage systems resulted in lower plant populations, yields among tillage treatments did not differ. Lint yields from the clover soils (558 lb/acre) were higher than yields from the vetch (460 lb/acre) and fallow (449 lb/acre) soils. Field tests conducted on a Dothan Soil were initiated to determine the cause of seedling failure and to establish the optimum management system for cotton seedling survival and lint production. Starter fertilizer and EDB increased yields 464 lb/acre in tilled plots and 711 lb/acre in no-till treatments.

A512

SUSCEPTIBILITY OF NATIVE PLANTS TO THREE SOIL BORNE FUNGI ENDEMIC TO THE SOUTHWESTERN UNITED STATES. **Rotkis, P.T. and S.M. Alcorn, Department of Plant Pathology, University of Arizona, Tucson, AZ 85721.**

Because they are potential sources of hydrocarbons, Amsonia grandiflora, Asclepias albicans, A. lineria, A. subulata, Chrysothamnus paniculatus, Grindelia camporum, and Xanthocephalum gymnospermoides (all native to the southwestern United States) were tested for susceptibility to three soil borne fungi which occur in this area. In the greenhouse, replications of 20 plants of each group were inoculated after emergence by soil drench with mycelial suspensions of Pythium aphanidermatum, Macrophomina phaseolina, or Rhizoctonia

solani. Inoculated plants were observed over 5 wk, unless symptoms developed earlier. This experiment was repeated once. All plant species were susceptible to M. phaseolina and P. aphanidermatum while all but A. grandiflora were susceptible to R. solani. Susceptibility varied with plant age.

A513

TOBACCO BLACK SHANK CONTROL WITH METALAXYL AND CULTIVARS. L. J. Herr and P. Sutton, Departments of Plant Pathology and Agronomy, The Ohio State University/OARDC, Wooster, OH 44691.

In a greenhouse test of metalaxyl placement, 0, 1.1, 2.2 and 4.4 kg a.i./h were applied (drench) to 'Ky 14' tobacco plants in 10-cm-diam pots of autoclaved soil mix. Plants with rootballs were then inserted into 7.6 L-containers of soil infested with Phytophthora parasitica f. sp. nicotianae. Other plants were drenched with metalaxyl (same rates) and then stem-wound-inoculated. Disease ratings (DR, 0=healthy, 4=dead) of this infested-soil series were 4.0, 3.3, 2.5, and 1.0 with increasing metalaxyl rates, whereas, all treated stem-inoculated plants had low disease (DR=4.0, 0.3, 0.3 and 0). In a field test, main treatments were 0, 1.1 and 2.2 kg a.i. metalaxyl/h, with the 1.1 kg rate applied in a band over the row and the 2.2 kg rate broadcast. Sub-plots were five burley cv: Ky 14, susceptible to race 0 and 1; Ky 14 x L8, susceptible to race 1; and Ky 17, Clay 501, Va509, all tolerant to both races. Data were expressed as % dead and diseased plants. The combination of tolerant cv and broadcast metalaxyl gave the best control.

A514

TRANSLOCATION OF METALAXYL IN SOYBEAN PLANTS AND CONTROL OF STEM ROT CAUSED BY PHYTOPHTHORA MEGASPERMA F. SP. GLYCINEA (PMG). J. P. Gupta, D. C. Erwin, J. W. Eckert and A. I. Zaki. Department of Plant Pathology, University of California, Riverside, CA 92521.

When hypocotyls of soybean plants from seed treated with 200 mg metalaxyl/100 g seed were wound-inoculated with PMG, 100% of the plants survived; at 100 mg, 80%; at 60 mg, 60%; at 30 mg, 40%, and at 0 mg none survived. In 7-day-old plants grown from seed treated with 200 mg metalaxyl, 79% was recovered by GLC in the cotyledons, 21% in the leaves and stems, and none in roots. When 7-day-old plants grown from seed treated with ^{14}C-metalaxyl were extracted and assayed by thin layer chromatography, 40% of the radioactivity corresponded to rf of metalaxyl. Of the ^{14}C absorbed by the plant from treated seed, 91% accumulated in the cotyledons, 6% in the other aerial parts, and about 3% in the roots. After application of ^{14}C-metalaxyl to a cotyledon or to a leaf, over 99% of the ^{14}C remained in the cotyledon or leaf. After soil drench, 81% of the ^{14}C was detected in cotyledons, 14% in the other aerial parts, and 4% in the roots.

A515

STUDIES ON THE IN VITRO AND IN VIVO ANTIFUNGAL ACTIVITY OF FOSETYL-AL AND PHOSPHOROUS ACID. M. E. Fenn and M. D. Coffey, Department of Plant Pathology, University of California, Riverside, CA 92521.

In a low-phosphate medium fosetyl-Al showed a much higher activity in vitro against Phytophthora than previously reported in the literature. Both fosetyl-Al, and more particularly phosphorous acid (PA), were highly inhibitory in vitro against several species of Phytophthora. PA was much less inhibitory in vitro against Pythium and had only low activity against a selection of non-oomycetous fungi. The EC_{50} values for an isolate of P. cinnamomi, cultured on a low phosphate medium, were 0.05 PO_3 meq/L of PA (4 µg/ml) and 0.45 PO_3 meq/L of fosetyl-Al (54 µg/ml). An increase in the level of phosphate reduced the inhibition of mycelial growth due to fosetyl-Na, but there was little or no effect of phosphate on the inhibition caused by PA. In vivo, either 12.7 PO_3 meq/L of PA (1.0 g/L) or 12.7 PO_3 meq/L of fosetyl-Al (1.5 g a.i./L), applied as a foliar spray or soil drench, gave equivalent control of root rot of Persea indica seedlings caused by Phytophthora cinnamomi.

A516

EVALUATION AND USE OF SLOW-RELEASE PROPICONAZOLE FORMULATIONS IN CONTROLLING PHYMATOTRICHUM OMNIVORUM ON COTTON. T. M. Small and S. D. Lyda, Department of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843

The effective use of soil-applied fungicides to control Phymatotrichum omnivorum on cotton has been marginal because the fungus attacks cotton late in the season and the persistence of organic fungicides in the soil is limited. A slow-release system was developed for propiconazole using starch as the entrapping matrix. Starch xanthide-propiconazole or starch borate-propiconazole formulations were synthesized in a ratio of 20 parts starch to one part propiconazole. The formulations were evaluated for persistence in soil, phytotoxicity and efficacy in controlling Phymatotrichum root rot of cotton. The starch matrix degrades up to 80% (dry wt) over 2 wks in the soil (28 C), releasing up to 69% of the propiconazole as determined by monitoring 14C-labeled propiconazole. The slow-release formulations of the fungicide reduced growth inhibition to cotton seedlings commonly seen with the emulsifiable concentrate (0.43 g a.i./ml) and granular formulations (2.5% w/w) in the greenhouse.

A517

THE EFFECT OF METALAXYL AND EFOSITE-AL APPLIED THROUGH THE DRIP IRRIGATION SYSTEM ON PHYTOPHTHORA PARASITICA IN THE SOIL AND ON THE YIELD OF NAVEL ORANGES. E. Pond, J. A. Menge and H. D. Ohr, Department of Plant Pathology, University of California, Riverside, CA 92521, and J. E. Pehrson, Extension Service, University of California, Lindcove Field Station, Exeter, CA 93221.

Navel orange trees on sweet orange rootstock at 3 locations in southern California with histories of Phytophthora root rot treated through the drip system with metalaxyl (1.25 g a.i./m^2/applic.), or Efosite-Al (14 g a.i./m^2/applic.), were compared to untreated trees. Eight trees/treatment were treated monthly from May to October for 3 years at each of two locations and 2 years at one. During this period the rhizosphere populations of Phytophthora parasitica were reduced 48-71% by metalaxyl and 34-38% by Efosite-Al. Root length was increased by 178% by metalaxyl in a grove treated for 3 years, but neither fungicide significantly increased root length in the other two groves. Yield was increased 37-59% by metalaxyl and 20-88% by Efosite-Al in 2 groves treated for 3 years. Fruit size was also increased by the fungicide applications.

A518

SENSITIVITY OF RHIZOCTONIA SOLANI TO EXPERIMENTAL FUNGICIDE NTN19701. D. L. Roberts and C. T. Stephens, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

Laboratory bioassays were used to determine the efficacy of experimental fungicide NTN19701 against Rhizoctonia solani. Seventy isolates of R. solani, representing five anastomosis groups, were obtained from different regions of the United States and tested on potato-dextrose agar (Difco) amended with 0, 1, 10, and 100 µg/ml of NTN19701 or pentachloronitrobenzene (PCNB). Fifty percent of the isolates exhibited 90-100 percent growth inhibition by 1 µg/ml NTN19701. While NTN19701 was generally effective at lower concentrations than PCNB, growth inhibition by NTN19701 was highly variable and ranged from 0 to 100%. This highly variable sensitivity of R. solani isolates to NTN19701 was not correlated with specific anastomosis groups but may explain contradictory results obtained in field, turfgrass and greenhouse testing of NTN19701 for management of R. solani-incited diseases.

A519

CONTROL OF FUSARIUM YELLOWS OF CELERY IN AN ORGANIC SOIL. R. T. Awuah and J. W. Lorbeer. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Methyl bromide-fumigation (1.36 kg/9.3 m^2; 3 lb/100 ft^2) of an organic soil naturally infested with Fusarium oxysporum f. sp. apii eradicated the pathogen from the soil and celery root pieces in the soil. Celery seedlings and transplants grown for 4 and 12 weeks respectively in the fumigated soil were disease free. Autoclaving (120 C, 6 hr) the soil eradicated the pathogen and increased growth of celery seedlings compared to the control. The growth response resulted from death of the pathogen as well as increased availability of nutrients. Pasteurization (70 C, 30 min) of the soil also killed the pathogen. Celery seedlings (3 1/2 weeks old) transplanted to the pasteurized soil grew better than those transplanted to the autoclaved soil. However, when direct seeded, seedlings grew better in autoclaved soil than in pasteurized soil. Levels of soluble salts in the soil released by autoclaving were significantly higher than levels released by pasteurization.

A520

THE USE OF METALAXYL TO CONTROL PHYTOPHTHORA ROOT ROT OF GINSENG (PANAX QUINQUEFOLIUM). M.L. Putnam and J.E. Mitchell. Dept. of Plant Pathology, Univ. of Wisconsin, Madison WI 53706.

Commercial ginseng production in north-central Wisconsin is at times limited by a devastating root rot caused by Phytophthora cactorum. To determine if metalaxyl would be efficacious in

controlling this disease, a single application of Ridomil 5G was made in June to beds known to be infested the previous season. Two plots were laid out in a latin square design with 4 replicates per plot; treatment levels were 0, 0.06, 0.12, and 0.25 kg a.i./ha. Each plot measured 3 x 1.8 m, with 1 m between plots within the same bed and 0.3 m of aisle between plots. Disease incidence was determined in August and September; and in October yield (dry wt. of roots) was assessed. Reduction in mean disease incidence was 55, 56, and 66% at 0.06, 0.12, and 0.25 kg a.i. of metalaxyl/ha, respectively. Metalaxyl at 0.25 kg a.i./ha increased yield by as much as 50% over non-treated controls.

A521

CONTROL OF PHYTOPHTHORA ON SWEET ORANGE ROOTSTOCK USING SYSTEMIC FUNGICIDES. L. W. Timmer and W. S. Castle, Univ. of Florida, Citrus Research and Education Center, Lake Alfred, FL 33850.

Sweet orange is a valuable rootstock, but its susceptibility to Phytophthora foot rot and root rot impedes its widespread use. In a 3-yr study, metalaxyl and fosetyl were evaluated as soil drenches, trunk paints, and foliar sprays for control of Phytophthora in newly planted Pineapple sweet orange on Ridge Pineapple sweet orange rootstock. After application of metalaxyl as a soil drench or trunk paint, fungicidal activity, as determined by bioassay, persisted for 3-4 mo in twigs and roots during the first 2 yr. In the third year, activity was less probably because of dilution of the fungicide in the larger trees. Fungicidal activity was less when metalaxyl was applied as a foliar spray. Application of fosetyl by all methods only occasionally produced fungicidal activity detectable by bioassay. Both fungicides reduced foot rot incidence, but none of the treatments increased growth of the trees compared to nontreated controls, indicating that root rot was of minor importance.

A522

CHEMICAL CONTROL OF PHYTOPHTHORA CROWN ROT OF PEPPER IN FLORIDA. D. F. Myers and R. Subramanya, University of Florida, Everglades Research and Education Center, Belle Glade, FL 33430.

Foliar applications of Aliette 80W (Fosetyl-Al) to bell pepper (Capsicum annuum L.) prior to inoculation reduced the rate of crown rot development caused by Phytophthora capsici Leonian in greenhouse tests. The most effective treatments of Aliette (4.3 & 5.8 g a.i./l) were applied at 1, 2, & 3 da before transplanting peppers into naturally infested soil and at 4-6 da intervals after inoculation. In one field test, Aliette applied to run-off on a 10-da schedule beginning 1 da before inoculation significantly reduced disease development at 2.9g a.i./l but not at 3.6 or 5.8 g a.i./l. Comparable applications of Difolatan 80W (sprills) (2.9 & 5.8 g a.i./l) were significantly better than the two higher Aliette treatments but not significantly different from Aliette applied at 2.9 g a.i./l. The relative efficacy of Ridomil 2E, Previcur N, and Curzate in this test will also be discussed.

A523

CONTROL OF PHYMATOTRICHUM ROOT ROT OF COTTON BY THE BASIPETAL TRANSLOCATION OF PROPICONAZOL. Whitson, R.S., and R.B. Hine. Department of Plant Pathology, University of Arizona, Tucson, AZ 85721.

Field trials in 1982 determined that foliar applications of propiconazol (0.56 kg/ha) provided significant control of Phymatotrichum Root Rot of cotton. In 1983, leaves of 5-wk-old container-grown cotton plants were treated with C-14-labeled propiconazol to determine whether this control was the result of basipetal translocation of the fungicide. Plant tissues were completely oxidized to carbon dioxide, and the C-14 analyzed by liquid scintillation. Over an 8-wk period, 0.07-0.23% of the applied radioactivity was translocated to the roots. Using these percentages, the concentration of propiconazol in the roots from a foliar application of 0.56 kg/ha was calculated to be 1.52-4.31 ug/g, or more than 250X the in vitro concentration necessary to inhibit mycelial growth of Phymatotrichum omnivorum (EC-50 = 0.03-0.06 ug/ml). These findings indicate that the control achieved from foliar applications resulted from the translocation of propiconazol to the roots.

A524

PRODUCTION OF SPOROPHORES OF ARMILLARIA MELLEA IN ISOLATED AND PURE CULTURE. R. D. Raabe, Department of Plant Pathology, University of California, Berkeley, CA 94720.

Armillaria mellea was established on moist sterilized pieces of fruit tree (cherry, apple, pear, etc.) branches approximately 1.9 to 3.2 cm in diameter x 10 cm in length in gallon jars covered with cheesecloth-covered cotton tops, and on sterile ground fig wood in 1 and 2 liter flasks sealed at the top with cigarette paper or covered with cheesecloth-covered cotton tops. When exposed to external temperatures in the shade, the fungus produced sporophores in early winter at approximately the same time that the fungus sporulated on naturally infected plants. To simulate this is the laboratory, the fungus was established on similar branch pieces in 1892 ml jars with cheesecloth-covered cotton tops and put into a growth chamber set at 20 C for 12 hr and at 8 C for 12 hr. These temperatures were chosen because they respresent the average mean daily day temperature and the average mean daily night temperature for the 2-month period prior to the time when sporophores are produced naturally. From sporophores, viable basidiospores were produced and from these some single spore cultures were grown.

A525

INFECTION STRUCTURES OF PLASMOPARA HALSTEDII ON SUNFLOWER SEEDLING ROOTS AND IN CELL SUSPENSION CULTURES. A. B. Gray, W. E. Sackston, and Louis Thauvette. Department of Plant Science, Macdonald College of McGill University, 21111 Lakeshore Road, Ste. Anne de Bellevue, Que., Canada H9X 1C0.

Infection structures produced on radicles of 2-day-old sunflower seedlings by three races of Plasmopara halstedii appeared similar when examined by scanning electron microscopy. Germ tubes produced by zoospores encysted in the root hair zone were short and straight, those in the zone free of root hairs were longer and twisted. Flattened appressoria formed at the ends of germ tubes in both zones. Infection structures were also seen apart from host tissues in cell suspension cultures. Cells derived from a susceptible sunflower cultivar and grown in culture 6 months were washed three-times in sterile distilled water. Washed cells (0.5 mL packed volume) were resuspended in 10 mL water containing 30000 zoosporangia per mL and incubated in 90 mm plastic petri dishes 24 h at 15C, then fixed for scanning electron microscopy. Zoospores adhering to the petri dishes germinated by long twisting germ tubes which swelled at the ends to form appressoria, a few of which developed a peg.

A526

RESISTANT ALFALFA PLANTS AS SYMPTOMLESS CARRIERS OF VERTICILLIUM ALBO-ATRUM. B.W. Pennypacker, K.T. Leath, and R.R. Hill, Jr., Penn State Univ. and U.S. Reg. Pasture Res. Lab., University Park, PA 16802.

Resistant (Vertus, NAPB 110, NAPB 108, CW 8015, WL 316, Apollo II) and susceptible (Cimmaron, Saranac AR) alfalfa varieties were grown in the greenhouse, inoculated with V. albo-atrum by spraying the stubble with conidia, and evaluated for percent symptomless plants. Eighty plants were tested per variety and were harvested 4 times at 42-day intervals prior to assignment of symptomless status. Seven months after inoculation, symptomless plants were checked for V. albo-atrum by stem isolations. Distribution of symptomless plants was: Vertus-54%, NAPB 108-54%, NAPB 110-48%, Apollo II-36%, CW 8015-36%, WL 316-35%, Cimmaron-22%, and Saranac AR-10%. All symptomless plants in the varieties Vertus, NAPB 110, Apollo II, CW 8015, WL 316, Cimmaron, and Saranac AR and 95% of the plants in NAPB 108 were infected with the pathogen. Absence of symptoms is not a reliable indicator of a Verticillium-free alfalfa plant.

A527

GROWTH OF RESISTANT VARIETIES OF ALFALFA INFECTED BY VERTICILLIUM ALBO-ATRUM. B.W. Pennypacker, K.T. Leath, and R.R. Hill, Jr., Penn State Univ. and U.S. Reg. Pasture Res. Lab., University Park, PA 16802.

Resistant alfalfa varieties (Vertus, NAPB 110, NAPB 108, CW 8015, WL 316, Apollo II) were evaluated to determine their response to V. albo-atrum (VAA). Eighty greenhouse-grown plants per variety were stubble inoculated with VAA conidia and harvested 4 times at 42-day intervals. Plot dry weight, plant height, number of stems per plant, and disease rating were recorded at each harvest; flowering data were noted during the 4th harvest. Number of stunted and dead plants was significantly higher in the VAA treatment. Plant height was significantly less in the inoculated plants as compared to control plants. Height was also significantly reduced in symptomless, unstunted inoculated plants. Plot dry weight and number of stems per plant were not affected. Flowering was significantly reduced in infected plants regardless of variety. Results suggest that VAA can elicit modification of several growth parameters in resistant plants regardless of symptoms.

A528

PREVALENCE AND PATHOGENICITY OF PYTHIUM PAROECANDRUM ON ALFALFA IN CALIFORNIA. J. G. Hancock, Department of Plant Pathology, University of California, Berkeley 94720.

Pythium paroecandrum was the most common member of its genus isolated from feeder-rootlets in surveys of alfalfa fields during the cooler months in the Central Valley of California. In January and April, infections averaged 6 (range: 0 to 14) and 11 (range: 1 to 26) per 100 cm of rootlets, respectively. Where successive samples were taken, frequencies of isolation of P. paroecandrum were reduced during midsummer. Other Pythium spp. (e.g. P. ultimum and P. vexans) were isolated more commonly in the warmer months. At similar inoculum densities, P. paroecandrum did not cause as severe damping-off as P. ultimum. However, shoot growth and development were strongly inhibited when alfalfa (cv. Moapa 69) transplants were grown in fumigated soils reinfested with either species. Thus, P. paroecandrum is a prominant rootlet-infecting fungus of alfalfa which may cause economic losses.

A529

THE EFFECT OF HOST GROWTH STAGE AND INOCULATION TECHNIQUE ON INFECTION OF WHEAT BY TILLETIA INDICA. M.H. Royer and J.L. Rytter, USDA-ARS PDRL, Ft. Detrick, Bldg. 1301, Frederick, MD 21701.

Primary and secondary sporidial inoculum was produced by germinating T. indica teliospores from Sangrur, India and the Yaqui Valley in Mexico on water agar petri plates at 20 C over 10-20 days. The spores were dislodged in distilled water, and the suspension was filtered through a 60 µm sieve. The inoculum was calibrated to contain $3x10^3$ to $3x10^5$ primary and secondary sporidia per ml. The wheat plants were inoculated either by injection into the boot or atomization with inoculum and then placed in a misting tent for 4 days at 20 C. The plants were examined for infection 3-5 wk after inoculation. Either injection of inoculum into the boot or atomization prior to anthesis yielded infection of 10-40% of all the plants that were inoculated. The average percent infection of each infected spike varied between 35-85%. Wheat cultivars Butte, Olaf, Waldron, Alex, Chris, and Len were all susceptible.

A530

SYSTEMIC INFECTION OF MAIZE SEEDLINGS BY ASPERGILLUS FLAVUS Sandra M. Kelly and Jack R. Wallin, 312 Curtis Hall, Univ. of Missouri, Columbia, 65211.

Systemic growth of Aspergillus flavus in plant tissue has been demonstrated by culturing viable mycelia from 8 segments of maize seedlings. Kernels were decapped, surface sterilized with 1% NaOCl, plated with an inoculum density of 3,000 conidia/ml, and incubated 13 days at 26°C. Uninoculated checks were grown along side inoculated seedlings. Infected kernels were again surface sterilized and scrubbed of all visible mycelia and conidia, planted in steamed soil and grown in growth chambers. Germination rate and seedling survival were recorded and surface sterilized tissue plated on Aspergillus Differential Media. Fungal mycelia grew from plant tissue segments in 71% of the leaves, 79% of the stems and 75% of hypocotyls and roots of the inoculated plants. Systemic infection may occur through the life cycle of the plant and ultimately cause infection and contamination of the carbohydrate sink - the ear and kernels.

A531

TEMPERATURE REQUIREMENTS BY ISARIOPSIS GRISEOLA (IG) FOR INFECTION AND DISEASE DEVELOPMENT ON RED KIDNEY BEANS. D. A. Inglis and D. J. Hagedorn, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Angular leaf spot has been serious in red kidney bean growing areas of Wisconsin for the last 3 yr. A temperature series was used to determine whether temperature requirements for infection and disease development are different. Infection occurred most rapidly at 24 C followed by 20 C, and was lowest at 16 & 28 C. Expansion of lesions was most rapid at 24 C followed by 20 & 28 C, and was lowest at 16 C. Ratings for leaf chlorosis were higher and days to defoliation were fewer when infection was allowed to occur at 16, 20, & 24 C and when the disease was allowed to develop at 20, 24, & 28 C than when the same temperatures were maintained throughout infection and disease development. Although cooler temperatures favor infection and warmer temperatures favor disease development, cool followed by warm conditions in the field may lead to greatest disease severity.

A532

CYTOLOGICAL EVENTS DURING THE COURSE OF APPRESSORIUM DEVELOPMENT IN UREDOSPORE GERMLINGS OF UROMYCES PHASEOLI VAR. TYPICA. T. M. Bourett, H. C. Hoch and R. C. Staples*, New York State Agr. Exp. Sta., Cornell University, Geneva, NY 14456 and *Boyce Thompson Institute for Plant Research, Ithaca, NY 14853.

Uredospore germlings proceed through a specific series of biochemical and morphological events during appressorium formation. The exact timing and interrelationships of certain events (e.g., nuclear migration, ballooning of the germling tip, mitosis, septum formation) have not been well documented. Also, the involvement of the microtubule (MT)-microfilament (MF) cytoskeleton in this cell differentiation event has not been elucidated. Using immunofluorescence microscopy and TEM, we determined that the germling tip balloons laterally upon induction for cell differentiation, accompanied by a dispersal of the apical vesicles and a reorientation of MT's and MF's along the inducive scratch. DNA synthesis is timed closely with this event. Mitosis was observed later, only in the differentiating germling tip. Septum formation did not begin until anaphase-telophase. MT's were not seen in the mature appressorium.

A533

INCIDENCE AND DISSEMINATION OF THE TALL FESCUE FUNGAL ENDOPHYTE. M. R. Siegel, M. C. Johnson, D. R. Varney and R. C. Buckner. Plant Pathology and Agronomy Departments University of Kentucky, Lexington, Ky., 40546.

Thirty seven fields in 4 Kentucky counties were surveyed for the tall fescue endophyte in 1982. Distribution of infested fields by percent were 10-33% (10 fields), 34-66% (6 fields) and 67-100% (21 fields). There was no correlation between the age of the stand and incidence. The endophyte is disseminated by seed via the maternal parent. No significant changes in the incidence of endophyte occurred during 4 years in experimental plots and fields managed for seed production (SP). This suggests that the endophyte is not disseminated by wind, rain or mowing. Higher endophyte levels were found in SP plots than those managed for hay-pasturage (HP). Hay yields and seed production in Kenhy tall fescue HP and SP plots were independent of the incidence of the endophyte. The origin of the endophyte in tall fescue and the mutualistic or commensal relationship between fungus and host grass will be described.

A534

DEVELOPMENT OF LOOSE SMUT GALLS IN BARLEY. E. S. Luttrell, Department of Plant Pathology, University of Georgia, Athens, GA 30602.

Florets in six-rowed barley are borne in triplets with one terminal floret and two lateral florets on a short rachilla. Differentiation of galls on plants infected with Ustilago nuda is apparent at an early stage when stamens and pistils are recognizable but are not yet enclosed by lemma and palea initials. Differentiation of florets is arrested at this stage. Hypertrophy of the rachilla and its branches produces the greater part of the three-lobed gall. The tips of the lobes are formed by the hypertrophied lemmas. The aborted florets occupy shallow recesses in the adaxial surfaces of the lemmas. Awns on lemmas and glumes are reduced but still conspicuous. The mycelium, which is inter- and intracellular, develops within gall tissues derived mostly from the rachilla cortex. The epidermis forms a membrane over the spore mass.

A535

EFFECT OF BLAST FURNACE SLAG (BFS) AND POTASSIUM (K) ON THE GROWTH AND RESISTANCE OF RICE TO BROWN SPOT DISEASE. K. L. Khew[1], N. P. Lee[1], M. D. Coffey[2] and G. A. Zentmyer[2], [1]Sch. of Biol. Sci., Univ. Sains Malaysia, Penang, Malaysia and [2]Dept. of Plant Pathology, Univ. of California, Riverside, CA 92521.

Different levels of blast furnace slag (0, 7.5, 15, 30 g) were applied with 3 levels of K (0.5, 1, 2 g K_2O) to 6.8 kg of soil in pots to study their effects on growth and resistance of rice plants to leaf spot infection caused by Helminthosporium oryzae. Results indicated that different levels of BFS and K affected both yield and disease resistance. In general, the higher levels of BFS and K increased disease resistance to brown spot and produced a higher weight and number of grains and mean yield per head as well as an improved height and dry weight of plants than the lower levels. Larger numbers of silicified cells and a thicker cuticle and epidermal wall were also observed in plants treated with the highest levels of BFS and K. Since BFS increased both yield and resistance to brown spot and is readily available in Malaysia, we recommend its inclusion in a routine rice planting together with judicious use of other fertilizers.

A536

A COMPARISON OF INOCULATION METHODS FOR KARNAL BUNT, NEOVOSSIA INDICA. E. J. Warham, Centro Internacional de Mejoramiento de Maiz y Trigo, Londres 40, Apdo. Postal 6-641, 06600 Mexico, D.F. Mexico.

Two inoculation techniques for Neovossia indica are compared: 1) boot inoculation--injection of a water suspension of secondary sporidia with a hypodermic syringe into the boot and 2) spray inoculation--a water suspension of secondary sporidia sprayed at various growth stages between heading and anthesis. Both inoculation techniques were investigated using susceptible cultivars to determine the optimum inoculum concentrations, ideal plant growth stage and humidity requirements for successful infection. Boot inoculation required no humidity and gave reliable infection with low secondary sporidia concentrations (1,000-10,000/ml). The ideal plant growth stage for inoculation was mid-boot. In contrast, spray inoculation required high secondary sporidia concentrations (50,000/ml) and 48 hours of humidity, but infection was initiated over a range of growth stages.

A537

PROGRESSION OF PHYMATOTRICHUM OMNIVORUM ON COTTON CULTIVARS AND EFFECT ON YIELD AND FIBER QUALITY. K. M. El-Zik, L. S. Bird, and P. M. Thaxton, Dept. of Plant Pathology and Microbiology, Texas A&M University, College Station, TX. 77843.

Twelve cotton cultivars were evaluated for root rot, yield and fiber quality in three tests in the Phymatotrichum root rot nursery at Temple, Texas during 1982 and 1983. Progression of root rot was slower in 1983 than in 1982. A higher incidence of dead plants (54%) was obtained in 1982 compared to 40% in 1983, 128 days from planting. The most critical period for P. omnivorum infection in the field was from mid-July to mid-August. Significant differences in root rot, yield and fiber quality among the 12 cultivars were obtained. Range of percent root rot among cultivars was 15-83%. Correlation coefficients between percent dead plants and yield, earliness and fiber traits were computed. Increased severity of the disease was highly correlated with decreased yield, lower gin turnout and fiber fineness. Maturity was positively correlated with root rot. P. omnivorum did not influence fiber length or strength.

A538

THE TELEOMORPH OF RHIZOCTONIA ORYZAE. P. S. Gunnell and R. K. Webster, Department of Plant Pathology, University of California, Davis, CA 95616.

Bright orange hymenia of the teleomorph of Rhizoctonia oryzae Ryker and Gooch, the causal organism of bordered sheath spot of rice, were found encircling diseased rice sheaths just above the water line in California. This is the first description of the sexual state of R. oryzae and the first report of the occurrence of bordered sheath spot in CA. The teleomorph is morphologically similar to Waitea circinata Warc. and Talb. Fructifications are resupinate, waxy-pruniose, and easily detached from the substratum. Basidia are cylindraceous, often bent to one side, and often constricted at one or more points, at times appearing somewhat urniform, (4.8-)6.8(-9.4) X (21.5-) 27.5 (-38.0) µm. Basidia bear 4 stout, hornlike sterigmata, 2-3 X (6.5-)8.4(-10.5) µm. Basidiospores are smooth, hyaline, broadly ellipsoid with one side usually flattened, with a prominent truncate apiculus, (4.5-)5.7(-7.4) X (7.0-)9.5(-11.7) µm. No repetitive spore germination has been observed.

A539

SEEDBORNE INFECTION OF COWPEA DETERMINED BY LYSOTYPE DISTRIBUTION OF XANTHOMONAS CAMPESTRIS PV. VIGNICOLA. R. D. Gitaitis and D. K. Bell, Department of Plant Pathology, University of Georgia, Tifton, GA 31793-0748.

Two lysotypes of Xanthomonas campestris pv. vignicola equal in virulence to cowpea were used to study seed-borne infection and movement of inoculum in field plots of Mississippi Silver. Lysotype A was used as inoculum in nontreated plots and lysotype B was distributed in plots treated with copper ammonium carbonate (0.65 kg ai/ha). Although the bactericide significantly reduced bacterial leafspot (38.5% in nontreated plants versus 1.5% in treated plants), high levels of seed-borne bacteria were detected in seed from copper-treated as well as nontreated plants. Bacteria recovered from seed harvested from symptomless plants in copper-treated plots were identified as lysotype A indicating interplot interference from neighboring plots.

A540

A MACHISMO-LIKE DISEASE OF SOYBEANS IN MEXICO. J. Fletcher, M. E. Irwin, O. E. Bradfute, and G. A. Granada. Department of Plant Pathology, Oklahoma State University, Stillwater, OK 74078; INTSOY, University of Illinois, Urbana, IL 61801; Ohio Agricultural Research & Development Center, Wooster, OH 44691; Department of Plant Pathology, Instituto Colombiano Agropecuario, Palmira, Colombia.

Soybean plants in southwestern Mexico had symptoms similar to those reported for machismo disease in Colombia including delayed senescence, flower phyllody and virescence, lateral bud proliferation, and deformed pods. Phloem of diseased plants from both countries displayed positive Dienes' staining. Electron microscopy revealed many mycoplasmalike organisms in sieve elements of diseased samples from Mexico and Colombia. Leafhoppers of the species Scaphytopius fuliginosus, known vector of the machismo disease agent, were found in affected Mexican fields. Machismo disease of soybeans may be more widely distributed than previously thought.

A541

PATHOGENICITY OF ERWINIA HERBICOLA VAR. ANANAS TO HONEYDEW MELONS. J.M. Wells and M.J. Ceponis, USDA-ARS, Market Pathology Lab, Rutgers University, New Brunswick, NJ 08903.

Strains of yellow bacteria causing firm, brown lesions on honeydew melons were isolated from shipments originating from South and Central America. Bacteria were Gram-negative, non-acid fast, motile, facultative anaerobic rods (0.5 - 1.0 X 1.0 - 2.5 µm) that produced yellow pigment on nutrient agar. Strains were positive for catalase, β-galactosidase, gelatinase, β-lactamase, phosphatase, H_2S production, and utilized asparagine. Type strains Erwinia herbicola var. ananas, (ATCC 11530 and 23822) a pathogen of pineapple fruits, were identical to the yellow bacteria from melons. All strains were pathogenic to pineapple fruits and to melons. Fatty acid profiles, electrophoretic protein patterns, DNA mol % G + C and genome size of melon and type strains were identical.

A542

PATHOGENIC RELATIONSHIPS BETWEEN PIERCE'S DISEASE AND PHONY PEACH BACTERIA. B.C. Raju, J.M. Wells, S.M. Mircetich, and G. Nyland, Yoder Bros., Inc., Alva, FL 33920; USDA, ARS, NER, Rutgers Univ., New Brunswick, NJ 08903; and Dept. of Plant Pathology, Univ. of California, Davis, CA 95616.

Pierce's disease (PD) and phony peach (PP) bacteria were inoculated into almond, grape, plum and peach seedlings or transmitted by grafting and vectors. The PD bacteria caused symptoms on 18 of 20 grapes, leaf scorch on 16 of 20 almonds, and no symptoms on peach or plum. Three months after inoculation bacteria were reisolated from all symptomatic grape and almonds, from 5 of 20 plums, but not from peach. Similar results were obtained from graft-inoculated almond and peach. The vector Draeculacephala minerva transmitted PD bacterium from almond to almond but not to peach. The PP bacterium caused symptoms on peach and plum but none on almonds or grapes, nor could it be reisolated from almonds or grapes. Bacteria from leaf-scalded plum caused phony on peach. The PD and PP bacteria were distinguishable serologically. We conclude the PD bacterium is pathogenically different from the PP bacterium.

A543

IDENTIFICATION OF THE BACTERIUM CAUSING LETTUCE CORKY ROOT. C. M. Waters and R. G. Grogan. Department of Plant Pathology, University of California, Davis, CA 95616

A bacterium isolated in pure culture from Salinas Valley soil produced severe symptoms on the corky root susceptible lettuce cultivar Salinas and mild symptoms on the corky root tolerant cultivars Marquette and Montello. Colonies of the bacteria appear umbonate, translucent, circular, compact and often develop a raised edge and wrinkles. The corky root bacterium has the following characteristics: Gram positive wall structure, rod shaped (0.3-0.6 by 0.6-1.4 µm), single lateral flagellum, oxidase (+), catalase (-), acid production from glucose but not sucrose, optimum growth at 28-30 C, growth at 36 C but not at 39 C, survival after 10 min exposure to 53 C but not after 10 min exposure to 56 C, aerobic and microaerophilic growth and fatty inclusions which stain with Sudan Black B. These characters indicate the corky root bacterium belongs in the genus Corynebacterium.

A544

AN ERWINIA STEM-ROT OF HYDROPONIC CUCURBITS AND CRUCIFERS. Andrew C. Schuerger and Jean Carlson Batzer, The Land, EPCOT Center, P. O. Box 40, Lake Buena Vista, Florida 32830

A gram-negative, pectolytic and facultatively anaerobic Erwinia bacterium was consistently isolated from hydroponically grown crucifer and cucurbit plants. Abundant exudate from stem wounds, vascular discoloration, maceration of stem tissue, and foliar

wilt are the prominent symptoms. The bacterium is oxydase negative; produces acid from alpha-methylglucoside, xylose, palatinose, maltose and lactose; produces reducing substances from sucrose; does not utilize phosphatase; is resistant to erythromycin; and grows at 36°C. Thirteen crop cultivars were screened in a pathogenicity host range test. Stems were inoculated by injecting 0.1 ml of a 1 X 10^7 cfu/ml suspension of bacteria (from a 24-hr nutrient agar plate). Maceration of stem tissue, beginning at the site of inoculation, advanced most rapidly in squash, kale, pak-choi, and chinese cabbage. Stems of luffa gourd and chinese bitter melon (cucurbit cultivars) remained free of disease following inoculation.

A545

Bacterial leaf spots of Hibiscus rosa-sinensis. A. R. Chase, IFAS, University of Florida Agricultural Research Center - Apopka, 32703.

Hibiscus rosa-sinensis L. with a foliar disease were collected from several Florida nurseries. Lesions were scattered across the leaf and were dark-brown to black, angular to irregular and surrounded by a chlorotic halo. Several isolates of Pseudomonas sp., P. cichorii, and Xanthomonas sp. were recovered singly and in pairs. Healthy plants were inoculated with single isolates after 24 hr in mist, by wounding with insect pins and spraying to runoff with 1 x 10^8 CFU/ml. After 72 hr, plants inoculated with P. cichorii had 1 mm lesions at wound sites. Within 1 wk, characteristic angular lesions also formed in non-wounded tissue of older leaves. Pseudomonas sp. caused pinpoint to 1 mm lesions with chlorotic halos in nonwounded tissue of all ages 2 wk after inoculation. Xanthomonas sp. did not cause any symptoms after 4 wk. Distortion of new leaves was common on Pseudomonas sp. inoculated plants and leaf abscission was present for P. cichorii inoculated plants. Both pathogens were reisolated from their respective treatments but not controls.

A546

EVALUATION OF THE ADULT PLANT RESISTANCE OF *Xa-6* TO BACTERIAL BLIGHT OF RICE

Zhang Qi and T.W. Mew, The International Rice Research Institute Los Baños, Laguna, Philippines

Malagkit Sungsong, Zenith, IR944 and IR1695, all having the *Xa-6* gene for resistance to bacterial blight, were susceptible at seedling stage, but resistant at the 11th and 12th leaves. Malagkit Sungsong showed resistant on earlier leaves than did the other cultivars. By stagger planting to synchronize inoculation, resistance increased with ascending leaf position. Lesions on a susceptible cultivar (TN1) progressed faster at 33/25 C than at 25/20 C. The overall resistance of IR1545-339 was not altered by high temperature. These cultivars therefore have adult plant resistance and resistance was not affected by changes in temperature.

The bacterial population in the 6th, 9th and 10th leaves of Zenith was maximum 3 days after inoculation. On the 12th leaf, the trend was lower and declined faster.

A547

ROLE OF MOTILITY IN APPLE BLOSSOM INFECTION BY ERWINIA AMYLOVORA AND STUDIES OF FIRE BLIGHT CONTROL WITH ATTRACTANT AND REPELLENT COMPOUNDS. R. G. Bayot and S. M. Ries. Department of Plant Pathology, University of Illinois at Urbana-Champaign, 1102 S. Goodwin Ave., Urbana, Illinois 61801.

A nonmotile isolate (Nm22) and its motile revertant (MR22) of Erwinia amylovora were obtained after treating cells of the wild type (Isolate 110) with 0.3 M ethyl methanesulfonate. The pathogenicity of both isolates was similar to the wild type when inoculated to young shoots of apple seedlings. Significantly more infection was recorded in blossoms inoculated with MR22 than with the Nm22 isolate. The possibility of using attractant and repellent compounds to minimize apple blossom infection was explored. Benzoate and salicylate were used as repellents while malate and tartarate were used as attractants. None of these compounds applied to apple blossoms as 1 or 10 mM solutions, provided consistent protection from fire blight infection at an inoculum concentration of approximately 500,000 cells per ml.

A548

CHARACTERISTICS OF STRAINS OF PSEUDOMONAS SOLANACEARUM FROM TOBACCO IN NORTH CAROLINA. M. A. Haque and E. Echandi, Dept. of Plant Pathology, North Carolina State University, Raleigh, NC 27695-7616.

Forty nine strains of P. solanacearum isolated from flue-cured tobacco collected in 8 counties of N. C. were compared in terms of: physiological characteristics, bacteriocin production, pathogenicity in 6 hosts and virulence in 8 tobacco cultivars. All strains belonged to biotype I of Hayward's classification, produced bacteriocins and were clustered in 8 bacteriocin types, 12 pathogenicity groups and 9 virulence groups. No correlation was found between the bacteriocin types, pathogenicity groups and virulence groups, nor between the geographical locations and the bacteriocin types, pathogenicity groups and virulence groups.

A549

A SEMISELECTIVE AGAR MEDIUM FOR ISOLATING XANTHOMONAS CAMPESTRIS PV. TRANSLUCENS FROM WHEAT SEEDS. **N. W. Schaad and R. L. Forster, Dept. of Pl., Soil and Ent. Sci., University of Idaho, Moscow, ID 83843 and Kimberly, ID 83341.**

Black chaff of wheat causes serious losses in spring wheat under sprinkler irrigation in southern Idaho. Xanthomonas translucens is seedborne but no assay is available. An agar medium is described for isolating X. translucens from seeds of Triticum aestivum L. The medium, XTS agar, contains Difco nutrient agar, glucose, cycloheximide, gentamycin, and cephalexin. XTS agar inhibited 91% or more of those bacteria growing on nutrient glucose agar (NGA) from seed washings of five seed lots. X. translucens was isolated from 13 of 17 on XTS but from only three of 17 on NGA. Comparison between 1) assaying seed washings of naturally contaminated seedlots on XTS agar and 2) subsequent disease development in the field showed the former to be more sensitive. Such frequent isolation of X. translucens from seeds suggests that seed contamination is responsible for recent epiphytotics of black chaff in southern Idaho.

A550

Survival of Xanthomonas campestris pv. vesicatoria in Florida. J. B. Jones, R. E. Stall, J. P. Jones and K. L Pohronezny. University of Florida, IFAS, GCREC, Bradenton.

Survival of Xanthomonas campestris pv. vesicatoria (XCV) causal agent of bacterial spot of tomato and pepper, in crop residue was determined in 1982 and 1983. From residue of a fall crop, XCV was recovered after 6 months from tissue placed on the soil surface and after 5-6 months from buried tissue. After spring crops, the bacterium was detected at Homestead in tissue buried or placed on the surface after 1-2 months. At Bradenton, the bacterium was detected after 3 and 2 months from infested crop residue buried or placed on the soil surface, respectively. Three of six old fields had volunteer plants with at least 50% having bacterial spot. Only the fields that had cover crops of sorghum or where the plant beds with plastic mulch remained, contained volunteers. Fields that were disked periodically were free of volunteers. Epiphytic populations of XCV were found on 11 of 202 weed samples. Of those, 3 samples each were from Physalis pubescens and Solanum nigrum. Based on these results, XCV survives in crop residue, soil and possibly weeds for extended periods.

A551

RESISTANCE OF FIELD STRAINS OF XANTHOMONAS CAMPESTRIS PV. VESICATORIA TO COPPER BACTERIOCIDES. Adaskaveg, J.E. and Hine, R.B., Dept. of Plant Pathology, Univ. of Arizona, Tucson, Ariozna 85721

Bacterial spot, caused by Xanthomonas campestris pv. vesicatoria, was not controlled on tomato and pepper crops in Sinaloa, Mexico following routine applications in 1983 of the copper containing bacteriocide, Cosmocel 200. Pathogenic strains of the bacterium isolated from the two solanaceous crops were tested in vitro against several copper containing bacteriocides (Copper Sulfate, Kocide 101, Count-N, and Cosmocel 200) and antibiotics (Streptomycin, Tetracycline, Neomycin, Penicillin G, and Erythromycin). The Mexican strains were resistant while Arizona strains, isolated from pepper, maintained in culture since 1976, and recently shown to be pathogenic, were susceptible to all the copper formulations at recommended field rates. All isolates showed variable susceptibility to the antibiotics tested.

A552

A SEVERE STRAIN OF TOBACCO ETCH VIRUS RECOVERED FROM TOMATO IN MARYLAND. H. E. Moline, R. W. Goth and J. O. Kuti. USDA, ARS, HSI, Beltsville, MD and Hort. Dept., Univ. of MD, College Park

Tomato plants displaying symptoms which include severe stunting and failure to set fruits, were found to be infected with tobacco etch virus (TEV). There are no previous reports of TEV pro-

ducing symptoms in tomato plants. The severe tomato strain produced abundant laminar nuclear and cylindrical cytoplasmic inclusions in infected tomato leaves typical of TEV. The virus was difficult to transmit mechanically and had a restricted host range, including Lycopersicon esculentum, Datura stramonium, Nicotiana tabacum, and Chenopodium quinoa. Mechanical transmission and symptom expression was easiest to achieve at greenhouse temperatures of 30-35C and long days. Enzyme-linked immunosorbent assay (ELISA) revealed that the severe tomato TEV isolate (ATCC #PV364) reacted more strongly than the ATCC #PV69 TEV strain against TEV antiserum (PVAS69). No reaction was observed between the tomato isolate and antiserum prepared against potato leafroll virus, potato virus A, M, S, Y, tobacco mosaic virus, or tomato aspermy virus.

A553

DETECTION OF SUGARCANE MOSAIC VIRUS BY DOT-BLOT HYBRIDIZATION. R. C. French, S. H. Simon, and K. S. Derrick, Dept. of Plant Pathology and Crop Physiology and Dept. of Biochemistry, La. Agric. Exp. Sta., La. State Univ. Agr. Ctr., Baton Rouge, LA 70803.

Dot-blot hybridization, which involved spotting plant extracts on nitrocellulose filters and hybridizing with ^{32}P-labeled recombinant plasmid DNA, proved to be a rapid and sensitive method for detecting SCMV-H. DNA complementary (cDNA) to the RNA genome of sugarcane mosaic virus strain H (SCMV-H) was synthesized using avian myeloblastosis virus reverse transcriptase and oligo (dT) as primer. Double-stranded cDNA was inserted into the PstI site of plasmid pBR322 by the G-C tailing method and cloned in Esherechia coli HB101. Twenty recombinant clones containing SCMV-H sequences were obtained. Two plasmids with inserts of 1.2 kpb and 2.7 kbp were used for dot-blot assays. Dot-blot hybridization also revealed that SCMV strain I, but not SCMV strains A, B, D, M, and J has sequences in common with the strain H-derived clones.

A554

TRANSMISSION AND CYTOPATHOLOGY OF JATROPHA MOSAIC DISEASE CAUSED BY A WHITEFLY-TRANSMITTED GEMINIVIRUS. J. BIRD, K.S. KIM, R.L. RODRIGUEZ, E.M. MARTIN, AND J. ESCUDERO, Agri. Experiment Station, Univ. of Puerto Rico, Río Piedras, Puerto Rico and Dept. of Plant Pathology, Univ. of Arkansas, Fayetteville, AR 72701.

The causal agent of the mosaic of Jatropha gossypifolia was transmitted to healthy Jatropha plants by the whitefly, Bemisia tabaci. The agent was transmitted either mechanically or by the insect to Nicotiana benthamiana. Electron microscopy revealed cytopathic effects characteristic of known whitefly-transmitted geminiviruses in both hosts. These were 1) electron-dense fibrillar bodies which were often ring-shaped and 2) virus-like particles, 15-20 nm in diameter, in loosely clustered aggregates or a closely packed extremely large body occupying more than 3/4 of a nucleus. Many ring-shaped fibrillar bodies were composed of orderly aligned spherical bodies of high electron density alternating with less electron-dense fibrillar material. Cytochemical studies indicated that the bodies are composed of DNP. Virus particles occurred only in the nuclei of phloem associated parenchyma cells or in the lumen of mature sieve elements.

A555

LOCALIZATION OF FIRST APPEARANCE OF BARLEY STRIPE MOSAIC VIRUS (BSMV) PROTEIN IN INFECTED WHEAT CELLS. Na-Sheng Lin and W. G. Langenberg, Dept. of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722.

The location of viral capsid protein in root tips systemically infected with BSMV was studied by immunoelectron microscopy (J. Ultrastruct. Res. 84:16-24). By following infection from cell to cell from the meristematic point back to older cells, we distinguished 4 cytological stages of infection. Stage I, plastids contain peripheral vesicles (= infection initiation marker). No BSMV virions are seen, but BSMV protein can be first detected on vesiculated plastid membranes by gold-IgG complexes. Stage II, BSMV virions appear mostly perpendicular to the plastid membrane. All virions label specifically with immunogold. Stage III, BSMV virions appear attached to endoplasmic reticulum and BSMV crystals form in the cytoplasm. Stage IV, a large amount of viral protein, with few or no virions, can be detected in nuclei by immunostaining. Viral protein is limited to euchromatin and never occurs in heterochromatin or in nucleoli. The fourth stage is several cells behind the first stage.

A556

AGGREGATION OF POTYVIRAL RNA. C. Luciano, M. Siaw, S. Ballard, Z. Xu, and J. Shaw, Department of Plant Pathology, University of Kentucky, Lexington, KY 40546.

The RNA of the potyvirus tobacco vein mottling virus (TVMV), isolated from purified virus by disruption in SDS at pH 9 and sucrose density gradient centrifugation, contains discrete high molecular weight aggregates. These aggregates appear as low-mobility bands upon agarose gel electrophoresis of native RNA and RNA denatured by glyoxal or glyoxal plus DMSO. The mobility of these aggregates in 0.8% denaturing gels suggests they represent dimers, trimers, etc. Aggregates are stable to extraction with phenol or perchlorate and to treatment with SDS plus β-mercaptoethanol at 65° C. Aggregates are converted to monomer RNA mobility by treatment with 50 µg/ml Proteinase K for 30 min at 37° C, indicating protein is responsible for maintaining intermolecular association. This protein is probably a Vpg which we have detected in TVMV-RNA preparations. Similar aggregates have also been observed in potato virus Y (PVY) RNA.

A557

INFECTION OF TOBACCO PROTOPLASTS WITH A POTYVIRUS. Z. Xu, S. T. Ballard, C. S. Luciano & J. G. Shaw, Dept. of Plant Pathology, University of Kentucky, Lexington, KY 40546.

Little is known about in vivo genome expression of potyviruses because of difficulties in establishing infections with these viruses in protoplasts. We have found that a modification of standard inoculation procedures provides efficient infection of tobacco protoplasts with tobacco vein mottling virus (TVMV). After inoculation with 1 µg/ml TVMV (preincubated with 1 µg/ml poly-L-ornithine), protoplasts were washed once with 200 mM $CaCl_2$ and once with culture medium, and incubated at 25°C. After 2-3 days, protoplasts were shown with specific antibodies to contain the TVMV cylindrical inclusion protein (CIP) and about 1.5×10^5 virus particles per protoplast. Protoplasts were applied to nitrocellulose and analyzed by anti-CIP IgG and enzyme-conjugated secondary antibodies. Up to 75% of the protoplasts were revealed by microscopy to be infected.

A558

A COMPARATIVE STUDY OF THE CAPSID PROTEINS AND THE 3'TERMINAL NUCLEOTIDE SEQUENCES OF THE POTYVIRUSES TOBACCO ETCH AND PEPPER MOTTLE. R. Allison, R. Johnston, F. Armstrong, R. Horton, and W. G. Dougherty, North Carolina State University, Raleigh, NC 27695-7616.

The capsid protein of tobacco etch virus (TEV) and pepper mottle virus (PeMV) have molecular weights of 30,000 and 32,000 respectively. Polyclonal antisera to each protein revealed no antigenic homology in ouchterlony tests and only limited homology by ELISA. However, considerable amino acid sequence homology is predicted from the nucleotide sequence of the molecularly cloned capsid protein genes. Accounting for molecular weight differences between the two capsid proteins, 244 amino acids of the primary protein sequence could be compared directly. Of these, 65% were identical. The observed differences in predicted amino acid sequence were near the amino and carboxy termini of the two capsid proteins.

A559

NUCLEOTIDE SEQUENCE DETERMINATION OF A GENE PROXIMAL TO THE 3' TERMINUS OF PEPPER MOTTLE VIRUS GENOMIC RNA. W.G. Dougherty, R. Allison, F. Armstrong, R. Horton and R. E. Johnston, North Carolina State University, Raleigh, N.C., 27695-7616

The RNA of pepper mottle virus (PeMV), a potyvirus, was purified and complementary DNA (cDNA) was synthesized using oligo dT_{12-18} as a primer in the reverse transcriptase reaction. Double-stranded cDNA was inserted into the bacterial plasmid pBR-322. The nucleotide sequence of the 1400 nucleotides at the 3' terminus was determined. This sequence contained an open reading frame of 1005 nucleotides commencing with an AUG codon near the 5' terminus of the cloned fragment and ending with a single UGA stop codon. An untranslated region of 300 nucleotides, containing 21 stop codons in all 3 reading frames and a 3' polyadenylate segment, which varied in length from 20 to 120 residues followed. The open reading frame could encode a 36,000d protein. The predicted amino acid composition of this protein compared to PeMV capsid protein (32,000d) will be reported.

A560

GOLD-LABELED ANTIBODIES FOR THE DETECTION OF PLANT VIRUS ANTIGEN ON NITROCELLULOSE PAPER. Y. H. Hsu and M. K. Brakke, Agricultural Research Service, U.S. Department of Agriculture and Department of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722.

Gold-labeled antibodies yield a pink color which permit visual

detection of plant virus antigen on nitrocellulose paper. Virus antigen bound on nitrocellulose paper is first reacted with primary antibody raised in rabbits and then with gold-labeled goat anti-rabbit IgG. As little as 1-5 pg of Tobacco Mosaic Virus (TMV) protein is specifically stained either purified or in plant extracts. Gold-labeled antibody is relative inexpensive and easily prepared in laboratory. The staining technique is simple and it avoids sources of non-specific staining which limit other techniques. The procedure has been used either as a dot blot with 1 µl samples or a electro blot with samples transfered to nitrocellulose sheet after gel electrophoresis.

A561

SOME INTERACTIONS OF TMV-COMMON, TMV-LEGUME, AND SOUTHERN BEAN MOSAIC VIRUS. Thomas M. Zinnen and R. W. Fulton, Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706.

In inoculum interference tests, TMV-legume (TMV-L) and TMV-common (TMV-C) interfered with each other in Pinto bean and in Nicotiana sylvestris; interference between southern bean mosaic virus (SBMV) and TMV-C or TMV-L was less than the interference between TMV-C and TMV-L. In local cross protection tests, TMV-C and TMV-L locally cross protected against each other in N. sylvestris and in Pinto. TMV-L did not protect locally against SBMV, but SBMV caused some protection against TMV-C in Black Turtle bean. In systemic cross protection tests, darkening plants 24-48 h before challenge inoculation increased TMV-C lesions on healthy Pinto but decreased TMV-C lesions on Pinto infected with TMV-L; darkness increased SBMV lesions on both healthy Pinto and Pinto infected with TMV-L.

A562

NEURON LOCATION IN THE MANDIBULARY STYLETS OF THE APHID, MYZUS PERSICAE. R. L. Mernaugh, Dept. of Microbiology, Iowa State Univ., Ames, Iowa 50011

An EM study was done to determine the orientation of the neuron pair within the mandibulary stylets of M. persicae. Continuous serial sections of the stylets indicate that the neuron pair is contained within a single central duct throughout most of the stylet's length. However, approximately 7 microns from the stylet tip, each neuron becomes encompassed in a separate duct. Both neurons remain separate until they terminate 3 microns from the tip of the stylet. Electron dense structures appearing within the individual ducts 3 microns from the stylet tip appear to be mechanical impulse receptors for the neurons. No pores were seen connecting the neurons to the exterior environment of the stylets. These results suggest that the mandibulary stylet neurons act as mechanoreceptors rather than chemoreceptors.

A563

FURTHER STUDIES ON THE ROLE OF DOUBLE-STRANDED RNA (dsRNA) IN VIRULENCE OF RHIZOCTONIA SOLANI. D. H. Zanzinger, B. P. Bandy, and S. M. Tavantzis, Dept. of Botany and Plant Pathology, University of Maine, Orono, ME 04469.

Sixty-six isolates of R. solani obtained from Maine soils and several locations within the U.S. and representing five anastomosis groups (AG 1 through AG 5) were characterized for dsRNA content to determine whether presence of dsRNA were associated with virulence. Sixty-three isolates containing one to several segments of dsRNA exhibited a wide range in virulence from hypovirulent to highly virulent. The number of dsRNA segments was not correlated with virulence. Size of dsRNA segments was not isolate specific nor was it characteristic for a particular AG. Electrophoretic patterns of dsRNA were consistent and stable over more than twelve months following several transfers. Despite multiple transfers in culture, no significant changes in virulence were detected. These findings, which represent a broad spectrum of isolates of R. solani, suggest the mere presence of dsRNA is not necessarily associated with hypovirulence.

A564

CHARACTERIZATION OF A dsRNA CONTAINING VIRUS FROM RHIZOCTONIA SOLANI. S.M. Tavantzis, and B.P. Bandy, Department of Botany and Plant Pathology, University of Maine, Orono, ME 04469.

We have previously reported the high frequency of dsRNA in virulent and hypovirulent isolates of R. solani. Isometric virus-like particles (VLP's), 33 nm in diameter, have been purified to apparent homogeneity from a dsRNA containing isolate (Rhs 717) of R. solani. The dsRNA segments isolated from purified VLP preparations had the same size (~6.5, 1.3, 1.2, 0.9, and 0.8x 10^6) as those isolated directly from mycelial tissue of Rhs 717 by phenol extraction. The VLP's were purified by two cycles of differential centrifugation in 0.05 M phosphate-citrate buffer, pH 5.8, using sucrose cushions, rate-zonal, and isopycnic gradient centrifugation. Purified VLP's had an average sedimentation coefficient of 139 S, a buoyant density of 1.37 g/cm^3 in CsCl, and an A_{260}/A_{280} absorbance ratio of 1.45. The melting curve of the mixture of dsRNA segments isolated from the Rhs 717 VLP's in standard saline citrate (1xSSC) buffer had a sharp thermal transition with Tm = 95 C.

A565

TWO DOUBLE-STRANDED RNAs OF UNKNOWN ORIGIN IN VEGETATIVE TISSUES OF BEAN. D.A. Wakarchuk and R.I. Hamilton, Agriculture Canada Research Station, 6660 N.W. Marine Drive, Vancouver, B.C. Canada V6T 1X2.

Two double-stranded RNAs (dsRNA) were consistently associated with extracts of apparently healthy seedlings of Phaseolus vulgaris cv Black Turtle Soup (BTS) from seedlots obtained in 1968 and 1983. No such RNAs were detected in seedlings of several other bean varieties. Each of 15 BTS plants contained the same RNAs. Their molecular weights estimated by agarose gel electrophoresis, were 8.8 and 10.0 x 10^6 (13.3 and 15.2 kbp). Both RNAs were hydrolyzed by ribonuclease A (3µg/ml) in 0.1 xSSC (0.15 M NaCl, 0.015 M sodium citrate, pH7.0) but were resistant to the enzyme in 2.0 x SSC and to deoxyribonuclease. No cytopathic evidence of virus infection was obtained by electron microscopy although chloroplasts were atypical, containing dense strands in the stroma. Sap or total nucleic acid extracts of BTS seedlings did not induce symptoms of virus infection in test plants of other varieties and no dsRNA was isolated from them.

A566

NON-CAPSID PROTEIN ASSOCIATED WITH WHEAT STREAK MOSAIC VIRUS INFECTION by M. K. Brakke, Ellen Ball, Y. H. Hsu, and J. Joshi, Agricultural Research Service, U.S. Department of Agriculture and Department of Plant Pathology, University of Nebraska, Lincoln, Nebraska 68583-0722.

Wheat streak mosaic virus has flexuous rod-shaped virions 700nm long with a single-stranded RNA genome of about 2.8x10^6 Mr. Virus-infected tissue contains a 66K Mr protein in about ten times the concentration of virions. Antibody was prepared to the 66K protein purified by centrifugation and SDS gel electrophoresis. The antibody does not react with WSMV virions and reacts only slightly with extracts of uninfected plants, but reacts strongly with extracts of infected plants in agar double diffusion and other tests. Translation of purified WSMV RNA in vitro in a rabbit-reticulocyte cell-free system yielded more than 20 polypeptides, several of which were precipitated with antibody to virions. Experiments on precipitation of in vitro translation products with antiserum to the 66K Mr protein are in progress.

A567

USE OF MONOCLONAL ANTIBODIES IN SEROLOGICALLY SPECIFIC ELECTRON MICROSCOPY OF BARLEY YELLOW DWARF VIRUS. R. Diaco[1], J. H. Hill[2] R. M. Lister[3], and D. P. Durand[1]. [1]Dept. of Microbiology, Iowa State University, Ames, IA 50011. [2]Dept. of Plant Path., Seed and Weed Sciences, Iowa State University. [3]Dept. of Botany and Plant Path., Purdue University, West Lafayette, IN 47907.

Monoclonal antibodies (M-Ab) independently produced against three isolates (PAV, MAV, and RPV) of barley yellow dwarf virus (BYDV) were able to capture the heterologous and homologous isolates of BYDV in serologically specific electron microscopy. Nitrocellulose-coated carbon-stabilized copper grids coated with protein-A purified M-Ab preparations were used to capture virus from purified preparations of each viral isolate. M-Ab prepared against individual isolates were able to specifically bind all three isolates. Identically treated grids, however, were unable to bind the unrelated soybean mosaic virus. Decoration experiments using ferritin-labelled M-Ab suggested the M-Ab bound to external epitopes on the capsid. This is the first report of successful use of M-Ab in SSEM of plant viruses.

A568

VIRUS-FUNGUS INTERRELATIONSHIPS IN A FUSARIUM ROOT AND CROWN ROT COMPLEX IN ASPARAGUS. T.A. Evans and C.T. Stephens. Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312.

Declining asparagus yields in Michigan are a consequence of crown and root rot incited by Fusarium oxysporum f. sp. asparagi (FOA) and F. moniliforme (FM) in stressed plants. Asparagus Virus II (AV II) has been detected in a high percentage of

asparagus plants within most commercial plantings in Michigan and may be an important stress factor. In greenhouse studies, asparagus seedlings infected with AV II had more severe crown and root rot when challenged with FOA. The difference in disease severity was smaller between virus-infected and virus-free asparagus seedlings when challenged with FM. Root exudates, collected from AV II-infected asparagus clones grown in liquid culture, increased germination of FOA microconidia whereas root exudates of virus-free clones had less effect on the germination of FOA or FM microconidia.

A569

ELECTROPHORETIC PATTERNS OF SEVERAL MULTIPLE FORMS OF ENZYMES IN HEALTHY AND BARLEY STRIPE MOSAIC VIRUS (BSMV) INOCULATED BARLEY CULTIVARS. F. C. Wu and R. G. Timian, Department of Plant Pathology, USDA-ARS Plant Pathology, North Dakota State University, Fargo, ND 58105.

Multiple forms of enzymes (isozymes) of three classes (hydrolases, oxidoreductases, and isomerases) from BSMV inoculated and virus-free barley seedlings were studied. Two barley cultivars, Black Hulless (C.I.666) (susceptible) and C.I.4197 (resistant), were inoculated with two strains of the virus, CV52 (ND18) (virulent) and CV42 (ND159) (mild). After polyacrylamide disk electrophoresis of extracts and specific histochemical staining, significant changes in some of the isozyme patterns were seen in Black Hulless barley seedlings 7 days after inoculation. Variations were found in acid phosphatase, alcohol dehydrogenase, and glucosephosphate isomerase. The change of isozyme activities were correlated with virulence of the virus strains. Distinct differences in isozyme patterns were also observed between the susceptible and the resistant barley cultivars.

A570

HOMOLOGY AMONG NUCLEIC ACIDS FROM MICHIGAN HYPOVIRULENT STRAINS OF ENDOTHIA PARASITICA. C. P. Paul, M. A. Estelle and D. W. Fulbright, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

Hypovirulent strains of Endothia parasitica isolated from recovering American chestnut trees (Castanea dentata) in Michigan exhibit many diverse dsRNA banding patterns on agarose gels. Total dsRNA isolated from the GH2 strain was 5' end labeled with ^{32}P and used to probe dsRNA isolated from GH2 and other hypovirulent strains which had been immobilized on nitrocellulose. Hybridization results indicate that there is homology between the dsRNA of the GH2 strain and the dsRNA of some strains isolated from other groves which exhibit different banding patterns. Hybridizations of labeled GH2 dsRNA to variously fractionated GH2 nucleic acids indicate that dsRNA from the GH2 strain binds to oligo(dT) cellulose.

A571

SOUTHERN BEAN MOSAIC VIRUS-INDUCED HYPERSENSITIVE REACTION IN PHASEOLUS VULGARIS L. 'PINTO': ANALYSIS OF SOLUBLE AND PLASMALEMMA PROTEINS. F. Mohamed, Carole Sieckman and O.P. Sehgal. Department of Plant Pathology, University of Missouri, MO 65211.

Sodium dodecyl sulfate-polyacrylamide gel electrophoretic analyses of soluble proteins from diseased leaf tissue showed three additional proteins of mol. wt. 36K, 30K (virus coat protein) and 20K. Furthermore, two proteins, mol. wt. 17K and 15K, were present in amounts 3 to 5 times greater in the diseased than in the healthy tissue. The relative proportion of a mol. wt. 41K protein was reduced slightly as a result of viral infection. Approximately 12-15 proteins (mol. wt. range, 92K to 12K) were associated with the leaf plasmalemma of which proteins of mol. wt. 54K, 15.5K and 13.5K were the most prominent. No qualitative or quantitative differences were detected between plasmalemma proteins from diseased and healthy leaves.

A572

GENE VI OF CAULIFLOWER MOSAIC VIRUS (CaMV) CONTROLS SYSTEMIC SPREAD IN SOLANACEOUS HOSTS. J.E. Schoelz, S.D. Daubert, R. J. Shepherd. Dept. of Plant Pathology, Univ. of Calif. Davis, CA

A region of the CaMV genome that controls systemic development of the virus in a solanaceous host (Datura stramonium) has been mapped. Whereas most strains of CaMV, e.g. CM1841, infect D. stramonium with the production of necrotic local lesions, one strain, D4, induced chlorotic local lesions and developed systemically. To determine which portion of the DNA genome controlled systemic spread in D. stramonium, hybrid genomes were constructed in vitro by cleaving cloned viral DNA with restriction endonucleases, mixing selected fragments from each parent strain, ligating, and transforming E. coli. Hybrids were also made in planta by subcloning portions of the CaMV genome, excising the subcloned fragments from the cloning vector, then inoculating complementing segments from the two strains onto turnips. Recombinant genomes were identified by screening for characteristic combinations of restriction sites. The region conferring systemic infection of D. stramonium was associated with the first half of open reading region VI in every hybrid tested.

A573

COMPARATIVE DETECTION OF CARNATION ETCHED RING VIRUS (CERV) USING MOUSE MONOCLONAL ANTIBODIES AND CHICKEN AND RABBIT ANTISERUM. H. T. Hsu and R. H. Lawson, American Type Culture Collection, Rockville, Md. 20852 and Beltsville Agricultural Research Center, USDA, Beltsville, Md. 20705

Mouse hybridoma-derived monoclonal antibodies (McAbs) against CERV were evaluated. About 10^3 to 10^4 times more antibodies were produced in ascites than in culture fluid. The minimum concentration of purified CERV detected by double antibody sandwich ELISA (McAb coating and McAb-enzyme conjugate detecting) was about 0.25 µg/ml. In a triple antibody sandwich procedure (McAb coating, chicken ∝ CERV antibody, and enzyme labeled goat ∝ chicken globulin detecting) similar sensitivity was obtained when compared with the McAb double antibody sandwich procedure. About 8 times more CERV was detected from Saponaria vaccaria than from Dianthus caryophyllus by both methods. Grids pretreated with protein A followed by either McAb or rabbit antiserum showed a significant difference in the number of virions attached compared to grids treated only with antibodies.

A574

IN VITRO MESSENGER RNA ACTIVITY OF SEVERAL SATELLITE RNAS OF CUCUMBER MOSAIC VIRUS. Maria Avila-Rincon, C. W. Collmer, and J. M. Kaper. Plant Virology Laboratory, PPI, U.S.D.A., Beltsville, MD 20705

Three different cucumber mosaic virus satellite RNAs (CARNA 5s), (S1)CARNA 5 from CMV strain S, (1)CARNA 5 from CMV strain 1, and (n)CARNA 5 from CMV strain D, have been purified from mixtures and translated in a wheat germ protein-synthesis system. All three CARNA 5s produce polypeptides that migrate to unique positions in SDS-polyacrylamide gels containing 8M urea. Only (S1)CARNA 5, a nonnecrotic satellite RNA, shows the translational characteristics previously reported by Owens and Kaper (Virology, **80**, 196-203, 1977) for CARNA 5 isolated from CMV strain S. Similar results are obtained with (S1)CARNA 5 isolated from virions or from its double-stranded form. This CARNA 5 has been sequenced and contains an open reading frame capable of coding for a polypeptide of 5,400 daltons, the estimated size of its larger translational product.

A575

SYNTHESIS OF DOUBLE-STRANDED RNA BY A VIRUS-ENRICHED FRACTION FROM AGARICUS BISPORUS. C. Peter Romaine and A. Sriskantha, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Partially-purified virus preparations from LaFrance disease-affected sporophores of Agaricus bisporus have up to a 15 fold higher RNA polymerase activity than comparable preparations from healthy sporophores. Enzyme activity is dependent upon the presence of Mg^{++} and the four nucleoside triphosphates (K_m=2.14 X 10^{-6} M), and is insensitive to actinomycin D, α-amanitin, and rifampicin. The ^3H-labelled enzyme reaction product is double-stranded RNA(dsRNA) as judged by its behavior upon CF-11 cellulose chromatography and ionic strength-dependent sensitivity to hydrolysis by RNase. The principal dsRNA products have estimated molecular weights of 4.3 and 1.47 X 10^6 and correspond in size and share nucleotide sequences with the major dsRNAs present in the virus preparations. The data suggest that the RNA polymerase associated with the virus fraction is a replicase which functions to catalyze the synthesis of genomic dsRNAs.

A576

ROLE OF THE HINDGUT IN APHID ACQUISITION OF BARLEY YELLOW DWARF VIRUS (BYDV). F. E. Gildow, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

An ultrastructural study of Rhopalosiphum padi L. fed on oats infected with the RPV-NY isolate of BYDV indicated that the hindgut is the site for BYDV ingress into the aphid homocoel. Virions were consistently associated with membranes of the hindgut in 15 R. padi fed on RPV-infected oats, but not with the midgut regions

of the same aphids, or in hindguts of 8 aphids fed on healthy oats. Adsorption of virus to the hindgut apical plasmalemma and endocytosis by coated pits was suggested as a mechanism for virus uptake. Virions appeared to be released into the hemocoel by fusion of virus-containing vesicles with the basal plasmalemma. Unidirectional transport of virus from the gut lumen to the homocoel was indicated by immuno-labelling experiments.

A577

PRODUCTION OF MONOCLONAL ANTIBODY SECRETING HYBRIDOMA CELL LINES TO PRUNE DWARF VIRUS BY IN VIVO AND IN VITRO IMMUNIZATION. Ramon L. Jordan, Nancy Elliott, and H. T. Hsu. USDA, Beltsville, MD 20705 and American Type Culture Collection, Rockville, MD 20852.

Seven hybridoma cell lines secreting monoclonal antibodies (McAbs) to prune dwarf virus (PDV) have been produced and evaluated. Five cell lines were generated by a standard in vivo immunization protocol. Two additional hybridomas were developed by an in vitro immunization procedure. Spleen cells from a BALB/c mouse previously injected with 20 µg PDV were cultured (in vitro) in the presence of 100 µg PDV antigen. The spleen cells were sensitized for 4 days in thymocyte conditioned serum-free media prior to fusion with NS1 myeloma cells. Analysis of the immunoreactivity and relative binding characteristics of the McAbs with immunogen and related and unrelated viruses in direct and indirect ELISA procedures will be presented.

A578

RELATIONSHIPS BETWEEN VIRUS INFECTION, DISEASE RESISTANCE, AND EARLY GROWTH OF WINTER WHEAT. H. J. Larsen, M. K. Brakke, and W. G. Langenberg. Agric. Res. Service, USDA, Lincoln, NE 68583

Varieties of hard red winter wheat field-resistant ('Homestead and 'Newton') or field-susceptible ('Scout-66') to soilborne wheat mosaic virus (SBWMV) were each susceptible in the laboratory to the vector fungus and, through sap inoculation, to SBWMV and wheat streak mosaic virus (WSMV). Infection with either virus reduced root growth more than shoot growth, and secondary root growth more than primary root growth regardless of variety. Root biomass of 'Scout-66' and 'Homestead WSMV-infected seedlings was 39% & 45% of normal, respectively, and shoot biomass 68% & 61% of normal. However, SBWMV infection reduced secondary root numbers and root biomass of 'Homestead' and Newton to only 11-52% of normal while root biomass of infected 'Scout-66' plants was 73-78% of normal. Shoot growth of SBWMV-infected 'Homestead' and 'Newton' was unaffected, but that of Scout-66 was reduced to 83% of normal. Field resistance to SBWMV may result from hypersensitivity in the root to virus inoculation by the fungal vector.

A579

INCIDENCE OF INFECTION BY THE GROUNDNUT (= PEANUT) CHLOROTIC SPOT VIRUS IN THE COTYLEDONOTROPHIC STAGES OF SUSCEPTIBLE HOSTS. Talisetty Haragopal, Department of Botany, S.V.Jr. College, Tirupati, India 517501

The time of susception was studied in the different hosts susceptible to the groundnut chlorotic spot virus. In all the cases susceptibility was found confined to only the 'cotyledonotrophic stage' of the 'growth history' of these hosts. Beyond this stage, infection was not detected in any of the hosts. These studies suggest a nexus between cotyledonotrophy and susceptibility in these plants.

A580

ON THE BIOCHEMISTRY OF CHLOROTIC SPOT-MOTTLE PATHOLOGY OF THE LEAVES OF DATURA FASTUOSA LINN. Vedam Chandrasekharam and Nadadur S. Srikanth, Research Centre in Biology, Department of Zoology, S.G.S. Arts College, Tirupati, India 517501

Examination of the organic composition of the leaves of Datura fastuosa showing chlorotic spot-mottle pathology suggested host-protein to be the source of biosynthesis of the pathogen. Besides, utilization of carbohydrates also was evident in relation to symptom development. Activity levels of some enzymes studied were in accordance with the changes in the organic composition mentioned above.

A581

DETECTING THE INCLUSION FORMING BLUEBERRY RED RINGSPOT VIRUS WITH ELISA. J.M. Gillett and D.C. Ramsdell, Dept. of Botany and Plant Pathology, Michigan State Univ., East Lansing, MI 48824

The standard assay for blueberry red ringspot virus (RRSV) is to bud graft to susceptible varieties and wait for symptom development. Since this can take months, the suitability of enzyme linked immunosorbent assay (ELISA) for detecting RRSV was tested. In preliminary studies it was suspected that many of the virions were not released from their inclusions in normal ELISA extraction. To overcome this, samples were taken through a partial purification before being used in ELISA. Blueberry leaves were extracted 1:5 (w/v) in 0.1 M Na_2HPO_4, KH_2PO_4, pH 7.2 after which 2.5% triton-x, 6% urea, and 8% n-butanol were added. Samples were then shaken overnight at 4 C. After straining the samples through cheesecloth, virions were concentrated by adding 8% polyethylene glycol and 1% NaCl followed by a low speed centrifugation. The pellets were resuspended in 1 ml of PBS-Tween containing 0.5% ovalbumin, 20% polyvinylpyrrolidone and tested by ELISA. Twenty-five of 25 (100%) symptomatic and 0 of 25 (0%) non-infected leaf samples were positive.

A582

A VIRUS FROM HIBISCUS ROSA-SINENSIS WITH PROPERTIES OF A TOMBUSVIRUS. S. S. Hearon. USDA-ARS, Beltsville, MD 20705.

An icosahedral virus (HTV) was isolated from distorted and cupped leaves of a varigated hibiscus cultivar. Chenopodium quinoa, C. amaranticolor, and Tetragonia expansa were local lesion hosts for HTV. HTV was purified from C. quinoa by ammonium sulfate precipitation and differential and cesium chloride density gradient centrifugation. The buoyant density of HTV in CsCl at pH 6 was 1.35 g/cc, but virus stability was affected by the pH of the gradient. A major protein, MW 38 kd, and a minor protein, MW 66 kd, were detected by SDS-PAGE. HTV migrated as multiple electrophoretic components in agarose at pH 5, 6, and 7. Extracts of infected C. quinoa contained one genomic and two subgenomic ds-RNAs. HTB was serologically related to hisicus chlorotic ringspot virus. In ultrathin sections of C. quinoa, virions occurred in the cytoplasm and the central vacuole. Mitochondria cristae contained stranded material. Swollen or disrupted peroxisomes contained membrane-bound bodies of granular and stranded material. These data are evidence that HTV is a tombusvirus.

A583

INDUCTION OF SUPPRESSIVENESS TO RHIZOCTONIA SOLANI IN AN UNMODIFIED LOAMY SOIL. C. Wijetunga, R. W. Stack (Dept. of Plant Pathology, NDSU, Fargo, ND 58105) and R. Baker (Dept. of Botany and Plant Pathology, CSU, Ft. Collins, CO 80525).

Previously, induction of soil suppressiveness to large (>589µm) propagules of Rhizoctonia solani has required artificial modification of soil pH. A loamy soil with a natural pH 6.0 collected from a grain field near Erie, North Dakota was artificially infested with small (<425µm) and large (>600µm) propagules of R. solani. When radishes were grown at weekly intervals in this soil in a growth chamber at 26 ± 2°C, the soil became suppressive to both small and large R. solani propagules. The damping-off of radish increased initially and gradually diminished. Rhizoctonia populations, determined by soil pellet sampler on Ko-Hora selective medium, also declined. Trichoderma populations, as determined by dilution plate method on selective medium (Elad and Chet), increased from low levels to very high levels over a 6 week period when cropped to radishes; but not when wheat, a non-host, was planted.

A584

SURVIVAL CURVES TO EVALUATE BIOLOGICAL CONTROL OF RHIZOCTONIA SOLANI BY TRICHODERMA HARZIANUM. C. Wijetunga, R. W. Stack (Dept. of Plant Pathology, NDSU, Fargo, ND 58105) and R. Baker (Dept. of Botany and Plant Pathology, CSU, Ft. Collins, CO 80525)

Survival rates of large (>589µm) and small (<250µm) propagules of Rhizoctonia solani in soil were used to determine the amount of biological control obtained by Trichoderma harzianum. Populations of R. solani were assessed using Ko-Hora selective medium and a multiple pellet soil sampler. Dilution plates on selective medium (Elad and Chet) were used for populations of T. harzianum. Increase in inoculum density was greater from large propagules of R. solani than from small propagules. Induced suppression was assessed in loamy sand, loam, clay loam and silty clay soils. Trichoderma affected the growth phase of the survival curves of R. solani in some treatments. There were interactions among the Trichoderma treatments, soil physical properties and cropping. Analysis by survival curves allowed separation of these factors and the determination of their

individual and combined contributions to the decline of R. solani in soil.

A585

EDAPHIC PARAMETERS ASSOCIATED WITH ESTABLISHMENT OF THE BIO-CONTROL AGENT TALAROMYCES FLAVUS. D. R. Fravel[1], J. J. Marois[2], and D. M. Benson[3]. [1]Univ. of Maryland, College Park, MD 20742, [2]Soilborne Diseases Laboratory, Plant Protection Institute, ARS, USDA, Beltsville, MD 20705, and [3]North Carolina State Univ., Raleigh, NC 27650.

The widespread use of Talaromyces flavus to control Verticillium dahliae may depend on the ability of this antagonist to establish itself in soils having different chemical, physical and biological characteristics. This study was undertaken to determine which edaphic parameters are associated with T. flavus survival and proliferation. T. flavus populations were monitored for 13 weeks in 25 soils to which ascospores had been added. Six of 23 physical and biological parameters measured in the 25 soils were related to survival and proliferation of T. flavus in a multivariate principal axis factor analysis. These parameters were cation exchange capacity, potassium, sodium, zinc, soluble salts, and total soil bacteria.

A586

FACTORS AFFECTING BIOLOGICAL CONTROL OF PYTHIUM ULTIMUM ON ALFALFA USING SEED TREATMENT WITH GLIOCLADIUM VIRENS, TRICHODERMA HARZIANUM AND T. HAMATUM. D.A. Abdelwahab and G.W. Buchenau Plt. Sci. Dept., South Dakota State University, Brookings, SD 57007

Gliocladium virens applied as a seed treatment controlled alfalfa damping-off in natural soils artifically infested with high levels of Pythium ultimum,but Trichoderma hamatum and Trichoderma harzianum were not effective at the relatively high soil pH levels tested. Biocontrol with G. virens was nearly as effective as metalaxyl seed treatment,and better than Captan in most environments. Biocontrol was optimum at soil temperatures between 15 and 30 C, soil moistures of 15% to 30%, soil pH of 6 to 8 and when seeds had been treated with conidial concentrations of 10^9 spores/ml or greater. In a parallel study in infested steamed soil, biocontrol was most effective between 25 and 35 C at relatively low soil moistures, and pH levels of 6 to 8. Unbleached chitin in the spore suspension nullified biocontrol: bleached chitin had no effect. G. virens also controlled damping-off in field tests.

A587

GENETIC ANALYSIS OF PYTHIUM ANTIBIOSIS BY PSEUDOMONAS FLUORESCENS. Paul Gill, Lee Anderson, Trevor Suslow and Gareth Warren. Advanced Genetic Sciences, Inc., 6701 San Pablo Ave., Oakland, CA 94608

A number of field isolates of P. fluorescens have been identified which exhibit antibiosis toward Pythium ultimum on defined media. Several of these provide protection against seedling damping-off in greenhouse studies. Subsequent analysis of one of these isolates, NZ130, indicated that the expression of the antibiosis appears to be independent of the carbon or nitrogen source, and is evident only when the Fe+++ concentration is below 10 µM. The size of the zone of P. ultimum inhibition is proportionally increased as the Fe+++ concentration decreases from 10 µM to 0.1 µM. To begin molecular analyses of the mechanism of fungal antibiosis and its role in the soil environment, we are screening a genomic library of NZ130 for the ability to synthesize antifungal compounds. We are also isolating and testing mutants for altered expression of the antibiosis.

A588

CLONING AND EXPRESSION OF SERRATIA CHITINASE GENES IN E. COLI AND PSEUDOMONAS RHIZOBACTERIA. J. Jones, J.S. Ziegle, and T.V. Suslow. Advanced Genetic Sciences, Inc., 6701 San Pablo Ave., Oakland, CA 94608

Many soil bacteria with anti-fungal properties produce chitinase and some have been reported to be effective biological control agents. As a first step in determining the role of chitinase in disease control and fungal inhibition, the genome of Serratia marcescens QMB1466 was cloned into the cosmid vector pLAFR1. Chitinase-producing E. coli clones were identified by the ability to dissolve colloidal chitin in a solid medium. Analysis of chitinase-positive strains revealed that chitinase-encoding cosmids fell into 2 classes, based on restriction enzyme analysis, indicating two DNA regions encoding for chitinase in S. marcescens. These cosmid clones were subsequently conjugated into Pseudomonas rhizobacteria and novel chitinolytic ability was observed.

A589

FUNGI ASSOCIATED WITH SCLEROTIA OF PHYMATOTRICHUM OMNIVORUM IN TEXAS SOILS. C. M. Kenerley, M. J. Jeger, R. W. Jones, Depart. of Plant Pathology and Microbiology, and D. A. Zuberer, Depart. of Soil and Crop Science, Texas Agricultural Experiment Station, College Station, TX 77843

Sclerotia of Phymatotrichum omnivorum (PO) in nylon bags were buried at a depth of 46 cm at three locations at the Blacklands Research Center, Temple, Texas, on 1 September 1983. The crop history at each location was 1) native prairie; 2) agricultural rotation of cotton, corn, and sorghum; or 3) continuous cotton. Sclerotia were retrieved at monthly intervals and assessed for viability and associated fungi by washing, disinfesting, and then plating sclerotia onto minimal salts agar, PO sclerotial extract agar, or moist filter paper. At all locations, subsequent germination of PO sclerotia was highest when no associated fungi were isolated. Isolation of associated fungi from non-germinating sclerotia was greatest at locations 1 and 2. Species of Gliocladium, Penicillium, Aspergillus, Trichoderma, Fusarium, and Paecilomyces (listed in descending frequency of isolation) were most frequently associated with non-germinating sclerotia during the sampling period October1983-February 1984.

A590

WITHDRAWN

A591

INTERACTIONS OF VERTICILLIUM ALBO-ATRUM AND THREE FUNGAL ANTAGONISTS. J. Feinstein and A. L. Morehart, Dept. of Plant Science, Univ. of Delaware, Newark, DE 19717-1303.

The capacity of three antagonistic fungi (Talaromyces flavus, Trichoderma harzianum and Trichoderma viride) to inhibit spore germination in V. albo-atrum (dark mycelial form) was evaluated by germinating spore suspensions of each antagonist on agar-coated slides to which pathogen spores were added. The effects of aqueous infusions of leaf litter from two host and eight non-host tree species on spore germination and hyphal growth of both V. albo-atrum and the antagonists were compared. Only pre-germinated antagonist spores significantly inhibited spore germination of V. albo-atrum. This suppression increased with the length of the pre-germination period. Leaf litter infusions from each tree species significantly stimulated or inhibited spore germination and/or hyphal growth in at least one of the four fungi. Combinations of introduced antagonists with leaf litter or with the interplanting of selected tree species may be useful in the control of Verticillium wilt.

A592

VARIATION IN SENSITIVITY TO GLIOTOXIN AMONG RHIZOCTONIA SOLANI ANASTOMOSIS GROUPS. R.W. Jones and R.E. Pettit, Dept. of Plant Pathology and Microbiology, Texas A&M, College Station, TX 77843

Fungal and bacterial inhibitory properties of gliotoxin, produced by Gliocladium virens have been considered important factors in the biological control of various pathogens by G. virens. Studies were undertaken to determine the effectiveness of gliotoxin in inhibiting growth in seven anastomosis sections of R. solani. Modified Czapek broth (pH 5.8) was prepared for flask cultures. Agar diffusion plates (0.75%) were prepared with the same medium and wells cut at the periphery for application of gliotoxin. The gliotoxin, dissolved in ethanol, was added to the broth cultures, providing solutions of 2 and 4 ppm, with equal amounts added to the wells of the agar plates. Dry wts. of each isolate at seven days, expressed as a percentage of the controls, for 2 and 4 ppm respectively were: AG 1 microsclerotial (83)(67); AG 1 sasakii (0)(0); AG 2-1 (0)(0); AG2-2 (0)(0); AG 3 (80)(68); AG 4 (91)(74); AG 5 (93)(70). Linear growth suppression at five days on agar diffusion plates was similar for all isolates, averaging at 2 and 4 ppm reductions of 31 and 47% respectively.

A593

BACTERIA ANTAGONISTIC TO BACTERIAL AND FUNGAL PATHOGENS P. S. Randhawa, USDA, BARC-W, Beltsville, MD 20705 and N. W. Schaad, Dept. Pl., Soil and Ent. Sci., Univ. Idaho, Moscow, ID 83843.

A seedling bioassay (SBA) chamber was used to select bacteria antagonistic to Pseudomonas solanacearum and Rhizoctonia solani on tomato and cucumber roots, respectively. Tomato and cucumber seeds were treated with 28-35 bacteria isolated from various plant seeds and germinated for 24-60 hrs. Germinated seeds were inoculated with pathogens, placed into SBA chambers and incubated for 4-8 days. Twelve of 28 strains of bacteria

inhibited P. solanacearum on tomato roots by 95% or more whereas only 1 of 35 strains prevented damping-off symptoms on cucumber. In another experiment root colonizing bacterial strains were selected from 14 different field soils directly onto roots of cucumber in SBA chamber. Nine of 24 strains prevented damping-off symptoms when tested separately. Failure to isolate R. solani from excised roots of protected seedlings on PDA containing streptomycin confirmed the absence of pathogen on roots.

A594

INHIBITION OF XANTHOMONAS CAMPESTRIS PV PRUNI BY BACTERIA AND PRUNIPHAGE ON DETACHED PEACH LEAVES. P. S. Randhawa and E. L. Civerolo, BARC-W, USDA, Beltsville, MD 20705.

Inhibition of Xanthomonas campestris pv pruni (X. pruni) by bacteria isolated from apricot and peach leaves and a pruniphage was studied on young detached leaves of greenhouse grown peaches. Detached leaves were washed with 70% ethanol for 1 min and placed on 0.5% water agar with their adaxial side up. Suspensions of test bacteria (A_{600}=0.1) or pruniphage ($2x10-2x10^9$ pfu/ml) were mixed with suspension of X. pruni ($1x10^8$ cfu/ml) in 1:1 ratio and placed on 100 sites on 5 leaves @ 5 ul/site. After incubation for 0 and 120 hr at 25C under 16 hr photoperiod, leaves were assayed for surface and internal population of X. pruni. Four of 47 bacterial strains reduced surface population and 6 reduced internal population by 95% or more. Inhibition on nutrient agar and leaves was not related. Pruniphage at pfu/cfu ratio of 2 reduced surface as well as internal population by more than 95%.

A595

QUANTITATIVE DESCRIPTION OF COLONIZATION OF CERCOSPORIDIUM LEAFSPOT OF PEANUTS BY THE MYCOPARASITE DICYMA PULVINATA. James K. Mitchell, Michael J. Jeger, and Ruth A. Taber, Department of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843.

Colonization of Cercosporidium personatum by Dicyma pulvinata on detached peanut leaves followed a logistic growth pattern. The initial relative rate and the asymptotic level of colonization were calculated by plotting the relative rate against percent colonization. These two parameters, independent of the time lag in colonization, were used to compare growth of the mycoparasite at constant and cycling temperature regimes. No colonization was observed at constant temperatures equal to or greater than 31.5 C. The time lag in colonization increased and the asymptotic level declined markedly at constant temperatures greater than 28 C. The initial relative rate and the asymptotic level of colonization were directly related to the difference between maximum and minimum temperatures especially at maximum temperatures greater than 28 C.

A596

EFFECTS OF 2 NATURALLY OCCURRING BACTERIA ON TILLETIOPSIS WASHINGTONESIS, A BIOLGICAL CONTROL AGENT OF POWDERY MILDEW. Riggs[1], W.A., H. Hartmann[2] and B.R.Currie[1]. M.B. Research & Development Ltd., Box 263, Saanichton, B.C.[1], Saanichton Research & Plant Quarantine Station, 8801 East Saanich Rd., Sidney, B.C.[2].

Two bacteria (Azotobacter sp and Pseudomonas sp) consistantly isolated from greenhouse cucumbers interacted with T. washingtonesis (Tw) and with each other in a complex manner. The bacteria were applied singly, together or in combination with Tw to cucumbers grown under sterile conditions. Growth curves were deliniated by sampling every 72 hours for 12 days. Populations of Pseudomonas were reduced in the presence of Tw, Azotobacter and the two combined. Azotobacter numbers increased in the presence of Tw whether Pseudomonas was present or not. Populations of Azotobacter were reduced in the presence of Pseudomonas only. Numbers of Tw were not changed in the presence of Pseudomonas, however, the average colony size on agar was greatly reduced. Tw increased when in combination with Azotobacter or with both Azotobacter and Pseudomonas. Tw and Azotobacter together grew synergistically.

A597

SCREENING FOR BIOLOGICAL CONTROL AGENTS OF POWDERY MILDEW (SPHAEROTHECA FULIGINEA) ON CUCUMBERS. Hartmann, H., W.A. Riggs and J.W. Hall. Saanichton Research & Plant Quarantine Station, 8801 East Saanich Rd, Sidney, B.C., M.B. Research & Development Ltd., P.O. Box 263, Saanichton, B.C., Research Station, Agric. Canada, Vancouver, B.C.

Eight Tilletiopsis spp and 1 Ampelomyces sp were tested as biological control agents for powdery mildew. In order to maximize number of treatments and account for variability in disease development, plants at 2 leaf stage were laid out in five 10 x 10 latin squares along a greenhouse bench. Each plant was enclosed in a polyethylene bag and sprayed with a control agent. Powdery mildew conidia were applied 24 hr later. Numbers of powdery mildew colonies on each cotyledon and leaf were determined after 7, 10 or 14 days. Reproducable results were obtained with 1st or 2nd leaves, 10 or 14 days after inoculation applied as a proctectant spray. All Tilletiopsis spp suppressed powdery mildew colonies by 90-97% compared to the untreated controls. Ampelomyces did not significantly control the pathogen.

A598

HOST SPECIFICITY OF MELAMPSORA EUPHORBIAE, A PATHOGEN OF EUPHORBIA CYPARISSIAS. E.M. Sutker[1], W.L. Bruckart[2], [1]Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802, [2]USDA, Plant Disease Research Laboratory, Ft. Detrick, Bldg. 1301, Frederick, MD 21701.

Melampsora euphorbiae, an autoecious rust fungus which infects leafy and cypress spurges, may be useful in biological control of these weed species. To date, only 1 of 18 isolates collected from Europe in 1982 was successfully established and evaluated for its potential in biocontrol. The isolate (E20) is capable of infecting two collections of cypress spurge (D49 from Hungary and CCH from Switzerland). None of 22 crop species, 8 economically important species in the Euphorbiaceae, 24 collections of leafy spurge (Euphorbia esula-virgata) or 6 additional collections of cypress spurge were susceptible to E20. Urediniospore germination and formation of appressoria were observed consistently on the non-susceptible cypress spurges. The isolate E20 caused infection of stems as well as leaves, often killing acropetal stem portions.

A599

INDUCTION OF TOLERANCE TO BENOMYL IN COLLETOTRICHUM GLOEOSPORIOIDES F.SP. AESCHYNOMENE BY ETHYL METHANESULFONATE. David O. TeBeest, Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701.

The fungus, Colletotrichum gloeosporioides f.sp. aeschynomene, incites an anthracnose of northern jointvetch (Aeschynomene virginica) and is used as a biological control agent for that weed. Spores were treated with ethyl methanesulfonate (EMS) and plated onto potato-dextrose-agar (PDA) amended to contain 1 ug benomyl per ml media. Four mutant isolates obtained after a single EMS treatment grew and sporulated on PDA and in modified Richards V8 juice media containing from 1 to 100 ug benomyl/ml media. Growth and sporulation of the parent culture was completely inhibited at 1 ug benomyl/ml. Laboratory and greenhouse tests show that these mutants appear to be genetically stable, pathogenically specific and potentially useful as biological agents for A. virginica. The four isolates have been repeatedly transferred in culture and re-isolated from plants without loss of tolerance to benomyl.

A600

ADDITIONS TO THE HOST RANGE OF COLLETOTRICHUM GLOEOSPORIOIDES F.SP. AESCHYNOMENE David O. TeBeest, Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701.

The fungus, Colletotrichum gloeosporioides f.sp. aeschynomene is currently used as a mycoherbicide, COLLEGO™, to control northern jointvetch, Aeschynomene virginica, a leguminous weed. Additional host range tests were conducted in 1983 and a total of 66 species from 44 genera and 11 families have been inoculated in these tests. Crop species within Gossypium, Hibiscus, Cucumis, Cucurbita, Citrullus, Oryzae, Sorghum, Zea, Triticum, Lycopersicon, Spinacia, Brassica, Raphanus, Helianthus, Lactuca, Daucus, Fragaria, Phaseolus, Glycine, Arachis and Vigna were immune. Only Aeschynomene virginica was killed by infection. Several species of Aeschynomene, Lathyrus and Lupinus and 13 of 15 Pisum sativum varieties tested were susceptible to infection. Lesions were found on leaflets, petioles, and stems of Lathyrus, Lupinus and Aeschynomene and also on the tendrils of Pisum. The host range is thus larger than originally described.

A601

SCLEROTINIA SCLEROTIORUM TO CONTROL CANADA THISTLE. B.S. Brosten and D.C. Sands, Plant Pathology Dept., Montana State University, Bozeman, MT 59717.

In field trials conducted on cultivated land infested with

Canada thistle (Cirsium arvense), up to 73% of the thistle shoots died within nine weeks of treatment with Sclerotinia sclerotiorum infested wheat kernel inoculum. Besides attacking the thistle crown and causing wilting and death of the shoots, S. sclerotiorum infected the roots, extending more than 35 cm into the root system. The damage to the root system reduced the thistle shoot density by 8 to 51% the following year, while the density increased over 40% in the control plots. Incorporation of the inoculum into the soil reduced the thistle density to lower levels than surface application. At a pasture site the thistle density was reduced 95% the year following S. sclerotiorum treatment versus a 32% reduction in the control plots. Greenhouse tests have shown this fungus also causes a wilt of spotted knapweed (Centaurea maculosa). Research into the host range and genetics of S. sclerotiorum is underway.

A602

IDENTIFICATION OF TELIOSPORE GERMINATION STIMULATORS IN CANADA THISTLE ROOTS. R. C. French, S. K. Turner, PDRL, USDA, Fort Detrick, Frederick, MD 21701 and E. Piotrowski, ERRC, USDA, Philadelphia, PA 19118.

Root extracts of canada thistle (Cirsium arvense) contain compounds which stimulate germination of teliospores of canada thistle rust, Puccinia punctiformis (syn. P. obtegens), (Turner, Kwiatkowski & Fay, Phytopath. 72 (6) 711:82). Our studies further indicate that the stimulus is volatile and fat soluble, physically similar to other rust spore stimulators such as nonanal, beta-ionone, and 5-methyl-2-hexanone (J. Agr. Food Chem. 31:423:83), the latter most active on urediniospores of P. punctiformis. Active material was obtained from hexane extracts of steam distilled canada thistle roots. Thin layer and gas chromatography revealed four clearly separated spots, with activity in all four spots. GLC-mass spectrographic analysis indicated the presence of 1-heptadecene in spot four, and the other spots were closely related. Bioassays indicated stimulatory activity from 1-hexadecene and 1-octadecene.

A603

FACTORS DETERMINING EFFICACY OF GREENHOUSE WHITEFLY CONTROL WITH VERTICILLIUM LECANII. Riggs, W.A., H. Hartmann and D.P. Elliott. M.B. Research & Development Ltd., P.O. Box 263, Saanichton, B.C., Saanichton Research & Plant Quarantine Stn., Sidney, B.C., Applied Bio-Nomics Ltd., P.O. Box 2637, Sidney, B.C.

Conflicting results have been reported for control of whitefly (Trialeuodes vaporariorum) using V. lecanii. In this study, pathogenicity of 4 V. lecanii strains to eggs, 2nd-3rd instar nymphs, and pupae of whitefly were tested in greenhouses under various relative humidities (RH). The 4 V. lecanii strains were equally pathogenic to whitefly. Under high RH (95-100%), 20-25C) 14% of eggs, 70-100% of nymphs and 80-97% of pupae were diseased within 8 days of inoculation. No significant differences were observed in efficacy of V. lecanii when subjected to 60-70, 70-80, 85-95% RH, 20-25C. Ten to 28% of eggs, 25-50% of nymphs and 20-40% of pupae were diseased. Inspite of the low disease rates achieved under low greenhouse humidities (60-85% RH, 25C), the pathogen's ability to grow saprophytically and its tolerance of some pesticides suggests that it has good potential for use as a biological control agent for whitefly.

A604

EFFECT OF DIFFERENT ISOLATES OF BACILLUS SUBTILIS USED FOR BIOLOGICAL CONTROL OF BEAN RUST UNDER FIELD CONDITIONS. C. Jacyn Baker and J. R. Stavely, Plant Pathology Laboratory, USDA, Beltsville, MD 20705, and Norton Mock, Department of Plant Science, U of DE, Newark, DE 19717-1303.

Previous studies under greenhouse conditions demonstrated biological control by Bacillus subtilis of bean rust, caused by Uromyces phaseoli (Phytopathology 73:1148-1152). In 1982 and 1983, further tests demonstrated that B. subtilis effectively reduces the severity of bean rust under field conditions. When 7-day-old culture filtrates of two different isolates, APPL-1 and PPL-3, were sprayed onto field-grown plants, greater than 75% control was obtained with both isolates. Unlike APPL-1, PPL-3 had marked side effects on field plants and resulted in greener, more succulent plants late into the season with a marked decrease in yield. This apparent hormonal effect by PPL-3 filtrates on the treated plant was reproduced in the greenhouse on bean, as well as on tobacco and cucumber.

A605

RESISTANT REACTION TYPE AGAINST ALTERNARIA LEAF BLIGHT IN CUCUMIS MELO. C. E. Thomas, USDA, ARS, U. S. Vegetable Laboratory, Charleston, SC 29407

Four reported sources of resistance in Cucumis melo against Alternaria leaf blight incited by Alternaria cucumerina were tested for their reaction to the pathogen. The cultivar, Honey Dew Green Flesh, and plant introductions 140471, 164364, and 164756 along with the susceptible cultivar, Perlita, were inoculated with 5.0×10^3 conidia/ml of A. cucumerina. Plants were placed in a high humidity tent for 16 hr at 20°C in the dark and subsequently moved to the greenhouse bench. Lesion number was only slightly reduced on leaves of the four resistant lines compared to Perlita. However, reaction type was markedly different in resistant lines. Lesions were restricted to $\leq$ 1 mm in diameter so that at 10 days total lesion area per unit of leaf area was reduced to 2-6% of that on Perlita. Sporulation per unit of lesion area was reduced to 9-47%. Necrotic lesions on resistant lines remained restricted while those on Perlita continued to expand and coalesce until leaves died.

A606

THE INFLUENCE OF MELOIDOGYNE INCOGNITA ON VASCULAR WILT OF SUMMER SQUASH CAUSED BY FUSARIUM OXYSPORUM F. SP. NIVEUM. M. Caperton, R. D. Martyn, and J. L. Starr. Dept. of Plant Pathology and Microbiology, Texas A&M University, College Station, TX 77843

Greenhouse experiments were conducted to determine if the root-knot nematode, Meloidogyne incognita, influenced the total percentage wilt or the timing of the wilt symptom caused by Fusarium oxysporum f. sp. niveum on yellow summer squash (Cucurbita pepo var. melopepo). Seedlings of Early Prolific Straigthneck (EPS), moderately wilt resistant, and Goldneck (GN), highly resistant, were evaluated with each pathogen alone and in combination. No nematode interaction was observed with Fusarium on GN; however, with EPS there was both an increase in percentage wilt development and an earlier onset of symptoms when both pathogens were present. Wilt was observed in 14 days on EPS when both pathogens were present and totaled 85%. With Fusarium alone, EPS wilted at 19 days and totaled only 35% wilt. No significant wilt was observed with any of the pathogen treatments on GN.

A607

THE EFFECT OF LIGHT ON SPORULATION OF PERONOSPORA PARASITICA ON CABBAGE SEEDLINGS. Proctor, R.H., H. Hartmann and C.J. French. Dept. of Biology, University of Victoria, Victoria, B.C., Saanichton Research & Plant Quarantine Station, 8801 East Saanich Road, Sidney, B.C.

Inhibitory effects of light on Peronospora parasitica sporulation were investigated. All tests were carried out under fluorescent and incandescent light (75% : 25%) 110uE $s^{-1}m^{-2}$ at 13 $\pm$ 2C. Contin ous illumination reduced sporulation of P. parasitica on intact seedlings by 95%. Doubling the light intensity made no significant difference in the number of sporangia produced. Inhibiting photosynthesis in infected seedlings by application of 10^{-4} M Diuron (DCMU) 2, 4 or 6 days after inoculation did not significantly affect sporulation. Interrupting the scotophases following the first or second day of sporulation by 1 hr resulted in 55-70% inhibition. Reversal of inhibition occurred when both scotophases were interrupted in sequence.

A608

CERCOSPORA BLIGHT OF ASPARAGUS IN OKLAHOMA. K. E. Conway and J. E. Motes. Departments of Plant Pathology and Horticulture and L. A., Oklahoma State University, Stillwater, OK 74078

Cercospora blight, caused by Cercospora asparagi, has potential to become a major disease in asparagus in central and eastern Oklahoma. It was first identified in 1980 at the Vegetable Research Station, Bixby, OK. Incidence of Cercospora blight increased as new production areas were established. The pathogen overwinters on fern and as residue on soil during harvest (March-May). Lesions on seedlings are prevalent during this period. Conidial densities were monitored using a Kramer-Collins 7 day sampler during 1982 and 1983. Fern growth closes between rows during July and coincides with increased aerial conidial densities. Lesion formation and defoliation are most prevalent at the base of ferns during initial stages of the epidemic. Ferns will be completely diseased by September. Use of a fungicide (chorothalonil) during 1982 to control disease in field plots resulted in significant yield increases compared to non-sprayed controls.

A609

THE EFFECTS OF CROP RESIDUES ON POPULATIONS OF FUSARIUM

OXYSPORUM F. SP. APII RACE 2 IN SOIL AND RESULTING DISEASE IN CELERY. W.H. Elmer and M.L. Lacy, Dept. of Botany and Plant Pathology, Michigan State Univ., East Lansing, MI 48824-1312

A color mutant of F. oxysporum f. sp. apii Race 2 (FOA2) was produced that gave distinctive orange-colored colonies on Komada's agar, allowing enumeration of FOA2 in soil without enumeration of F. oxysporum saprophytes. Muck soils were infested with FOA2 propagules and amended with 1 g of several crop residues per liter soil. FOA2 populations increased within 3 wk in celery-amended soils and decreased in soils amended with mint, sudex and sorghum when compared to nonamended soils. Soils amended with onion, potato and rye had populations of FOA2 similar to controls. After 9 wk, all residue-amended soil had similar FOA2 populations. However, Fusarium yellows was significantly more severe when celery 'Tall Utah 52-70R' was grown in naturally infested soil amended with celery residues than in other amended or nonamended soils, suggesting that the incorporation of celery trimmings into field soil at harvest should be avoided.

A610

ECOLOGY AND EPIDEMIOLOGY OF TOMATO SPOTTED WILT VIRUS (TSWV) AND ITS VECTOR, FRANKLINIELLA OCCIDENTALIS. J. J. Cho, W. C. Mitchell, L. Yudin, and L. Takayama. University of Hawaii, Maui Research, P. O. Box 269, Kula, HI 96790 and Honolulu, HI 96822.

TSWV causes severe losses of lettuce in Hawaii's major vegetable growing area in Kula, Maui. Data from monitoring and sampling at farm sites in our IPM program have identified interrelationships between TSWV, its insect vector, reservoir weed hosts and weather factors which may be useful in developing predictive models for disease outbreaks. Twenty five weeds found in Kula serve as reservoirs for F. occidentalis 17 of which may harbor TSWV. Seven weeds are new recordings as virus hosts including: Amaranthus sp., A. viridis, Bidens pilosa var. minor, Crotalaria incana, C. mucronata, Verbena rigida, and Verbescina encelioides. Thrips populations increase in mid to late May with peaks occuring during the summer. In lettuce fields there is a high correlation between thrips populations and TSWV incidence. Thrips populations associated with a major tree legume (Leucaena glauca) show a high correlation between numbers found in flowers and adjacent farm catches.

A611

EFFECT OF SOIL SALINITY ON DEVELOPMENT OF PHYTOPHTHORA ROOT ROT OF CITRUS. N. S. Blaker and J. D. MacDonald, Department of Plant Pathology, University of California, Davis, CA 95616

In the Coachella Valley, CA, a field survey indicated that root rot of citrus caused by Phytophthora parasitica increased with increasing soil salinity. In experiments with hydroponically-grown seedlings of Troyer citrange and Pineapple sweet orange rootstocks, plants were briefly exposed to high levels of salinity (EC=22dS/m) prior to inoculation. These treatments did not alter the susceptibility of Troyer to root rot, but Pineapple seedlings were predisposed to disease. When plants were grown for 9 wk in soil salinized to an EC_e of 3-4 dS/m, total root growth and production of new roots by Troyer seedlings was greatly inhibited. In the presence of P. parasitica, plants at an EC_e of 3-4 dS/m had 30% root rot, while plants in non-saline soil had only 10%. Similar results were obtained with Pineapple seedlings. These results suggest that in saline soils lack of root regeneration as well as predisposition may contribute to the severe root rot observed under field conditions.

A612

EVALUATION OF ISOZYMES FOR THE IDENTIFICATION OF ISOLATES OF PHYTOPHTHORA MEGASPERMA. C. K. Elliott and D. P. Maxwell, Dept. of Plant Pathology, Univ. of Wisconsin, Madison WI 53706

Eleven enzymes from isolates of Phytophthora megasperma (Pm) from several hosts and from various geographic origins were examined by means of starch gel electrophoresis. The three recognized forma speciales, f.sp. medicaginis (Pmm) on alfalfa, f.sp. glycinea (Pmg) on soybean, and f.sp. trifolii (Pmt) on arrowleaf clover, had zymograms distinct from each other and from isolates of Pm from other hosts. No isozyme variation for the eleven enzymes was detected within Pmg, regardless of race or geographic origin. Generally, zymograms for isolates of Pmm were the same except for one isolate from California and one from Australia. These two isolates had identical zymograms but differed from the other Pmm isolates by one isozyme band for malate dehydrogenase and malic enzyme. Cluster analysis of isozyme patterns indicated that Pmg, Pmm, and Pmt can be distinguished by their respective zymograms and that Pmg and Pmm are not closely related.

A613

COMPUTER SIMULATION OF EMERGENCE OF DRY BEANS AS AFFECTED BY RHIZOCTONIA SOLANI. Ariena H. C. Van Bruggen, C. H. Whalen and P. A. Arneson, Cornell University, Ithaca, NY 14853

Emergence of dry beans was simulated in a model written in the Continuous System Modeling Program (CSMP) language. The model consisted of two sub-models: (1) development and growth of an "average" seedling, simulating imbibition, activation of reserves, and conversion of reserves into roots, hypocotyl and primary leaves, and (2) establishment of a population of seedlings, simulating skewed distributed delays in development. The driving variables were soil water potential and temperature. Rhizoctonia solani increased the delays and their skewness; these effects were accounted for in sub-model 2. In sub-model 1 the effect of R. solani was simulated by reduced translocation and increased respiration. Death of seedlings was a function of inoculum level, soil moisture and temperature. Values of parameters and growth rates were obtained from incubator and growth chamber experiments. Results from field experiments were used to validate the model.

A614

EFFECT OF SEEDLING RATE ON PROGRESS OF DAMPING-OFF CAUSED BY RHIZOCTONIA SOLANI. M.L. Putnam. Dept of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706.

American ginseng (Panax quinquefolium) seedlings were planted such that adjacent plants were 1.2, 2.5, 3.8, 5, or 7.6 cm apart in 22.5 x 29.5 cm aluminum pans, with 4 replicates per seeding rate. A small portion (0.1 g) of a 10-day-old culture of Rhizoctonia solani, growing on a cornmeal-sand medium, was placed in an area about 1 x 1 cm at one point at the edge of the pans. Post-emergence damping-off was measured daily for 20 days. The rate of disease progress was greatest at the highest seeding rate and least when adjoining plants were 7.6 cm apart; i.e. there was a negative correlation between plant density and the rate of disease progress. Damping-off is a major problem in commercial ginseng production. This study indicates that by modifying seeding rate growers may reduce seedling mortality.

A615

EFFECT OF CHEMICAL TREATMENT OF SOIL OR SEED PIECE ON EMERGENCE AND YIELD OF POTATOES INOCULATED WITH RHIZOCTONIA SOLANI AG-3. D.E. Carling. Agricultural Experiment Station, Univ. of Alaska, Box AE, Palmer, Alaska 99645.

Potato seed pieces were inoculated in the field by placing R. solani AG-3 colonized barley kernels above the seed piece in the planting hole. Chemical treatments were applied to the soil (PCNB) or to the seed piece (thiophanate methyl) just prior to planting. Forty-five percent of inoculated plants failed to emerge due to activity of R. solani AG-3. Individual seed pieces in this category produced as many as 9 sprouts, each being killed prior to penetrating the soil surface. Of the non-inoculated plants, only 2.2% failed to emerge. Inoculation severely reduced per-plant yields. Inoculated plants yielded at approximately 10% the rate of non-inoculated plants. PCNB soil treatment and thiophanate methyl seed treatment had no effect on potato tuber yield, nor upon recovery of R. solani at seasons end.

A616

EFFECT OF BIOCIDAL TREATMENTS ON CATION EXCHANGE CAPACITY AND FUSARIUM BLIGHT OF SOYBEANS IN FIVE DELAWARE SOILS. H. A. Sandler, R. B. Carroll and D. L. Sparks, Dept. of Plant Science, Univ. of Delaware, Newark, DE 19717-1303.

The effects of autoclaving and Vapam fumigation on cation exchange capacity, organic matter content, pH and Fusarium wilt of soybean were determined for five different Delaware soil types. Essex soybeans were grown in the treated soils which were infested, prior to planting, with 6.3×10^4 viable spores/g dry soil of Fusarium oxysporum. Treatments were arranged on the greenhouse bench in a randomized block design with five replications. Soil cation exchange capacity generally increased with fumigation and decreased with autoclaving but differences were not significant for all soils. Organic matter content increased following fumigation in three soils and pH values were lowered by both biocidal treatments in all soils except a Matapeake loam. Disease incidence and severity was affected differently for each soil type. A wilt-conducive Sassafras soil was the only one to show a correlation between changes in the chemical properties caused by the treatments and the occurrence of wilt.

A617

THE RELATIONSHIP BETWEEN SHORT-TERM ADHERENCE AND LONG-TERM COLONIZATION OF ROOTS BY BACTERIA. Douglas W. James, Jr.,

Trevor V. Suslow, and Katherine E. Steinback. Advanced Genetic Sciences, Inc., 6701 San Pablo Ave., Oakland, CA 94608

Radish seedlings were incubated for 20 min. in various bacterial suspensions (10^4-10^8 cfu/ml). The seedlings were removed, washed, and the relative adherence was determined by homogenizing the roots and dilution-plating the homogenate. All rhizosphere and soil bacteria which under controlled lab and greenhouse tests were good long-term colonizers also showed strong, concentration-dependent adherence. A non-soil isolate (E. coli) showed 100X less adherence. Optimum adherence for Pseudomonas fluorescens E6-22 was at pH 7.0 in the presence of 5 mM $MgCl_2$ or $CaCl_2$. The monovalent cations Na^+ and K^+ had no effect. The hydrophobicity of the cell surface of the various strains, as measured by partitioning of bacterial suspensions into hexadecane, had no direct bearing on adherence to radish roots. Considering cation and pH effects, electrostatic phenomena may partially explain short-term adherence, which appears to be an important prelude to colonization.

A618

ROLE OF IN VITRO ANTAGONISTIC FLUORESCENT PSEUDOMONADS IN SOILS SUPPRESSIVE TO BLACK ROOT ROT OF TOBACCO. Stutz, E., Défago, G., and Kern, H. Institut für Phytomedizin , Eidgenössische Technische Hochschule, ETH-Zentrum, 8092 Zürich, Switzerland

Fluorescent pseudomonads (Flp) were isolated from soils conducive and suppressive to Black Root Rot of Tobacco (Thielaviopsis basicola, Tb). These soils occur in the same area but are of different geological feature. Only the Flp strains isolated from suppressive soils inhibited the growth of Tb on malt agar plates. Roots of cotton, peanuts, peas and cherry trees were exempt from Black Root Rot in suppressive soils and colonized by Flp antagonistic to Tb. In conducive soils, the roots were diseased and had no antagonistic Flp. Heat treatment (60 C, 20 min.) of the suppresive soils destroyed the Flp but only half of the suppressive capacity. Similarly soil samples taken from 60-120 cm depth were without Flp and showed only half of the suppressive capacity compared with samples from above layers of the same geological features. Therefore, Flp, which in vitro inhibit Tb, seem to play a role in the suppressive mechanism, but other factors must also be considered.

A619

POPULATION DYNAMICS OF THIELAVIOPSIS BASICOLA IN SOILS CONDUCIVE AND SUPPRESSIVE TO BLACK ROOT ROT DISEASE. Berling, C.H., Défago, G. and Kern, H. Institut für Phytomedizin, Eidgenössische Technische Hochschule, ETH-Zentrum, CH-8092 Zürich

A Thielaviopsis basicola strain C017 marked with benomyl resistance was used to study the dynamics of the fungal population in soils conducive and suppressive to Black Root Rot (T. basicola). Known amount of C017 endoconidia were added to each of the soils and, over a period of 3 weeks, strain C017 was reisolated and colonies enumerated. In the absence of the host plant (tobacco) no difference was observed between the populations of T. basicola in conducive and suppressive soils. Populations remained constant with a reisolation rate of 1/10 of the originally added population. In heat treated soils, the populations increased after two days and then decreased after 4 days. In the presence of the host the parasite population increased in conducive soils near the roots and remained small in suppressive soils. This indicates that the suppressive principle is bound to the rhizosphere.

A620

THE DISEASE-SUPPORTING CAPABILITY OF SOILS OF DIFFERENT FUNGISTATIC CAPACITY. T. Isakeit and J. L. Lockwood, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

Relative wilt incidence in three soils artificially infested with chlamydospores of Fusarium oxysporum f. sp. lycopersici and F. oxysporum f. sp. conglutinans was compared with degree of fungistasis in the soils. Tomato wilt incidence was significantly greater ($P=0.05$) in Capac loam (L) (30%) and Boyer sandy loam (SL) (20%) than in Colwood L (5%). Chlamydospore germination on soils amended with 300 µg glucose + 60 µg peptone/g was in the same order: 92% for Capac L, 76% for Boyer SL, and 54% for Colwood L, reflecting different intensities of fungistasis. Radish wilt incidence was significantly greater ($P=0.05$) in Boyer SL (77%) than in Capac L (45%), while Colwood L (52%) was intermediate. However, chlamydospore germination was 56% on Boyer SL as compared with 78% on Capac L and 8% on Colwood L. These results suggest that differences in disease incidence cannot solely be explained by differences in the levels of fungistasis.

A621

SILICA GEL AS A SUBSTRATE FOR STORING FUSARIUM SPECIES. Carol E. Windels and Thor Kommedahl. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

Over 450 single-spored cultures of Fusarium spp. were stored in both silica gel (SG) and sterile soil for 30-36 months at 5 C. The SG medium was prepared by placing 3 cc of crystals (6-12 mesh, Grade H, Type II) into 13x100 mm screw cap culture tubes, which were then dry heat sterilized at 180 C for 1.5 hr. Spore suspensions were prepared in 7% Difco skim milk from actively sporulating 7-14 day old cultures. About 0.25 ml of suspension was added to SG to moisten the crystals without causing them to clump. Numbers of cultures tested included: F. acuminatum (58), F. avenaceum (8), F. culmorum (11), F. episphaeria (14), F. equiseti (27), F. graminearum (18), F. lateritium (16), F. moniliforme (26), F. oxysporum (154), F. sambucinum (16), F. semitectum (2), F. solani (63), F. subglutinans (13), and F. tricinctum (35). Nearly all isolates grew from SG (94%) and from soil (95%), except for F. graminearum which was recovered from 6% of the SG cultures and 100% of the soil cultures.

A622

BAITING PHYTOPHTHORA CACTORUM FROM NATURALLY INFESTED SOIL. S. N. Jeffers and H. S. Aldwinckle, Dept. Plant Pathology, Cornell Univ., NYS Agr. Exp. Sta., Geneva, NY 14456.

Apple cotyledons were better than apple seedlings, apple leaf pieces, and pear or apple fruits for recovering Phytophthora cactorum from naturally infested soils. Air-drying and then moistening (ADM) soil samples prior to flooding and adding baits greatly enhanced detection. Successful baiting was optimum at 20 C when compared with 16, 25, and 28 C. The advantages of using apple cotyledons in an ADM baiting procedure were: (i) detection of P. cactorum was repeatedly more successful with cotyledons than with pear and apple fruits, the conventional baits; (ii) P. cactorum was consistently recovered from soils that tested negative when baited without ADM; (iii) cotyledons were easily produced and handled; (iv) P. cactorum was identified sporulating on necrotic cotyledons; (v) Pythium contamination rarely interfered with detection. In addition, apple cotyledons were effective for recovering P. cambivora and P. cryptogea from soil but apparently were ineffective for P. megasperma.

A623

PRODUCTION OF MONILIOID CELLS IN ROOT CELLS BY BINUCLEATE RHIZOCTONIA ISOLATES. R. S. Ferriss, A.-C. McGraw & J. W. Hendrix, Dept. of Plant Pathology, Univ. of Kentucky, Lexington KY 40546.

During examinations of stained roots for endogonaceous mycorrhizae, we have frequently observed the presence of isolated cortical cells containing structures resembling monilioid cells of Rhizoctonia spp. Incidence of these structures in roots of sorghum x sudangrass plants grown in soil samples from a sorghum x sudangrass field indicated populations of 0 to 0.5 propagules/g dry soil. Isolations from roots on Rhizoctonia-selective medium mainly yielded binucleate isolates resembling and anastomosing with a Ceratobasidium anastomosis group 2 tester. The isolates produced short chains of monilioid cells, but no true sclerotia, on several media. Inoculation experiments indicated that the isolates can produce monilioid cells in cortical cells of sorghum x sudangrass, soybean and tobacco roots, and can cause moderate growth reductions of these hosts. Groups of monilioid cells in host cells appear to function as a sclerotium-like dispersal and survival unit for these fungi.

A624

THE EFFECT OF ROOT ORGANS, EXUDATES AND EXTRACTS ON HYPHAL ELONGATION OF GLOMUS FASICULATUS IN AXENIC CULTURE. K.S. Elias and G.R. Safir, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

The effect of phosphorus (P) nutrition on root exudates, root extracts and root organs and subsequent effects on hyphal elongation of G. fasiculatus in axenic culture were investigated. P-deficient and normal white clover (Trifolium repens) plants were hydroponically grown. Root exudates were collected 3 times at 2-week intervals and aqueous extracts of roots were made at 6 weeks. A root organ culture medium amended with concentrated (10X) exudates or extracts from 50 plants per treatment was used. Surface-sterilized G. fasiculatus chlamydospores were placed on the agar surface with or without (1) exudates or extracts, (2) phosphorus or (3) P-deficient or normal root organs. Exudates and extracts from P-deficient whole plants and root organs increased hyphal elongation. This suggests

that under low P nutrition overall exudation is increased or specific metabolites are produced in sufficient amounts to improve hyphal growth from germinated mycorrhizal spores.

A625

PATHOGENICITY OF FUSARIUM TRICINCTUM, F. POAE AND F. SPOROTRICHIOIDES ON MAIZE EARS. A. A. Al-Heeti, R. W. Caldwell and E. B. Smalley, Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Pathogenicity of Fusarium tricinctum, F. poae and F. sporotrichioides was assessed on a range of maize lines including 4 sweet corn, 8 dent corn and 2 pop corn types. Artificial inoculation in the field 6 days after silking utilized either the toothpick method (TP) or a spore suspension injected into wounded kernels (SW). Disease severity at harvest was scored. Sweet corn was the most susceptible, and Style Pake developed the most severly damaged. Its mean ear rot varied from 16 to 50% depending upon the inoculation method and species of Fusarium used. Dent corn and pop corn were less susceptible. No significant differences in pathogenicity were observed between the Fusarium isolates. Within species, however, the pooled mean DR of all F. tricinctum isolates (1.08) was significantly different (p=0.05) from F. poae (0.89) and F. sporotrichioides (0.90).

A626

GERLACHIA NIVALIS (FUSARIUM NIVALE), A NEW SNOW MOLD ON WINTER CEREALS AND GRASSES IN ALASKA. Jenifer Huang McBeath, Agricultural Experiment Station, University of Alaska, Fairbanks, Alaska 99701.

A disease displaying the typical symptoms of pink snow mold (Gerlachia nivalis=Fusairum nivale) was found in Fairbanks in spring 1983, following a relatively mild winter. After snow melt, abundant pinkish-colored patches of infected plants were observed on lawns and winter cereal fields. Examination of the infected tissues revealed the presence of numerous salmon-colored sporulating sporodochia. Conidia were mostly curved with tapered tips, having one septate. Neither Chalmydospores nor perithecia were found. Gerlachia nivalis appeared to survive Alaskan enviromental conditions well. This disease has been observed on newly established lawns with out-of-state seed sources as well as on old farm lands where mostly Alaskan grown seeds were sown. No indication of antagonism was noticed between G. nivalis and other snow molds.

A627

GROUPS OF DIAPORTHE/PHOMOPSIS ISOLATES OBTAINED FROM CULTIVATED SUNFLOWER. S. M. Yang and T. J. Gulya, Jr. USDA-ARS, Bushland, TX 79012; North Dakota State Univ., Fargo 58105, respectively.

We studied sunflower isolates of Diaporthe/Phomopsis from Ohio (D108 and 12.82.4), North Dakota (Ph-6, Ph-7, and Ph-10), Texas (ATCC 52763), and Yugoslavia (MC-1). A total of seven isolates were grown on PDA and on autoclaved sunflower leaves and stems on moistened filter paper in a petri dish. The plates were incubated on laboratory benches (22 ± 2C) and illuminated 12 hrs daily with fluorescent light. The seven isolates were divided into five groups according to the formation of fruiting bodies: Group 1: perithecia had short beaks (<536 µm long) (Ph-7 and ATCC 52763); Group 2: perithecia had long beaks (>853 µm long) (Ph-10); Group 3: pycnidia had only "A" pycnidiospores and no perithecia formed (12.82.4); Group 4: pycnidia had only "B" pycnidiospores and no perithecia formed (Ph-6 and MC-1); Group 5: neither pycnidia nor perithecia formed (D108). These results indicate that isolates of Diaporthe/Phomopsis attacking sunflower may belong to more than one species or biotype.

A628

EVALUATION OF SORGHUM BICOLOR (L.) MOENCH ACCESSIONS UNDER NATURAL INFECTION WITH YELLOW SORGHUM STUNT MYCOPLASMA. M. A. Langham, R. W. Toler, J. D. Alexander, and F. R. Miller, Texas A&M University, College Station, Tx 77843.

Yellow sorghum stunt is the only disease of sorghum, Sorghum bicolor (L.) Moench, known to be incited by a mycoplasma. One hundred and seven sorghum accessions were rated for disease severity and incidence due to natural infection in 1983 field tests. One hundred percent incidence was observed in some lines. The fifty-five accessions which had no symptoms were classified as having a severity rating of 1. Twenty-seven accessions with localized mild yellowing were rated 2 with incidences of 5-60%. Six accessions with mild yellowing which spread were rated 3 and had incidences of 5-40%. Thirteen accessions with intense yellowing over large areas were rated 4 and ranged in incidence from 5-60%. Six accessions with severe yellowing covering the majority of the plant with stunting were rated 5 with incidences from 5-60%. The five accessions with 100% incidence had average severity ratings of 3.6, 3.0, 3.0, 3.0, and 3.0.

A629

Effects of Soil Moisture, Soil Temperature, and Soil-borne Organisms on Sorghum Seedling Vigor. G.A. Forbes, and R.A. Frederiksen, Dept. of Plant Pathology and Microbiology, Texas Agr. Exp. Sta., College Station, Tx., 77843

Variation in moisture levels (-1 bar through saturation plus 24 hr of flooding) was more important than variation in temperature levels (10 through 25C) in explaining reduction in leaf length, leaf weight, and final emergence of sorghum seedlings. Comparing seedlings grown in pasteurized field soil (PFS) with those grown in non-pasteurized field soil (NFS) demonstrated that the deleterious effect of high moisture levels (saturation and saturation plus 24 h of flooding) was indirect, as the moisture effect was more pronounced in the NFS than in the PFS. Moisture X temperature interactions for leaf length, leaf weight, and final emergence were very small compared to the main moisture effect on the same variables. These data suggest that high soil moisture may have a greater effect on seedling stand establishment in the field than low soil temperature.

A630

FIELD EVALUATION OF SEVERAL SYSTEMIC AND PROTECTANT FUNGICIDES TO CONTROL BENOMYL,-THIOPHANATE-AND THIABENDAZOLE-RESISTANT STRAINS OF Cercospora beticola ON SUGAR BEET. James A. Percich and Michael W. Hotchkiss, Dept. of Plant Pathology, Univ. of Minnesota, St. Paul 55108.

Six systemic and four protectant fungicides were evaluated for control of benomyl,-thiophanate-and thiabendazole-resistant strains of Cercospora beticola on sugar beet. The systemic fungicides imazalil, nuarimol and propiconazole on three sugar beet varieties resulted in greater disease control, higher yields and recoverable sugar, greater monetary return, lower impurities (Na, K and Amino-N µg/ml) at 1.42, 1.63 and 0.58 l/ha, respectively, than any other treatment. Statistically nuarimol and propiconazole were significantly better than imazalil but were similar to each other. Triphenyltin hydroxide at 0.7 l/ha provided significantly better disease control, sugar yield and quality than all fungicides tested, but was not statistically different from nuarimol and propiconazole.

A631

A HAND-HELD FORCE GAUGE FOR RAPID ASSESSMENT OF FIELD CORN STANDING STRENGTH. R. B. Carroll and K. J. Byrnes, Dept. of Plant Science, Univ. of Delaware, Newark, DE 19717-1303.

A force gauge was fitted with a Y-shaped extension to fit against corn stalks. Readings were made by placing the extension against a stalk at a standard height and pushing until the corn stalk reached an angle of 45° or greater from the vertical. The gauge records the greatest force encountered in the motion and can be returned to zero for the next stalk by pressing a reset button. During the 1983 growing season, 1890 stalks were evaluated for standing strength using this method. Natural lodging also was recorded and stalk rot was determined by splitting stalks longitudinally from the ear to crown and visually recording the incidence and severity of infected internodes. None of the variables measured were highly correlated but the force gauge was a better predictor of lodging than the stalk rot rating. An evaluation of 240 additional stalks indicated only a low correlation among stalk standing strength, visual stalk rot ratings and stalk firmness as determined by squeezing tissue between the fore-finger and thumb.

A632

FUNGI ISOLATED FROM ROTTED CORN STALKS ON TILLED AND NO-TILL SOILS IN DELAWARE. K. J. Byrnes and R. B. Carroll, Dept. of Plant Science, Univ. of Delaware, Newark, DE 19717-1303.

During the 1982 and 1983 growing seasons, 210 lower stalk sections were collected from tilled and no-till corn fields in Delaware on a random, state-wide basis. Stalks were selected by observing external symptoms and softness of lower internodes. Rind sections (1x1 cm) were taken from three different sites, surface disinfested and cultured on acidified potato-dextrose agar to facilitate identification of isolated fungi. Based on symptoms and isolations, the major stalk-rotting pathogen was determined for each stalk. Pathogens confirmed for corn grown in tilled soil were: 53% Gibberella spp., 20% Stenocarpella maydis (= D. maydis), 17% Colletotrichum graminicola and 11%

miscellaneous fungi. From no-till corn, the percentages were 33, 42, 17 and 8, respectively. Spore suspensions of selected isolates of these fungi were inoculated into stalks of six different corn hybrids grown in till and no-till soils. Disease ratings were in the order of C. graminicola > S. maydis > Gibberella spp. for both tillage systems.

A633

INVOLVEMENT OF CYLINDROCOPTURUS ADSPERSUS IN THE PREMATURE RIPENING COMPLEX OF SUNFLOWER. T. J. Gulya and L. D. Charlet, USDA-ARS, Research Plant Pathologist and Research Entomologist, North Dakota State University, Fargo, ND 58105

Previous studies have shown that several fungi may be involved in premature ripening (PR) of sunflower, including Alternaria helianthi, Phoma macdonaldii, and various Fusarium spp. This 1983 study investigated the possible involvement of various stalkfeeding insects. Twelve commercial sunflower fields in North Dakota were examined. The stem weevil, Cylindrocopturus adspersus, was found in every field exhibiting PR. Plants with PR had 29 larva/stalk compared to 12/stalk for non-PR plants. PR plants yielded 56% of that of non-affected plants. In another study, efficacy of the fungicides benlate and maneb and the insecticides aldicarb, carbofuran, and malathion were evaluated under conditions of natural infection. None of the treatments reduced leaf spotting or rate of senescence, but all insecticides significantly reduced the number of stem weevil larva/stalk and lodging, and increased yield.

A634

INCIDENCE OF ALFALFA ANTHRACNOSE IN OKLAHOMA. S. J. Allen, G. L. Barnes, and J. L. Caddel, Departments of Plant Pathology and Agronomy, Oklahoma State University, Stillwater 74078.

Oklahoma data on whether anthracnose (Colletotrichum trifolii Bain & Essary) can reduce stand and forage yield conflicts. Data on occurrence of anthracnose symptoms and successful isolation of C. trifolii were compared to precipitation patterns at 5 sites over a 3 yr period. Yearly variation in precipitation distribution affected symptomatology and degree of disease development. During 1973-1974, an unusually high amount of rainfall occurred during late summer and fall. Anthracnose damage became apparent in 1975. During subsequent years, precipitation distribution was near normal, and no yield reduction attributable to anthracnose was observed. Damage by anthracnose occurs in Oklahoma, and resistance to C. trifolii is an important consideration in development of semi-dormant cultivars for the state.

A635

A LABORATORY METHOD OF POSSIBLE USE FOR ASSESSING RESISTANCE OF SOYBEANS TO WHITE MOLD. D. Chun and J. L. Lockwood, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

An in vitro assay for testing susceptibility of soybean varieties to white mold is being evaluated. Greenhouse-grown soybean plants, 3 to 4 weeks old, are cut off at ground level. Lateral leaves are excised. The stems are placed on moist silica sand in 26 x 18 x 6 cm trays lined with plastic film. Inoculum consists of 5-mm-diam. disks taken from cultures of Sclerotinia sclerotiorum grown for 5 days on 2% millet seed agar in petri dishes. The inoculum disks are coated with 0.3% water agar, then placed at the crotch formed by the stem and petiole of the first trifoliolate leaf. The trays are covered to retain moisture and tilted to keep the sand moist but unsaturated. Lesion lengths are determined 6 days after inoculation. Results suggest that the method may be useful for assessing the susceptibility/resistance of soybean varieties to white mold.

A636

SEED SURVEY FOR KERNEL SMUT OF RICE IN CALIFORNIA. Matsumoto, T., D. Showers, D. Luscher, D. Higuera, C. Krass and A. French Department of Food & Agriculture, 1220 N Street, Room 340, Sacramento, CA 95814.

Kernel smut of rice, caused by Tilletia barclayana (=Neovossia horrida) has been found to be widespread in California, especially in the northern Sacramento Valley. By means of a seed wash-centrifuge technique, samples representing 382 rice seed lots, gathered from storage facilities in eight counties, were tested. Chlamydospores of the kernel smut fungus were recovered from 16% (61) of these samples. The identifications were confirmed using scanning electron microscopy and by chlamydospore germination. Other smut fungi from weed hosts (e.g. Ustilago utriculosa) and fungal spores of Alternaria spp. and Epicoccum spp. were also found in the seed samples. Thirteen rice varieties were found to be infested with kernel smut. The varieties S-201, M-9, M-201 and M-302 contained the highest spore concentrations. Pathogenicity tests and field surveys will be conducted.

A637

RELATION OF ENERGY RESERVES IN ULMUS AMERICANA TO SUSCEPTIBILITY TO CERATOCYSTIS ULMI. D. Zimel, R.J. Campana and A.L. Shigo. Dept. of Botany and Plant Pathology, University of Maine, Orono 04469.

Because high susceptibility of Ulmus americana to Ceratocystis ulmi, coincides with low energy reserves in June, a study was made of energy reserves before, during, and after this period. Xylem tissue samples were obtained by increment hammer monthly from April-August from 30 small elms (5-10 cm dbh). Starch and sugars were extracted from samples and determined colorimetrically. On June 15 trees were inoculated with C. ulmi. Extracted sugars were identified by descending paper chromatography. Starch levels were maximum in April and August, and minimum in June; and sugar was minimum in July. Starch reserves were less ($P = 0.05$) in inoculated than in control trees in June. Concentrations of sugars were not different from one another in any period. Concentrations of sucrose, glucose, and fructose were greater than those of raffinose or maltose. The data suggest that low energy reserves may enhance susceptibility to C. ulmi.

A638

LATENT COLONIZATION OF SEEDLING AND SPROUT OAKS BY HYPOXYLON ATROPUNCTATUM. P. Fenn, R. T. Holland and D. L. Nida, Dept. of Plant Pathology, University of Arkansas, Fayetteville, AR 72701.

Living asymptomatic tissue samples were surface-sterilized and plated on yeast extract-glucose agar to determine whether H. atropunctatum latently infected small (0.3-1.0m tall) understory oaks. The fungus was isolated from stems of 30-80% of the seedling sprouts of red, black, blackjack, white and post oaks. In more detailed studies, the fungus was isolated from stems (44%), petioles (51%), midveins (27%) and leaf blade tissues (7%) of white oak (Quercus alba) seedling sprouts, and from 29%, 48%, 19% and 8%, respectively, of black oak (Q. velutina) seedling sprouts sampled. H. atropunctatum was also isolated from living stem tissues of vigorous stump sprouts. The fungus was not isolated from stems or roots of 6-wk-old white and black oak seedlings grown from surface-sterilized acorns in sterile soil in a growth chamber. Latent infection of oak regeneration helps explain the latency of this fungus in healthy saplings and mature trees. Sprout and seedling oaks may be useful for studies of the survival and spread of this fungus in oak stands.

A639

BACTERIAL POPULATION CHANGES IN WHITE SPRUCE NURSERY SEEDLINGS. N.J.Phillips-Luckai, School of Forestry, Lakehead University, Thunder Bay, Ontario, CANADA P7B 5E1.

White spruce seedlings were sampled at several stages of development, under both storage and seedbed conditions, from 25 May 1983 to 4 Jan 1984. The goal was to determine if qualitative and/or quantitative changes in bacteria found in various seedling tissues were related to seedling age, stage of annual development or cultural treatment(including nursery transplanting and storage over winter). A total of 114 bacterial colony "types" were isolated; to date, 24 of these have been identified using the API Numerical Profile Index for Enterobacteriaceae. Those identified belonged to the genera Pseudomonas, Pasturella, Acinetobacter, Enterobacter, Flavobacterium, Serratia and Moraxella. Preliminary analysis of the bacteria counts per gram of seedling tissue (leader, stem, primary root and fine roots) indicate fluctuations in both bacterial types and numbers. Such information may eventually help to improve nursery storage conditions and provide another means to monitor seedling health prior to outplanting.

A640

INVESTIGATIONS ON THE DECAY RESISTANCE OF ELM WETWOOD. J.S. Coleman, C.W. Murdoch, R.J. Campana, and W.H. Smith. University of Maine, Orono, ME 04469 and Yale University, New Haven, CT 06511.

Studies have shown that elm wetwood is resistant to decay, but the mechanisms of resistance have not been demonstrated. A total of 120 sapwood and wetwood blocks were either surface sterilized, autoclaved or given no treatment, inoculated with Gleophyllum trabea or Coriolus versicolor, incubated for 90 days at 22 C and weight loss determined. The fungi were grown on potato dextrose

and malt extract agar amended to pH values of 4-8.5, and on malt extract agar amended with filter sterilized (0.45u) or autoclaved wetwood capillary liquid in ratios of: 1:4; 1:10; and 1:25 (v/v). Radial growth of the fungi was measured after 4 days. Wetwood decayed significantly less (P = 0.05) than sapwood. Wetwood fluid showed significant inhibition (P=0.05) of fungal growth both heated or not, while extreme pH values inhibited (P=0.05) fungal growth. Wetwood fluid appears to contain chemical components inhibiting growth of wood decay fungi.

A641

FUNGAL COLONIZATION OF MORIBUND AMERICAN ELM TISSUES. J.G. O'Brien and R.A. Blanchette. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul 55108.

More than sixty distinct fungal taxa were identified from the phloem and outer bark of a single American elm tree sampled during a five week period. The 687 isolates recovered comprised 48.8% from unsterilized outer bark, 27.5% from unexposed inner phloem, and 23.7% from the cambial zone. The proportion of isolates recovered from the innermost zone increased from 14.3% of the total in the first week of sampling to 27.6% in the fifth week. The most frequently recovered taxa were, in descending order: Trichoderma spp., Mucor microsporus (group), Rhizopus stolonifer, Fusarium solani, Rhizoctonia solani, Alternaria alternata, Gliocladium roseum, Phomopsis sp., Epicoccum purpurascens and Diplodia mutila. Fungi obtained are being evaluated as potential antagonists of Ceratocystis ulmi and elm bark beetles.

A642

VARIATION IN SPORE WALL STRUCTURE AMONG ISOLATES OF Sphaeropsis sapinea. Wang Cheng-guo, R.A. Blanchette, Department of Plant Pathology, University of Minnesota and M.A. Palmer, North Central Forest Expt. Station, St. Paul, MN 55108.

Spores from six isolates of Sphaeropsis sapinea (Fr.) Dyko & Sutton (Diplodia pinea) with two different cultural characteristics, fluffy and flat mycelial growth, were examined using scanning and transmission electron microscopy. Spores from cultures with fluffy colony growth had smooth surfaces. No ornamentation was present on or in the cell wall. The spore wall was characterized by an outer electron-dense layer of melanin deposits and an inner transparent layer. Mature spores from isolates with flat colony growth had a regularly distributed pattern of pits over the spore surface. Melanin deposits were also located in the outer layer of the cell wall. All spores contained numerous small lipid bodies in the cytoplasm. These cultural and morphological differences suggest that there are two distinct strains of S. sapinea.

A643

RELATIVE SUSCEPTIBILITIES OF FIVE SOUTHEASTERN PINE SPECIES TO THE PINEWOOD NEMATODE, BURSAPHELENCHUS XYLOPHILUS. L.D. Dwinell. USDA For. Serv., Southeast. For. Expt. Sta., Athens, GA. 30602.

Three-yr-old seedlings of eastern white, loblolly, pond, slash, and Virginia pines were inoculated in a greenhouse with 3 populations of the pinewood nematode (PWN) isolated from declining Virginia pines in Alabama, Georgia and South Carolina. Some 63 seedlings per pine species were inoculated with each PWN population. The bark was removed from a section of the 2-yr-old stem and a moistened plug of cotton with 4,000 nematodes from populations increased on Botrytis cinerea was attached with Parafilm. The data for the populations were pooled. After 12 wk, pine mortality and rankings were: slash (80%), highly susceptible; eastern white (44%), loblolly (47%), and pond (40%), moderately susceptible; and Virginia (2%), highly resistant. In the greenhouse, the rankings were probably due to the affect of PWN alone, but in the field interaction of PWN with insect vectors and the influence of stress factors could result in a change in the species' ranking.

A644

OZONE (O_3), SULFUR DIOXIDE (SO_2), AND ACIDIC RAIN EFFECTS ON GROWTH OF WHITE AND GREEN ASH SEEDLINGS. A.H. Chappelka, B.I. Chevone and T.E. Burk, Dept. Plant Path., Physiol. & Weed Sci., & Dept. For., VPI&SU, Blacksburg, VA 24061.

Nine week old white and green ash were exposed to combinations of O_3 and/or SO_2 (0.00 ppm O_3 and SO_2, 0.10 ppm O_3, 0.08 ppm SO_2 or 0.10 ppm O_3+0.08 ppm SO_2, 4h/d, 5d/wk) and simulated rain (pH 3.0, 4.3 or 5.6 1h/d, 2d/wk at 0.75 cm/h) for 6 wks. Green ash had greater shoot growth and biomass than white ash after 6 wks, regardless of treatment. No significant differences in height growth and total biomass occurred among pH treatments for green ash, but height growth was significantly reduced by O_3+SO_2 compared to controls. On white ash, compared to controls, leaf dry weight was significantly less with O_3 treatment, and root/shoot ratio (RSR) was significantly less at pH 3.0 compared with pH's 4.3 and 5.6. A significant pollutant X rain interaction for RSR also occurred with this species. At pH 3.0 vs. pH 5.6, a 4, 9, 20 and 25% decrease in RSR occurred for non-fumigated, O_3+SO_2, O_3 and SO_2, fumigated seedlings.

A645

SURFACE CHANGES TO PINE NEEDLES INDUCED BY AMBIENT PARTICLES Charles R. Krause, USDA,ARS and Leon S. Dochinger, USDA, USFS 359 Main Rd., Delaware, OH 43015

Field studies were conducted to determine if changes could be detected on needle surfaces of white pines grown in clean air or in air containing high levels of particles. Ten Pinus strobus ramets, each previously screened to be tolerant to SO_2 and O_3, were grown either in Delaware, OH (clean air) or in Cleveland, OH (polluted air) for one year. Trees exposed to polluted air had chlorotic symptoms while those exposed to clean air were uninjured. Needles of trees at each site were sampled monthly during 1983 and analyzed in a scanning electron microscope (SEM) equipped with an energy dispersive X-ray analyzer (EDX) and a cold stage. SEM observations of needles grown in clean air revealed downy, loosely arranged, tubular epicuticular wax crystals. Particles containing Ca or Si were detected with EDX. Needle surfaces exposed to polluted air had agglomerated wax adjacent to particles containing, S, Fe and trace elements.

A646

AMINO ACID ALTERATIONS IN SUSCEPTIBLE, TOLERANT, AND RESISTANT SOUTHERN PINES INOCULATED WITH THE FUSIFORM RUST FUNGUS. Vernon Ammon and Dallas Seifers.

Amino acids were extracted from 3- and 6-mo old tissues removed from untreated and inoculated southern pine trees susceptible, tolerant, and resistant to the fusiform rust fungus, Cronartium quercuum f. sp. fusiforme. Untreated susceptible tissues had the highest content of free amino acids (AA) at both sampling dates. All three host reaction types had prominent serine: arginine levels in both treated and untreated tissue. Inoculated 3- and 6-mo-old tissue had increased levels of serine, arginine, glutamic acid, alanine, tryptophan, and tyrosine. Amino acids extracted from inoculated 3-mo-old tissue were highest in resistant tissue and lowest in the susceptible selection. At 6-mo, the inoculated susceptible tissues contained the highest levels of AA. Similar levels were measured from tolerant tissues and lower concentrations from resistant tissues.

A647

TEM OBSERVATIONS ON DECOMPOSITION OF WESTERN HEMLOCK AND SWEETGUM BY THE BROWN-ROT FUNGUS PORIA PLACENTA. T. L. Highley, L. Murmanis, and J. G. Palmer, Forest Products Laboratory, Madison, WI 53705

Secondary cell walls of hemlock and sweetgum, degraded by the brown-rot fungus, Poria placenta, and observed by TEM show a similar mode of attack in both species. Attack is predominantly initiated by hyphae growing in the cell lumen rather than by penetration. The S_2-layer is extensively degraded while the S_3-layer remains relatively undegraded when attack is intiated from hyphae in the lumen. Degradation by penetrating hyphae differs from that of non-penetrating hyphae in that degradation is localized to the immediate vicinity of the hyphae. On occasion intense degradation in localized areas of the middle lamella and cell corners occurs without noticeable degradation of the surrounding secondary wall. Evidence that P. placenta produces lignin-degrading agents is demonstrated by the destruction of the lignin lamellar structure visualized by $KMnO_4$ fixation.

A648

RESISTANCE OF PINUS VIRGINIANA AND P. RIGIDA X TAEDA SEEDLINGS TO ENDOCRONARTIUM HARKNESSII. N. Wenner, W. Merrill, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802, and B. Towers, Pa. Dept. Environ. Res., Middletown, PA 17050.

The susceptibility of 2 year-old Pinus virginiana and P. rigida x taeda seedlings to natural infection by Endocronartium harknessii was evaluated in an isolated P. ponderosa planting in central PA. Pinus ponderosa trees at this site were severely infected with E. harknessii and bore numerous galls. One hundred potted seedlings of each species were positioned beneath the P. ponderosa trees from May to July 1982 (budbreak to shoot maturation). All of the seedlings were within 1 m of sporulating galls. Galls formed on

1982 shoots of P. ponderosa, but not on the P. virginiana or P. rigida x taeda seedlings. P. virginiana is known to be susceptible to E. harknessii when artificially inoculated. However, these results indicate that P. virginiana was not susceptible to natural infection in a forest stand and probably would not serve as a bridge between the present range of E. harknessii and the susceptible southern pines.

A649

BIODETERIORATION OF OAK TREES DEAD FOLLOWING GYPSY MOTH DEFOLIATION. D. Karasevicz, W. Merrill, and B. Towers, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802, and Pa. Dept. Environ. Res., Middletown, PA 17507

In a preliminary study of the biodeterioration of Quercus rubra and Q. velutina killed following defoliation by Lymantria dispar, 210 trees in central Pennsylvania that died in 1978 were surveyed in the fall-winter of 1983-84. Of 135 trees on a north-facing slope, 63% bore Polyporus gilvus, 56% P. pargamenus, and 48% Armillaria mellea. Smaller percentages of trees bore fruiting bodies of nine other species of decay fungi. In contrast, on a south-facing slope, 43% bore P. gilvus, 32% P. pargamenus, 40% A. mellea, and 29% Hypoxylon atropunctatum. Smaller percentages of trees bore seven other species of decay fungi. All fungi were involved in sapwood decay. On most trees deterioration had advanced several cm into the heartwood. Isolations from deteriorating heartwood, however, yielded fungi different from those fruiting on decayed sapwood mentioned above. This indicates a succession of organisms in the biodeterioration of dead standing oaks.

A650

COMBINED EFFECTS OF IRRIGATION AND PRECIPITATION ON BROWN STEM ROT DEVELOPMENT AND YIELD OF TWO SOYBEAN CULTIVARS. A. Mengistu, and C. R. Grau. Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Two soybean cultivars, BSR 201 (moderately resistant to brown stem rot (BSR)) and Corsoy 79 (susceptible), were grown under 4 irrigation regimes (irrigated at VC-R1, VC-R7, and R1-R7 growth stages and unirrigated). Plots were established in two areas: 1) naturally infested with the BSR pathogen, Phialophora gregata, using all 4 irrigation regimes and 2) uninfested (irrigated only at VC-R7). Assessment of internal stem discoloration, 6 times at 3 wk intervals during V1, V4, R2, R4, R5, and R7 growth stages, indicated that the rate of disease progress was significantly lower (5% level) on BSR 201 than on Corsoy 79 at all irrigation levels. Disease progress on both cultivars in VC-R1 irrigated and unirrigated plots was significantly lower than in plots irrigated in the R1-R7 or VC-R7 stages. Yield and seed weight reductions were also significantly higher under R1-R7 and VC-R7 irrigations. Disease severity at the R4 and R5 stages showed a very high and significant R^2 value with both yield loss and seed weight reductions.

A651

DISTRIBUTION OF LETTUCE ANTHRACNOSE IN THE FIELD. B. R. Delp, L. J. Stowell, and R. G. Grogan. Department of Plant Pathology, University of California, Davis, CA 95616

Lettuce fields were assessed for anthracnose caused by Marssonina panattoniana. A recently developed computer program titled "Field Runner" and a portable microcomputer were used to divide fields into sectors of equal area. A randomly-located transect of 30 adjacent plants within each sector was evaluated for anthracnose. Data were analyzed to determine the variance to mean ratio ($s^2/\bar{x}$) based on the number of diseased plants in each transect and the variation among transects. Consistant $s^2/\bar{x}$ ratios >1 indicated that disease was aggregated in the field. Ordinary runs analysis of data from transects with 20-80% disease indicated that disease was aggregated within the transects (Z-score of the runs analysis were consistantly <-1.64). We conclude that anthracnose spreads from plant to plant and that disease is aggregated in the field.

A652

DISEASE DISTRIBUTION AND CROP LOSS ASSESSMENT USING A FIELD-PORTABLE MICROCOMPUTER. L. J. Stowell, B. R. Delp, and R. G. Grogan, Department of Plant Pathology, University of California, Davis, CA 95616

We have developed a program for use with a portable computer that will facilitate assessment of disease distribution and crop loss. Conventional methods of assessing fields for disease loss have utilized simple paths for sampling (e.g. diagonal, "X" or "W"). More complex patterns of sampling have not been utilized due to the difficulty and time required to locate the sites for sampling within the field. With the aid of an Epson HX-20 microcomputer and a program titled "Field Runner", stratified random sampling is now feasible. The computer directs the operator to the locations in the field for sampling. In addition to assistance in sampling, this system analyzes the data and computes disease loss, prevalence, ordinary runs analysis and variance to mean ratios based on raw and transformed data. These analyses are important in determining the accuracy of the loss assessment and also the epidemiology of the disease.

A653

FIELD SAMPLING OF FUNGAL PATHOGEN POPULATIONS: A SIMPLE DEVICE FOR MONITORING FUNGICIDE RESISTANCE. N. Lalancette, Jr., J. M. Russo, and K. D. Hickey, The Pennsylvania State University Fruit Research Lab, Biglerville, PA 17307-0309.

A simple device was developed for field sampling of fungal spores from lesions. The sampler consists of a standard sized lipbalm tube containing a solidified water agar core. Spores are removed by rubbing the agar surface of the core against a lesion. After slicing off the top of the agar core, the agar discs are incubated in petri dishes and then observed microscopically for spore germination. Incorporation of a fungicide into the agar allows for determination of resistance. The device was used successfully for field sampling in epidemiological experimentation as well as for monitoring fungal populations in commercial settings. Other potential uses for the device involve determining of spore production of individual lesions sampled in the field and monitoring fungicide residues on plant material.

A654

THE INFLUENCE OF SOIL WATER STATUS ON THE EPIDEMIOLOGY OF TOBACCO BLACK SHANK. D.M. Ferrin and D.J. Mitchell, Dept. of Plant Pathology, Univ. of Florida, Gainesville, FL 32611.

The influence of soil water status (measured by the daily summation of vacuum gauge tensiometer readings in centibars at the 15-cm soil depth) on disease development in two tobacco cultivars by Phytophthora parasitica var. nicotianae was assessed in 1982 and 1983. Increase in disease proportion (y) was positively correlated with time (in centibar-days). The average rates of change of disease proportion ($\Delta y/\Delta t$) and soil water status between successive disease ratings were plotted against time and compared. Increases in $\Delta y/\Delta t$ were associated with periods of wetting following dry soil conditions and initial periods of drying following prolonged periods of wetting. Cyclic increases in $\Delta y/\Delta t$ coincided in time for the two cultivars but their magnitudes were greater for Hicks than for Speight G-28. The maximum $\Delta y/\Delta t$ coincided with the first wet period following a drought period early in the season for Hicks but was delayed until flowering for Speight G-28.

A655

PHYTOSCAN 83: A COMPUTER PROGRAM FOR QUANTITATIVE DISEASE ASSESSMENT. C. R. Bronson and W. M. Klittich, Department of Plant Pathology, Seed and Weed Sciences and Department of Agronomy, respectively, Iowa State University, Ames, IA 50011

Microcomputer controlled video image analysis has received increasing attention as a method of quantitative disease assessment. Such systems can be very sensitive to slight differences in light intensity, and improper system calibration can often result in erroneous measurements. Phytoscan 83 is a program written for the Apple II Plus microcomputer that, in addition to scanning the field of view and calculating disease severity, displays the digitized image to permit rapid verification and adjustment of system parameters. The program is flexible and may be easily adapted to analyze any field of view that can be meaningfully divided into two or three categories on the basis of light intensity.

A656

VALIDATION OF REGIONAL MODELS FOR PREDICTING STRIPE RUST ON WINTER WHEAT. S.M. Coakley and R.F. Line, NCAR, P.O. Box 3000 Boulder, CO 80307 and ARS-USDA, WSU, Pullman, WA 99164.

Statistical models for predicting stripe rust(caused by Puccinia striiformis) on winter wheat cultivars Gaines, Nugaines and Omar at Lind, Pullman, and Walla Walla, WA and Pendleton, OR were developed based on 1968 to 1981 data. The dependent variable was disease intensity index (DI). Model I used standardized negative degree days (NDDZ), and Julian day of spring (JDS) as independent variables. Model II used positive degree days (PDD) in addition to NDDZ and JDS. In 1983, Model I was used in early February and Model II in late June to predict DI on the three cultivars at the four locations and at Mt. Vernon. Actual DI was

within the standard error of prediction for Model I 12 out of 15 times and for Model II all 15 times. Although spring temperatures were important in reducing DI, the early predictions allowed growers the option of applying chemical control on susceptible cultivars; the differences in predictions from the two models did not negate the value of any disease control applied.

A657

THE CRITICAL PERIOD FOR CONTROL OF POTATO EARLY BLIGHT, CAUSED BY Alternaria solani. J.W. Pscheidt and W.R. Stevenson, Dept. of Plant Pathology, Univ. of Wis., Madison, WI 53706.

The critical period (CP), during which a protectant fungicide program must begin in order to control early blight effectively, was examined during the 1980-82 growing seasons. The CP was identified by spraying Russet Burbank potatoes with chlorothalonil (0.88 kg/ha) starting 0, 2, 4 and 6 weeks after plants were 20-25 cm tall and continuing weekly until vinekill. Leaf wetness, temperature, relative humidity, rainfall and airborne spores of A. solani were monitored throughout the growing season. The CP began 3, 4 and 6 weeks after plants were 20-25 cm. tall during 1982, 1980 and 1981, respectively. The CP was characterized each year by high concentrations of airborne spores and weather favorable for sporulation and infection. Similar weather and spore conditions occurred prior to the CP each year, which indicates that the timing of this period may also be related to host susceptibility. Since susceptibility to early blight increases as the potato plant matures, studies on host development were conducted in 1983. These data indicate that the CP may be the time when foliage production, as measured by fresh weight and an index of leaf area, reaches its maximum.

A658

REACTION OF SOYBEAN CULTIVARS TO STEM CANKER IN LOUISIANA. J. P. Snow, G. T. Berggren and E. C. McGawley. Dept. of Plant Path. and Crop Physiol.; H. K. Whitam, La. Coop. Ext. Serv.; and B. G. Harville, Dept. of Agronomy. La. State Univ. Agr. Ctr., Baton Rouge, LA 70803.

A severe outbreak of soybean stem canker, caused by Diaporthe phaseolorum (Cke and Ell) Sacc. var. caulivora Athow and Caldwell, occurred in south Louisiana in 1983. A total of two hundred four breeding lines and cultivars in maturity Groups V, VI, VII and VIII planted in replicated plots at the Burden Research Plantation at Baton Rouge were rated for stem canker resistance. The rating scale was 0 to 9 with 0 indicating no symptoms and 9 indicating all plants in a plot were dead. Ratings between 0 and 9 were based on numbers of plants showing symptoms (cankers on stems and leaves with characteristic interveinal browning), length of stem lesions and number of affected leaves per plant. In most cases, high stem canker ratings were correlated with low yields and low ratings were correlated with high yields.

A659

DISTRIBUTION OF SOYBEAN STEM CANKER IN LOUISIANA. G. T. Berggren, J. P. Snow, E. C. McGawley. Dept. of Plant Path. and Crop Physiol. B. G. Harville, Agronomy Dept., and H. K. Whitam, La. Coop. Ext. Serv. La. State Univ. Agric. Ctr., Baton Rouge, La. 70803.

Stem canker, caused by Diaporthe phaseolorum (Cke and Ell) Sacc. var. caulivora Athow and Caldwell, has been reported as a serious disease of soybean in the U.S. and Canada. The disease spread in recent years from the traditional soybean producing states of the upper midwest to the lower Mississippi valley and the southeastern U.S. The disease was first observed in Louisiana in 1981 at one location in the central part of the state. In 1982 stem canker was detected in three parishes with severe yield losses in isolated fields. A widespread epidemic of stem canker was noted during the 1983 growing season, with the disease observed in 24 soybean producing parishes. Losses were of economic importance in 18 of the parishes. A higher incidence of the disease was apparent in the southern half of Louisiana, where rainfall was generally above average.

A660

STRATEGIES FOR CONTROLLING SOYBEAN AERIAL BLIGHT IN LOUISIANA. Berggren, G. T., E. C. McGawley, M. E. Pace, J. S. Gershey and G. F. Joye. Dept. Plant Path. and Crop Physiol., La. Agric. Expt. Sta., La. State Univ. Agric. Ctr., Baton Rouge, LA 70803

Aerial blight of soybeans, caused by the fungus Rhizoctonia solani Kuhn was described by Atkins and Lewis in 1954. The recent dramatic increase in soybean acreage in Louisiana and the practice of rotation with rice in south Louisiana has increased the problem in both crops. The organism causes sheath blight, a major disease of rice, and has been reported on a wide range of common monocotyledonous and dicotyledonous weed hosts. Control practices for aerial blight have been designed using a system of varietal selection, fungicidal sprays and cultural practices. Several soybean cultivars were moderately tolerant to the fungus. In addition, foliar sprays of Benlate 50 WP (1.12 kg/ha applied twice) were effective in supressing the disease. Although yields were significantly higher in narrow row (25.4 cm) plantings, disease severity was not affected by row spacing.

A661

VIDEO IMAGE ANALYSIS OF LODGING AND YIELD LOSS IN WINTER WHEAT RELATIVE TO FOOT ROT. D. M. Gerten and M. V. Wiese, Dept. of Plant, Soil and Entomological Sciences, University of Idaho, Moscow, ID 83843.

Microcomputer-assisted video image analysis can be used to identify and quantify image areas differing in light intensity. This technique was used to measure the extent of lodging in winter wheat fields caused by foot rot (Pseudocercosporella herpotrichoides). An Apple IIe microcomputer equipped with a digitizer and interfaced to an RCA video camera reproduced images from 65,536 pixels categorized according to 64 grey values. Analysis of color and color infrared aerial photographs of 7 maturing wheat fields revealed that lodging encompassed from 2 to 32% of the total area of each field. Concurrent manual measurements within each field showed significant differences in yield and foot rot levels between erect and lodged areas. In erect areas, yields were 1.4 to 3.4 t/ha (21 to 51 bu/a) higher and disease levels (percent of tillers with severe lesions) 9% lower than in lodged areas.

A662

APPLICATIONS OF COMPETITION MODELS TO STUDIES OF YIELD IN DISEASED AND DISEASE-FREE CULTIVAR MIXTURES. Helen Miller Alexander, Dept. of Biology, Univ. of Louisville, Louisville, KY 40292; J. J. Burdon, Div. of Plant Industry, CSIRO, P.O. Box 1600, Canberra City, ACT 2601, Australia; and A. P. Roelfs, Cereal Rust Laboratory, Univ. of Minnesota, St. Paul, MN 55108

The use of mixtures of cultivars for disease control has become a familiar concept. However, little is known about the basic relationship between disease levels and yield in such mixtures. The yield of a mixture depends on the yielding ability of the component cultivars, interaction (such as competition) between components, and the effect of the mixture of components on disease levels. Competition models developed for disease-free situations can be used to show the relative importance of these factors in plots of constant density but differing proportions of the components. "Ratio diagrams" show the ratio of components sown to components harvested while "replacement series yield diagrams" depict component and total plot yields. Using these concepts, we have developed diagrammatic models that illustrate how mixtures of different composition may yield in the presence of disease.

A663

MULTISPECTRAL RADIOMETRY USING A 12-BIT ANALOG-TO-DIGITAL CONVERTER INTERFACED WITH A PORTABLE MICROCOMPUTER. Vernyl D. Pederson, North Dakota State University, Fargo, ND 58105

Reflection of solar radiation from barley crop canopies was measured in 8 discrete wavelength bands ranging from 500 to 850 nm using a 12-bit analog-to-digital converter interfaced with a Radio Shack TRS-80 model 100 portable computer. The outputs of the radiometer, which ranged from 0-5 volts, were input to a 12-bit successive approximation A/D converter and transmitted by a UART to the serial I/O port of the portable computer. The UART was set to operate at 19,200 BAUD with 8 bits, even parity, and 1 stop bit. The analog to digital converter required 13 clock periods to complete a conversion. Conversion time was 170 microseconds with clock frequency of 76,800 Hz. The time required for one channel was 1.78 milliseconds. Power for the system was sourced by 2 gel-cell lead acid batteries connected in series. Software for the Model 100 enabled selection of up to 29 channels, storage of digitized voltage values in RAM, and calculation of percentage reflection from crop canopies for each wavelength.

A664

YIELD LOSS MODELING:TOLERANCE TO PATHOGENS. F. W. Nutter, Jr. and V. D. Pederson. Dept. of Plant Pathology, North Dakota State University, Fargo, North Dakota 58105.

A general yield loss equation cannot be used if relative differences in tolerance to disease exists among cultivars. Tolerance can be measured by the regression coefficient relating yield loss to disease severity. However, comparisons of regression coefficients are valid only for the range in which

disease severities of cultivars overlap, otherwise, improper conclusions concerning the presence or absence of tolerance may be made. For example, when spores of Cochliobolus sativus (spot blotch) were applied at one time only to barley plants, the resulting disease severities of Larker (susceptible) and Dickson (resistant) did not overlap. However, using multiple inoculations at specific barley growth stages, considerable overlap of disease severities resulted. When only the range of overlap of Larker and Dickson disease severities were regressed against yield, regression coefficients for Larker and Dickson were equivalent, i.e. one yield loss equation can be used for both cultivars.

A665

INTERRELATIONSHIPS OF SEPTORIA GLYCINES, XANTHOMONAS CAMPESTRIS PV. GLYCINES AND HETERODERA GLYCINES ON SOYBEANS IN ILLINOIS. J. A. Appel, G.R. Noel, D.I. Edwards, and S.M. Lim. Dept. of Plant Pathology and USDA-ARS, University of Illinois, Urbana 61801.

Experiments were established on H. glycines infested land for which Pi's ranged between 2-160 and 30-540 gravid cysts/250 cm^3 of soil, respectively in central and southern Illinois. Foliar disease was evaluated throughout three growing seasons and nematode populations were sampled at planting and six weeks following planting. Foliage at R6 was 60-70% and 10-25% infected, respectively for treatments inoculated with S. glycines and X. c. pv. glycines. Yield losses associated with H. glycines were largest and foliage disease severity was lowest when soybeans grew under prolonged environmental stress. Depending on the year, yield losses at the central location were 10-15% for S. glycines and 6-15% for H. glycines. Depending on the year, yield losses at the southern site were greater than 50% due to H. glycines and 6-11% for S. glycines. The yield losses from H. glycines and S. glycines were additive while no significant yield loss was associated with X. c. pv. glycines.

A666

THE EXTRACTION, ASSAY AND GENERAL PROPERTIES OF AN INDUCING RESISTANCE FACTOR FROM CUCUMBER LEAVES INDUCED WITH PSEUDOMONAS LACHRYMANS. N. A. Garas and J. Kuć, Department of Plant Pathology, University of Kentucky, Lexington, KY 40546, U.S.A.

An inducing resistance factor (IRF) was extracted from cucumber leaves previously induced with Pseudomonas lachrymans. Healthy susceptible cucumber plants supplied with the active factor exhibited induced systemic resistance to Colletotrichum lagenarium similar to that elicited by localized infection with fungi, bacteria or viruses. The induced resistance activity, expressed as a decrease in the number of lesions developed on leaf 2 of IRF-treated plants, was highly significant when the extracts were made from leaves 3 or more days after induction with P. lachrymans. The protection was evident if the treated plants were challenged after two days and progressively increased to reach highest level of protection in plants challenged 5 or 6 days after the treatment with IRF. The IRF, in crude extracts, is nondialyzable, heat unstable and tending to lose activity if freeze dried or subjected to extreme pH's.

A667

CHARACTERIZATION OF ROUGH LEMON-SPECIFIC TOXINS FROM ALTERNARIA CITRI AND THEIR SITE OF ACTION. J. M. Gardner, University of Florida, Citrus Research and Education Center, Lake Alfred, FL 33850 and Y. Kono, Lab. of Phytochem., R.I.K.E.N., Saitama 351, Japan.

The A. citri rough lemon-specific (ACRL) toxins exist as multiple isomers. The structure of a crystalline inactive conversion product of toxins has been determined and from this we have deduced a probable general structure for toxin. The conversion product ($C_{18}H_{30}O_4$) has a substituted tetrahydropyran ring; side chains contain hydroxyl, carbonyl and allylic groups. The ACRL toxins appear to be isomeric keto alcohols which inactivate with formation of the pyran ring. The conversion product is not competitive with toxin isomers, which have equivalent toxicities (approx. end point 5 ng/ml). Mitochondria are markedly affected within 1 hr and in vitro effects on mitochondria have been observed. The effects on mitochondria are interesting in view of some possible structural analogies between H. maydis T. toxin and ACRL toxin.

A668

FLUORESCENCE-ACTIVATED CELL SORTING FOR LOCALIZATION OF PHYTOALEXINS. M. Pierce and M. Essenberg, Oklahoma State University, Stillwater, OK 74078; A. Birdsong and V. E. Scholes, Oral Roberts University, Tulsa, OK 74171.

During the resistant response of cotton line OK 1.2 to Xanthomonas campestris pv. malvacearum, leaf cells next to a bacterial colony turn brown and become yellow-green fluorescent. The resistant leaf produces the two phytoalexins 2,7-dihydroxycadalene (DHC) and lacinilene C (LC). LC is yellow-green fluorescent. Are the phytoalexins predominantly located in the fluorescent cells? If so, we calculate their concentrations to be high enough to account for the observed inhibition of bacterial growth. We are isolating mesophyll cells from infected leafy cotyledons. A fluorescence-activated cell sorter is used to deflect fluorescent cells and non-fluorescent cells into different tubes. Cells are isolated and sorted as quickly as possible (1-1.5 hr), since LC and DHC diffuse into aqueous medium. The first chemical analysis of sorted cells indicated that the fluorescent fraction contained 5 times more LC and DHC per cell than did the non-fluorescent fraction.

A669

LOCATIONS OF CORYNEBACTERIUM MICHIGANENSE pv INSIDIOSUM EXTRACELLULAR POLYSACCHARIDE ACCUMULATION WITHIN THE TRANSPIRATION STREAM OF ALFALFA. B. D. McMillan and N. K. Van Alfen, Department of Biology, Utah State University, Logan, UT 84322.

The extracellular polysaccharide (EPS) of Corynebacterium michiganense pv insidiosum consists of three different sized components, each postulated to act at separate locations within the plant to induce water stress. To determine the points of accumulation, each component was purified, radiolabeled with ^{14}C-lysine, and then introduced into alfalfa cuttings by transpiration. Autoradiograms of the cuttings revealed the smallest molecule (m.w. 25 x 10^3 daltons) accumulated primarily in the leaves and meristematic tissue. The second component (m.w. 5 x 10^6 daltons) could not pass beyond the first internodes. The largest component (m.w. >5 x 10^6 daltons), similarly, would not pass beyond the first internode. These data indicate that the location at which each component acts is a function of the size of the molecule and the diameter of the capillaries within the plant's transpiration stream.

A670

COMPARISON OF XYLEM BLOCKAGE STRUCTURES IN VARIOUS CITRUS TREE DECLINES. R. H. Brlansky, M. H. Collins, and R. F. Lee. Univ. of Florida, Citrus Research and Education Center, Lake Alfred, FL 33850.

Citrus tree declines such as tristeza, foot rot, psorosis, and citrus slump often mimic the symptoms of citrus blight. Xylem vessels in trunks of citrus trees with these decline or dieback diseases were examined for the occurrence of any blockage structures using light microscopy (LM), scanning (SEM), and transmission electron microscopy (TEM). Amorphous and filamentous plugs have been described previously in the xylem vessels of trees affected with citrus blight. Filamentous plugs were frequently present in the xylem vessels of healthy trees and in trees affected by other citrus declines. Using LM, occlusions similar to the amorphous plugs associated with citrus blight were seen in trunk xylem of trees affected by psorosis and concave gum. These amorphous-like plugs could be differentiated from those associated with blight using SEM and TEM.

A671

EFFECTS OF IONONE-TYPE COMPOUNDS ON GROWTH OF TOBACCO AND RESISTANCE TO BLUE MOLD. S. D. Salt and J. Kuć. Dept. Plant Pathology, University of Kentucky, Lexington, KY, U.S.A. 40546

Ionone-type compounds arise in vivo in tobacco via oxidation of carotenoid pigments. β-ionone and 3-isobutyroxy-β-ionone (quiesone) are extremely potent (ED_{50} 0.15 ppm and 0.0001 ppm, respectively) inhibitors of Peronospora tabacina spore germination. Administration of ionones and structurally related abscisic acid into stems of tobacco cv. Kentucky 14 resulted in marked changes in plant growth, morphology, and resistance to P. tabacina. Ionone-induced changes include accelerated growth and senescence, loss of apical dominance, and reduction in fungal sporulation. These changes resemble those elicited by immunization of tobacco via stem injection of spores of virulent P. tabacina strains (Y. Cohen and J. Kuć. 1981. Phytopathol. 71:783-787). However, unlike fungal immunization, ionone treatment apparently did not significantly reduce lesion areas.

A672

SYNTHESIS OF (+)-PISATIN BY A METHYLTRANSFERASE FROM PEA. J. A. Sweigard, D. E. Matthews, and H. D. VanEtten, Dept. of Plant Pathology, Cornell Univ., Ithaca, NY 14853-0331.

Precursor feeding studies suggest that the terminal step of (+)-pisatin biosynthesis in Pisum sativum is the O-methylation

of (+)-6a-hydroxymaackiain (HMK). We have found that extracts from $CuCl_2$-treated pea seedlings exhibit a methyltransferase activity dependent on S-adenosylmethionine and (+)-HMK. The enzyme activity was also detected in extracts from diseased epicotyls and, at lower levels, from healthy tissues. Methyl transfer was assayed as toluene-extractable radioactivity produced from [methyl-^{14}C]-S-adenosylmethionine in the presence of enzyme and a phenolic substrate. The low level of activity observed in the absence of substrate, presumably due to the presence of methyl acceptors in the enzyme extract, was markedly stimulated by addition of (+)-HMK. It has been reported that $CuCl_2$-treated pea tissue provided with (-)-HMK or (-)-maackiain can synthesize (-)-pisatin. Our extract showed no methyltransferase activity dependent on either of these substrates. (+)-Maackiain was utilized ca. 5% as well as (+)-HMK.

A673

EFFECTS OF N-ACETYL- AND AMINE-SUGAR DERIVATIVES ON INFECTION OF BARLEY EPIDERMIS BY ERYSIPHE GRAMINIS F. SP. HORDEI. W. R. Bushnell and C. Curran. USDA-ARS, Cereal Rust Laboratory, Dept. of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Twenty-seven sugars and sugar derivatives, including several lectin haptens, were applied at 0.1 M with 0.01 M $Ca(NO_3)_2$ to epidermal tissues from coleoptiles of barley (Hordeum vulgare) at the time of inoculation with Erysiphe graminis f. sp. hordei. N-acetylglucosamine, NN'-diacetylchitobiose, glucosamine, and galactosamine strongly inhibited the formation of haustoria. Appressoria were normal. N-acetylgalactosamine and methyl-N-acetylglucosamine were among the inactive compounds tested. The inhibition resembled that produced by NH_4^+ and other monovalent cations. Several nonsugars with amino groups also inhibited infection, including lysine, valine, and putrescine. Inhibition by 0.01-0.06 M N-acetylglucosamine or 0.001-0.01 M NH_4^+ was partially reversed by Ca^{2+} at 0.01 M. The results suggest that plus-charged amino moieties of effective compounds, or NH_4^+ released from them, inhibited development of competent infection pegs by the fungus.

A674

LIGNIFICATION OF LESION BORDERS IN BEAN STEM CANKER. V. Stockwell and P. Hanchey. Dept. Botany and Plant Pathology, Colorado State University, Fort Collins, CO. 80523

Previous studies failed to confirm the hypothesis that cell wall calcification is associated with lesion delimitation in Rhizoctonia solani-infected bean hypocotyls. At the lesion periphery, cell walls are autofluorescent and react with toluidine blue, azure B, phloroglucinol, ferric chloride, and the nitroso and Maule reagents as early as the penetration and water soaking stages. Increases in polyphenol oxidase and peroxidase activities paralleled lignification at lesion borders. Lesion border walls were not macerated when tissues were digested in a mixture of 5% macerase and 5% cellulysin, but digestion was complete if lignins were first extracted with sodium hypochlorite and sodium bisulfite. We suggest that the previously demonstrated shift of carbohydrate metabolism to the pentose phosphate pathway may be important in the production of phenolics and lignin precursors. Lignification appears to be important in both lesion delimitation and increased resistance of lesion border walls to maceration by fungal enzymes.

A675

GROWTH DYNAMICS OF PHYMATOTRICHUM OMNIVORUM AND GOSSYPIUM KLOTZIANUM IN A PARABIOTIC SYSTEM. D. H. Kruse and S. D. Lyda, Dept. of Plant Pathology & Microbiology, Texas A&M Univ., College Station, TX 77843

A system was developed to grow Phymatotrichum omnivorum (PO) and Gossypium klotzianum (GK) separately, but parabiotically. The chamber consists of two L-tubes with a central ground glass joint and a coarse ground-glass frit (exclusion limit of 40-60 um). Two compatible growth media have been used to grow both organisms, Murashige-Skoog's basal medium and Gamborg's B-5 medium, both with hormones necessary for growth of GK. The male L-tube was inoculated with a cell suspension of GK (5 ml) and the female L-tube with PO (8 mm hyphal plug), from a stationary liquid culture. The growth system was shaken at 80 rpm and harvests made every 3 days. Growth of GK was detemined by packed cell volume and growth of PO was determined by fresh and dry wt of hyphal mats. Viability of GK cells was determined with Evans's Blue vital stain. PO grew exponentionally for 10-13 days post inoculation in the controls and for 14-15 days in the parabiotic system. GK showed an exponential increase for 10-12 days in the controls, but no significant increase in the parabiotic system.

A676

IDENTIFICATION AND ELICITOR ACTIVITY OF PHOSPHOLIPIDS FROM PHYTOPHTHORA INFESTANS. J. R. Creamer and R. M. Bostock, Dept. of Plant Pathology, University of California, Davis, CA 95616

Phospholipids, extracted from the mycelium of Phytophthora infestans with a mixture of chloroform and methanol (2:1, v/v), were purified by liquid and thin-layer chromatography. The fatty acid composition of each phospholipid was determined by gas-liquid chromatography of their methyl esters. Phosphatidyl choline, phosphatidyl ethanolamine, phosphatidyl inositol and a phosphorus containing, ninhydrin positive unknown were characterized as the major phospholipids. Phospholipid fractions were assayed for elicitation of terpenoid phytoalexins from potato tuber discs. All phospholipids elicited rishitin and lubimin with phosphatidyl choline and the unknown phospholipid showing the highest activity. The phospholipids contained a high percentage of arachidonic and eicosapentaenoic acids, previously shown to possess elicitor activity. Arachidonic acid was the major fatty acid in the unknown phospholipid. A more complete characterization of the latter is currently under investigation.

A677

ASSOCIATION OF ENHANCED PEROXIDASE ACTIVITY WITH INDUCED RESISTANCE OF MUSKMELON AND WATERMELON. J. A. Smith and R. Hammerschmidt, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

Inoculation of one leaf of muskmelon or watermelon with Colletotrichum lagenarium induced a systemic enhancement of peroxidase activity. The enhanced peroxidase activity appears to be due to an increase in a group of acidic, cell wall associated isozymes. These isozymes are readily extracted with distilled water from the intercellular spaces of leaf tissue and are the major protein component of the intercellular extracts. The isozymes induced in muskmelon and watermelon exhibit electrophoretic mobility on high pH native polyacrylamide gels which is very similar to the mobility of the isozymes induced in systemically protected cucumber. Chemical characteristics of the induced peroxidase isozymes from the three species will be described. Possible roles these isozymes play in the development and expression of induced resistance in cucurbits will be discussed.

A678

ROOT SURFACE PEROXIDASE: A DEFENSE ROLE? Anne J. Anderson and Lloyd W. Bennett. Department of Biology, Utah State University, Logan, Utah 84322.

Roots of intact sterile-grown bean seedlings display surface peroxidase activity. Peroxidase was detected on all root surfaces by the hydrogen-peroxide dependent formation of chromogen from chloronaphthol or dianisidine. Other peroxidase functions, oxidation of IAA and NADPH, were catalysed by intact roots. Both oxidases were stimulated by Mn^{2+} and p-coumarate. Oxidation of NADPH involved superoxide anion (O_2^-), and hydrogen peroxide formation. Analysis of root washes demonstrated NADPH oxidase and peroxidase to be associated with higher molecular weight components than IAA oxidase. Root surface and root wash peroxidase displayed optimal activity between pH 7 to 8 whereas both sources of IAA oxidase were more active at acidic pHs. The peroxidase may constitute a defense strategy for the root because the potential products, phenoxy radicals, or hydrogen peroxide and O_2^-, have antimicrobial properties. Microorganisms which colonize the rhizosphere may require mechanisms to negate the consequences of peroxidase activity.

A679

PLANT PROTOPLAST DEATH CAUSED BY FUNGAL EXTRACELLULAR PRODUCTS. Helen M. Griffiths and Anne J. Anderson, Department of Biology, Utah State University, Logan, Utah 84322.

Colletotrichum lindemuthianum, causal agent of bean anthracnose, displays race-cultivar specificity. Spore inoculation of Great Northern bean stems with α race resulted in large spreading lesions whereas β_1 race lesions were small and limited. Extracellular products from culture filtrates of α and β_1 races reduced viability of protoplasts prepared from Great Northern stem epidermal tissue. Viability was determined by screening for protoplasts that were fluorescent when incubated with fluorescein diacetate. Fractions separated by size on Sepharose 6B from the β_1 race were more active than equivalent-sized materials from the α race. Protoplast suspensions (10^5 cells/ml) displayed 50% reduction in viability within 30 minutes after application of β_1 race products equivalent to less than 0.01 µg glucose per ml. Reduction in protoplast viability was time dependent and varied with concentration of the fungal product. Heat treatment of the extracellular components did not decrease their effectiveness.

A680

ELICITOR PRODUCTION IN TEMPERATURE-INDUCED COMPATIBILITY OF SOYBEAN AND PHYTOPHTHORA MEGASPERMA F.SP. GLYCINEA. D.Classen and E.W.B.Ward, Department of Plant Sciences, University of Western Ontario and Agriculture Canada, Research Centre, London, Ontario. N6A 5B7

Soybean (Glycine max) cv. Altona is resistant to race 4 of Phytophthora megasperma f.sp. glycinea (Pmg) at 25°C but susceptible at 32°C with reduced accumulation of the phytoalexin glyceollin. The possibility that the production and activity of elicitors is affected by temperature was examined. Active elicitor preparations were obtained from culture filtrates and cell walls from race 4 grown at 25° and 32°C. These preparations and the abiotic elicitor $AgNO_3$ were active at both 25° and 32°C. Elicitor activity was demonstrated also in inter-cellular fluids from interactions with race 4 both at 25° (incompatible) and at 32°C(compatible). It is concluded that temperature-induced susceptibility is not related to elicitor levels.

A681

VISUALIZATION OF FUNGAL FIMBRIAE USING FLUORESCENT AND GOLD-IMMUNOSTAINING. Svircev, A. M., Smith,, R. Gardiner, R. B. and Day, A. W. Dept. of Plant Sciences, University of Western Ontario, London, Ontario N6A 5B7

Surface protein fibrils (fimbriae) 7 nm wide and up to 20 μm long are extruded by cells of Ustilago violacea and many other basidiomycetes. While much shorter fibrils have been observed on the surface of cells of a wide variety of other fungi the relationships of these fibrils to the fimbriae of U. violacea is not clear, but can be determined by immunocytochemical methods. Using antisera developed against the 74,000 dalton M.W. fimbrial protein of U. violacea we have developed specific immunocytochemical stains for fimbrial proteins. The walls of a wide variety of fungal species bind FITC labelled antifimbrial antisera. Electron microscopic observations have confirmed that antibodies bind specifically to fimbriae rather than to other cell surface structures. We will describe a protein A gold immunocytochemical staining technique for fimbrial protein and tabulate fungal species which respond to this stain.

A682

TWO TOXIC COMPOUNDS PRODUCED BY HELMINTHOSPORIUM SATIVUM IN LIQUID CULTURE. Sang Sun Lee[1], Robert W. Stack[1], and Brady A. Vick[2]. 1) Plant Pathology Dept., and 2) USDA-ARS Biochemistry Dept., North Dakota State University, Fargo, ND 58105.

When Helminthosporium sativum P.K.&B. is grown in liquid culture, it produces toxic metabolites which inhibit plant root growth. Toxic metabolites from four-day-old cultures were separated from culture broth by affinity column chromatography and concentrated in methanol. From the methanol solution, two components (D,M) were separated, having 38% and 62% of the toxic activities, respectively. One component (D) was soluble in diethyl ether and had a molecular weight of 234 daltons. NMR and ultraviolet spectra of (D) were consistent with the structures of isomers of helminthosporal, a toxin previously described from H. sativum cultures. The second toxic component (M), insoluble in diethyl ether, was characterized as a peptide of 1000 to 2000 daltons and composed of at least eight different amino acids. Culture broths from different H. sativum isolates had differing proportions of the two components.

A683

THE IMPACT OF AMBIENT OZONE ON FIELD GROWN SOYBEAN. G. Smith and E. Brennan, Department of Plant Pathology, Rutgers University, New Brunswick, NJ 08903.

The impact of ambient ozone on leaf chlorophyll, leaf nitrate reductase activity (NRA) (in vivo assay) and root nodule nitrogenase activity (C_2H_2 reduction) was investigated in field-grown soybean (Glycine max (L.) Merr. var. Cutler and Williams). The antioxidant, EDU, was applied as a soil drench to half of the field plots to obtain ozone free control plants. Four times during the growing season leaf and root assays were performed on + and - EDU plants. EDU reduced the amount of visible ozone injury on both cultivars, increased leaf chlorophyll in Cutler but not in Williams, and had no effect on NRA. In contrast, C_2H_2 reduction was enhanced in +EDU plants. This effect was most apparent in both Cutler and Williams at the later pod filling stages of plant growth. EDU had no effect on plant growth, pod number or final seed yields. However, seed size was increased with EDU treatment. The data suggest that ozone was not a significant factor in the determination of soybean yields in the year this study was conducted. A prolonged dry spell mid-season may have minimized any ozone effect.

A684

PREVENTION OF THE HYPERSENSITIVE RESPONSE BY PERIPLASMIC SHOCK FLUIDS FROM ESCHERICHIA COLI CONTAINING A CLONED PECTATE LYASE GENE. C. J. Baker, USDA, Beltsville, MD 20705, and A. Collmer and M. Roy, U of MD, College Park, MD 20740.

To explore the ability of specific pectic enzymes to induce resistance in plants, the osmotic shock fluids from the periplasmic space of E. coli strains harboring recombinant plasmids were assessed for their activity in preventing the hypersensitive response (HR) of tobacco (Nicotiana tabacum 'MD201') infiltrated with Pseudomonas syringae pv. pisi. E. coli CSR2, which contains an Erwinia chrysanthemi 1237 pectate lyase isozyme (pI 8.3) gene inserted in plasmid vector PBR322 blocked HR at concentrations containing less than 1 μg/ml protein. E. coli HB101 with the original pBR322 plasmid did not block HR at protein concentrations greater than 25 μg/ml. Culture supernatants from E. chrysanthemi 1237 prevented confluent necrosis at concentrations of 2.5 μg/ml protein but was phytotoxic at concentrations greater than this.

A685

USEFUL MINIMAL MEDIA FOR XANTHOMONAS CAMPESTRIS PV. MALVACEARUM K.L. McNally, D.W. Gabriel and M.K. Essenberg, Dept. of Biochemistry and Dept. of Plant Pathology, Oklahoma State University, Stillwater, OK 74078.

Most pathovars of Xanthomonas campestris grow poorly on defined minimal media, require a large buffering capacity at neutral pH and are intolerant of high inorganic ion concentrations. In an effort to find a defined minimal medium that would support rapid growth of pathovar malvacearum (Xm), we compared a series of minimal media using inorganic phosphate or organic Good buffers and various combinations of amino acid supplements. A modified Neidhardt medium (NaCl omitted) (J. Bact. 119:736-47), using MOPS as the buffer, proved superior to the phosphate, TRIS and HEPES buffered media tested. Doubling times for different Xm races grown at 30°C with gentle aeration were about five times longer in MOPS minimal medium with glycerol than in King's medium B. Addition of alanine, arginine, asparagine, histidine, isoleucine, phenylalanine and threonine at a total concentration of 4.39 mM resulted in doubling times about twice as long as those observed in King's medium B.

A686

THE EXPRESSION OF RESISTANCE TO LEAF RUST IN Triticum aestivum 'ATLAS 66' IN THE SEEDLING STAGE. L. E. Browder; USDA-ARS, Dept. of Plant Pathology, KSU, Manhattan, KS 66506.

Atlas 66 (A66) has served as a source of leaf rust resistance in breeding programs. Heretofore, this has been viewed as an "adult plant resistance." We inoculated A66 and Thatcher (TC) seedlings with 9 Puccinia recondita cultures and exposed these materials to 5, 12, 19, and 26 C. We saw no indication of resistance at 12, 19, or 26 C. At 5 C, however, we saw differences between lines and between cultures. Infection types 56X to 78X occurred in A66 with some cultures but not others; only 99Ps were observed on TC at 5 C. At 26 C, some cultures produced a 78X with TC, indicating it also has a gene detectable in the seedling stage. In another experiment at 28-29 C, both A66 and TC produced a 78X with many, but not all, cultures. Different cultures produced X infection types with A66 at 5 C and at 28-29 C. Thus, Atlas 66 must have more than one gene associated with its adult plant resistance to leaf rust.

A687

GENETICS OF LEAF RUST RESISTANCE IN TRITICALE. Jeffrey Wilson and Gregory Shaner, Dept. Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907.

Several spring triticales were selected because of their high level of resistance to Puccinia recondita in the field. In subsequent greenhouse tests, they proved to have both an intermediate hypersensitive resistance and a long latent period. These lines were crossed with susceptible triticales. F_2 and backcross F_1 data indicate that PI 429120 possesses two dominant, additive, independent factors that confer a 0ln infection type, and that PI 429155 possesses one dominant factor that conditions a 1+c infection type. These three factors are different and independent. PI 429155 also appears to possess incompletely dominant factors for long latent period (14 days). The hypersensitive resistance of PI 429155 is expressed in the presence of long latent period factors derived from the slow-rusting line PI 434889 in the F_2 of PI 429155/PI 434889. F_2 and backcross F_1 data suggest that PI 429121 possesses two dominant, complementary factors that confer a 2- infection type and two recessive additive factors that condition a latent period of 18 days.

A688

INTEGRATION OF pUW942 INTO THE CHROMOSOME OF EXTRA-SLOW-GROWING RHIZOBIUM JAPONICUM AND FORMATION OF RECOMBINANT PLASMIDS. **A.K. Aruna and A.K. Vidaver, Dept. of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722.**

The chimeric 82 kb plasmid pUW942 was used to transfer transposon Tn501, which confers Hg^{++} resistance, into extra-slow-growing R. japonicum strains RJ19FY and RJ128. pUW942 was not maintained as a replicating plasmid in transconjugants. Southern hybridization with pUW942 and acquisition of antibiotic resistant markers of the vector plasmid showed either cointegrate formation with indigenous plasmids or integration into the chromosome of R. japonicum. Also, transconjugants with chromosomally integrated pUW942 transferred plasmids ranging from 82 to 88 kb into plasmidless Escherichia coli C. Restrictive enzyme digests of these plasmids showed not only the presence of pUW942 specific fragments but also additional fragments. Southern hybridization of the recombinant plasmids with R. japonicum plasmids showed no homology, thus the new fragments are likely to be chromosomal in origin.

A689

EFFECT OF ALLELES AT THE TOX1 LOCUS ON THE FERTILITY OF COCHLIOBOLUS HETEROSTROPHUS. C. R. Bronson, Department of Plant Pathology, Seed & Weed Sciences, ISU, Ames, IA, 50011.

Virulence genes have been hypothesized to have a pleiotropic effect on the fitness of plant pathogenic fungi. This hypothesis is being tested for the Tox1 locus of Cochliobolus heterostrophus (Helminthosporium maydis). Near-isogenic isolates of race T (TOX1) and race O (tox1) (7 backcrosses to a common parent) were crossed and backcrossed for 4 additional generations. Two sets of four siblings were mated in all combinations (within the set) to determine if there is any correlation between alleles at the Tox1 locus and fertility in the sexual cycle. The average percentage of asci with 7 or 8 mature ascospores was the same in the TOX1 x TOX1 and tox1 x tox1 crosses (29%). However, the TOX1 x tox1 crosses were significantly less fertile (an average of 2% of the asci with 7 or 8 mature spores), suggesting an effect of heterozygosity at the Tox1 locus on fertility.

A690

SECTORING IN COCHLIOBOLUS HETEROSTROPHUS. C. R. Bronson, Department of Plant Pathology, Seed & Weed Sciences, Iowa State University, Ames, IA, 50011.

Frequent sectoring has been observed in certain isolates of Cochliobolus heterostrophus and a study was undertaken to determine its cause. Single ascospores from crosses involving sectoring strains yielded cultures that sectored, suggesting that sectoring is genetically controlled but not due to a pre-existing heterokaryotic condition. To determine if the genetic elements responsible are in the nucleus or the cytoplasm, reciprocal crosses were made between sectoring and non-sectoring isolates. The frequency of sectoring in the progeny was similar regardless of which isolate acted as the female parent. This observation suggests that sectoring in these isolates is under nuclear control.

A691

GENETICS OF USTILAGO HORDEI: THE SELECTION OF FUNGICIDE RESISTANCE MUTANTS. Caroll E. Henry, Ronald W. Schaefer, Bethsheba Bullock. Department of Biological Sciences, Chicago State University, Chicago, Illinois 60628.

Mutations for the standard mating type strains I4A and E3a of Ustilago hordei (Pers.) Lagerh, were induced by UV and chemical mutagenesis. These mutants and the standard strains were tested for their resistance to chloroneb and thiabendazole using a gradient plate technique. Spontaneous fungicide resistance mutants were selected from the I4A and E3a populations. A survey of 23 induced mutants indicated different levels of resistance for chloroneb and thiabendazole.

A692

Construction of a cosmid gene library of Erwinia carotovora subsp. carotovora (Ecc). **R. T. Zink and A. K. Chatterjee, Dept. of Plant Pathology, Kansas State Univ., Manhattan, KS 66506.**

A genomic library of Ecc was constructed using the 6 kb cosmid pHC79. SalI partially digested, size fractionated Ecc DNA was ligated into the tetracycline resistance gene of pHC79 and resulting concatameric DNA was packaged in vitro at a frequency of 10^4 transducing particles per g of Ecc DNA. By interspecific complementation with E. coli (HB101), cosmids were identified which carried Ecc DNA fragments containing the gal^+, lac^+, xyl^+, pro^+, leu^+, thi^+ and $recA^+$ genes. In addition, clones were identified which produced either endopolygalacturonase, endopectate lyase or exopectate lyase which were indistinguishable from those produced by Ecc in enzymatic activity, isoelectric mobility and ability to macerate potato tuber tissue. No linkage was observed among the above phenotypes and each cosmid produced unique SalI fragments with an average insert of 30 kb.

A693

INHERITANCE OF VIRULENCE IN SINGLE ZOOSPORE PROPAGATIONS AND MASS VEGETATIVE TRANSFERS OF PHYTOPHTHORA MEGASPERMA F.SP. GLYCINEA. F.S.Rutherford and E.W.B. Ward, Agriculture Canada, Research Centre, London. Ontario, N6A 5B7.

Following zoospore inoculation of etiolated hypocotyls of 11 soybean cultivars carrying known resistance (Rps) genes to Phytophthora megasperma f.sp. glycinea (Pmg) both gain and loss of virulence was observed in successive single zoospore propagations involving two Pmg lineages. Variation observed following mass mycelial transfer was, however, significantly less than variation observed following single zoospore propagation. The data thus supports the hypothesis that expression of virulence is controlled by factors non-Mendelian in nature and possibly cytoplasmic in origin. Significantly, loss of virulence did not correlate with loss of pathogenicity since lines avirulent when tested against all 11 soybean cultivars maintained their ability to infect and elicit glyceollin production.

A694

CLONING OF VIRULENCE GENES FROM ERWINIA STEWARTII BY DIRECT COMPLEMENTATION OF AVIRULENT MUTANTS. D. L. Coplin, R. D. Frederick and D. Majerczak, Department of Plant Pathology, The Ohio State University/OARDC, Wooster, OH 44691.

Virulence genes from Erwinia stewartii have been cloned by direct complementation of avirulent mutants. A library of Hind III cleaved E. stewartii DNA was constructed in E. coli HB101 using cosmid vector pVK100. Cloned genes for leu, pro, gal and recA were expressed in HB101. Cosmids were mobilized to avirulent strains by plasmid pRK2013 and mixtures of transconjugants were inoculated into sweet corn seedlings by addition of bacterial suspensions to the whorls. Clones were obtained which restored the ability to cause watersoaked lesions to 12 mutants. In three cases, genes restoring capsule production and watersoaking were linked on the cloned fragment. Another clone complemented mutants with greatly reduced synthesis of capsular polysaccharide and UDP-galactose-4-epimerase. In many cases, comparison of clones by restriction analysis revealed a given mutant was complemented by several different inserts.

A695

FURTHER STUDIES IN TRANSFERRING LEAF RUST RESISTANCE FROM Aegilops Squarrosa TO COMMON WHEAT. W. J. Raupp, L. E. Browder, and B. S. Gill, Dept. of Plant Pathology and USDA-ARS, Kansas State University, Manhattan, KS 66506.

Five accessions of Aegilops squarrosa were tested with eleven single-pustule isolates of Puccinia recondita from the U.S. pathogenicity survey as well as culture PRTUS6. Three of the lines, TA 1649, TA 1675, and TA 1691, conditioned a 01C, 02C, or 03C infection type (IT) with all isolates. F_1 hybrids between these lines and the susceptible winter wheat Wichita (88P IT with respect to all isolates) were tested for resistance using PRTUS6. A low IT (14C) was expressed only in the TA 1649-derived hybrid. Resistant plants with 42 chromosomes in the BC_2F_2 have been obtained. Further characterization of resistance is under way. Transfer and expression of resistance in backcross-derived germ plasm from all three lines will be reported.

A696

SUBCLONING OF GENES ENCODING DIFFERENT ERWINIA CHRYSANTHEMI PECTATE LYASE ISOZYMES. Christianne Schoedel, Jeffrey L. Ried and Alan Collmer. Department of Botany, University of Maryland, College Park, MD 20742.

The use of ultrathin layer isoelectric focusing and a high resolution activity stain has revealed that the pectolytic enzymes active at pH 8.5 in Erwinia chrysanthemi strain 1237

culture supernatants can be resolved into seven bands rather than the two previously reported. Two particularly intense bands occur at pH 8.0 and 8.3. In order to determine the genetic basis for this multiplicity, a series of clones and subclones representing a 9.8 Kb region of the E. chrysanthemi chromosome has been constructed in plasmid vector pBR322 in Escherichia coli. All pectolytic E. coli transformants that are able to pit pectate semi-soft agar were found to produce the pI 8.3 pectate lyase isozyme. Prolonged activity stain incubation has revealed that at least four other bands of activity corresponding to E. chrysanthemi bands are produced by the clones. The pI 8.0 pectate lyase isozyme is not produced by subclones which lack a 2.5 Kb EcoRV fragment.

A697

EVIDENCE FOR INCREASED MELANIN CONTENT IN DICARBOXIMIDE-RESISTANT STRAINS OF MONILINIA FRUCTICOLA. S. E. Benes and D. F. Ritchie. Department of Plant Pathology, North Carolina State University, Raleigh, NC 27695-7616.

Dicarboximide-resistant strains of Monilinia fructicola could be distinguished from sensitive strains by their darker mycelial pigmentation. Extraction in hot KOH yielded pigments having absorption spectra and physical properties characteristic of melanins. The visible light absorbance of alkaline solutions of pigments extraced from fungicide-resistant strains was several times greater than that from fungicide-sensitive strains. Pigment formation in the resistant strains was sensitive to the melanin synthesis inhibitors tricyclazole (5-methyl-1,2,4-triazole (3,4-b) benzothiazole) and pyroquilon (1,2,5,6-tetrahydro-4H-pyrrolo (3,2,1-i,j)-quinolin-4-one). These data suggest that resistance to the dicarboximide fungicides is associated with increased melanin formation in M. fructicola.

A698

SECRETION OF LYTIC ACTIVITIES BY TRICHODERMA, A MYCOPARASITE OF PYTHIUM ULTIMUM. Neal Gutterson, Trevor Suslow and Gareth Warren, Advanced Genetic Sciences, 6701 San Pablo Ave., Oakland, CA 94608

Trichoderma spp. are necrotrophic mycoparasites of Pythium ultimum. To characterize the mechanism of parasitism, secretion of lytic activities has been analyzed. Trichoderma hyphae were grown in glucose minimal medium, then glucose was replaced by alternate carbon sources. Both glucanase and protease activites were derepressed in the absence of a carbon source. In addition both activities could be induced. Glucanase was induced by a) laminarin, b) Pythium cell walls, or c) Trichoderma cell walls. Protease was induced by a) BSA, b) Pythium cell walls, or c) Trichoderma cell walls. Glucanase and protease regulation is distinct. Several protease activites were resolved by DEAE column chromatography. Three activities were found under all conditions, and at least one activity was induced specifically by Pythium cell walls.

A699

ZEARALENONE INDUCTION IN FUSARIUM CULMORUM. Clarence Madhosingh and W. Orr. Agriculture Canada, Research Centre, University Sub Post Office, London, Ontario, Canada, N6A 5B7.

Total protoplast DNA from F. graminearum, a zearalenone producing species, was incubated with the protoplasts of a non-producing single spore isolate of F. c. The protoplasts were then regenerated to spores and then to individual cultures developed from single spore isolations. Each culture was examined for zearalenone production. Zearalenone was produced in 1 out of 5000 F. culmorum DNA-incubated protoplasts. Protoplasts from non-DNA incubations did not produce zearalenone and inoculations with DNA preparations did not produce cultures. The F. graminearum protoplasts regenerated zearalenone producing cultures. Radioactivity (100-300 cpm) was obtained in 1% of the tritium-labelled DNA-incubated and regenerated cultures of F. culmorum. The data indicate that the F. graminearum DNA induced zearalenone production in cultures of F. c. Further, the data suggest interspecies DNA transformation.

A700

IMMUNOCHEMISTRY OF THE EXTRACELLULAR ENZYMES OF PHYTOPHTHORA MEGASPERMA F.SP. GLYCINEA. **J.J. Goodell, L.C. Valenti and A.R. Ayers. Dept. Cell. and Devel. Biol., Harvard Univ., Cambridge, MA 02138.**

Extracellular glycoproteins of 3 races of P. megasperma f.sp. glycinea were examined immunochemically to test for predicted race-specific determinants. Polyclonal antibodies reacted more strongly with the homologous race than with antigens from other races. Relative immunogenicity of glycoproteins was assessed by western blotting and subsequent peroxidase-antiperoxidase assays or FITC tagged antibodies. Immunoprecipitation was used to remove antigens corresponding to bands on SDS-PAGE. Six degradative enzymes were assayed in fungi grown under 10 different nutrient regimes. Enzyme inactivation or binding assays were used to screen for monoclonal antibodies specific for either protein or glyco components respectively.

A701

High Performance Liquid Chromatography (HPLC) in the Purification of Kievitone Hydratase (KHase). S. W. Banks and D. A. Smith, Department of Plant Pathology, University of Kentucky, Lexington KY 40546-0091.

Previous work (Physiol. Plant Pathol. 22:129-142) indicated that KHase, the enzyme catalysing detoxification of the phytoalexin kievitone, is an extracellular, acidic glycoprotein (MW c. 102,000). Further attempts to purify KHase have involved HPLC in an aqueous system (100 mM potassium phosphate) using three Waters 300 SW "Protein Pak" gel permeation columns. The 280 nm-absorbance profile of the eluate revealed several peaks indicating partial resolution of different molecular species. Recovery of biologically-active enzyme (75% of the initial activity), with a retention time of 50 min at a flow rate of 0.5 ml/min, was achieved. The results suggest that HPLC, traditionally a technique for the separation of low MW compounds, is also of value in the purification of phytopathologically interesting macromolecules.

A702

PHENYLALANINE AMMONIA LYASE (PAL) ACTIVATION AND LIGNIFICATION AS AN INDUCED RESISTANCE MECHANISM IN TOBACCO MOSAIC VIRUS (TMV)-INDUCED TOBACCO PLANTS. B. J. McMaster and B-F. Huang. Allied Corporation, Syracuse Research Laboratory, Solvay, New York 13209

TMV infection on two local leaves of tobacco (Nicotiana tabacum cv. WS117) induced systemic resistance against subsequent challenge inoculation with either TMV or Collectotrichum destructivum as evidenced by reductions in lesion number and size. Lignification of induced plants upon challenge was demonstrated by both the change in total lignin content and histological staining. Earlier lignification occurred around infection sites on the induced tissue as compared to non-induced tissue when TMV but not C. destructivum was used as challenger. Lignification response against both challengers, however, appeared to be more intense by histological observation. Upon TMV challenge, an earlier PAL activation occurred and was prior to the lignification response in induced-as compared to non-induced tissue. The earlier PAL activation may result in earlier lignification and could be the resistance mechanism responsible for the expression of induced resistance of tobacco against TMV.

A703

THE INFLUENCE OF TEMPERATURE ON "SLOW-LEAF-RUSTING" IN Triticum aestivum 'SUWON 85.' L. E. Browder and M. G. Eversmeyer, USDA-ARS, Dept. of Pl. Path., KSU, Manhattan, KS 66506.

Suwon 85 (S85) and the spring wheat cultivar Thatcher (TC) were inoculated as seedlings with 4 cultures of Puccinia recondita Rob. ex Desm. and grown at 5, 12, 19, and 26 C. The tests were repeated 4 times. Results were variable, however, we saw definite trends. Differences in development of leaf rust between S85 and TC were greater with decreasing temperature. In some tests, differences in infection type (IT) were not classifiable at 26 C or 19 C. We saw differences in other tests at 26 C and 19 C. Large differences in IT were observed with all cultures at 12 C and 5 C in all tests. S85, with all cultures, consistently produced an X at 12 C and a 01C at 5 C in the time required to produce a 99P on TC. Cultures varied in their growth rate at 12 C and at 5 C. We conclude that the resistance of S85 is specific to culture and temperature. We expect that P. recondita variants that grow as fast on S85 as on Thatcher would occur and prevail if its resistance were deployed.

A704

ESTIMATION OF RELATIVE SPECIFICITY IN A MODEL HOST-PATHOGEN SYSTEM WITH QUANTITATIVE RESISTANCE. Anne E. Jenns, Department of Plant Pathology, North Carolina State University, Raleigh 27695.

Combinations of 50 hypothetical host and pathogen genotypes were

generated in a model system and the resulting disease severities calculated and subjected to analysis of variance. For subsamples of pathogen genotypes the highest percentage of total variance accounted for by host x pathogen genotype interaction was obtained when the most resistant genotypes in the host population and the pathogen genotype most virulent on each of those host genotypes were included in the set. A simplified method of analysis was developed to estimate the amount of specific resistance in a set of host genotypes. This method, based on the variance of disease severity adjusted to remove general virulence, was of comparable accuracy and much simpler than the previously proposed methods based on regression analysis.

A705

RESISTANCE OF WINTER BARLEYS TO LEPTOSPHAERIA NODORUM. Barry M. Cunfer, J. W. Johnson, and A. R. Brown, Dept. of Plant Pathology, Dept. of Agronomy, Georgia Station, University of Georgia, Experiment, 30212; and Dept. of Agronomy, University of Georgia, Athens, 30602.

During a five year period winter barley (Hordeum vulgare L.) cultivars and breeding lines have been evaluated for resistance to Leptosphaeria nodorum in the field at Athens and Griffin, GA. Field plots were inoculated at least twice each season with a mixture of virulent isolates. Plots were inoculated at one end only in order to observe the effects of artificial and natural inoculation. Natural infection occurred only on the most susceptible cultivars. Seedlings at the 4 to 5 leaf stage were inoculated with the same isolates in the greenhouse. Results from greenhouse and field evaluations were comparable. The diversity in reaction among barley germplasm to L. nodorum was more pronounced than that usually observed for wheat. Resistant cultivars included Dawn, Boone, Redhill, Miller, and Colonial 2. Highly susceptible cultivars included Henry, Milton, Anson, and Sussex.

A706

CHLORTETRACYCLINE BREAKS PAPILLA-MEDIATED RESISTANCE TO POWDERY MILDEW IN *ml-o* BARLEY COLEOPTILES. R. E. Gold, M. C. Stolzenburg, J. R. Aist, M. R. Marshall, C. A. Stockwell, B. E. Hazen, M. G. Smart, and H. W. Israel. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Treatment of *Ml-o* susceptible (S) and *ml-o* resistant (R) barley coleoptiles with the Ca^{2+} selective probe chlortetracycline (CTC), altered penetration efficiency (PE) of *Erysiphe graminis* f. sp. *hordei*. Compared to H_2O controls, both 100 µm CTC and 10 mM $Ca(NO_3)_2$ increased PE on (S), but only CTC increased it on (R). Time-course studies showed in coleoptiles incubated on $Ca(NO_3)_2$ that papillae were initiated in (R) 1.0 h before penetration pegs; in (S), 0.5 h after. These differences were correlated with a PE of 8% in (R) and 62% in (S). Incubation on CTC delayed papilla formation by 2.2 h and increased PE to 79% in (R), but caused no significant effects in (S). These results suggest that CTC alters some aspect of Ca^{2+} regulation and reduces resistance by delaying papilla formation. Ca^{2+} localization studies are in progress.

A707

REDUCTION OF CERCOSPORA ARACHIDICOLA SPORULATION ON PEANUT CV. 'TAMNUT 74' INFECTED WITH PEANUT MOTTLE VIRUS. H. A. Melouk and J. L. Sherwood, USDA-ARS and Plant Pathology Dept., Oklahoma State University, Stillwater, OK 74078.

Two-week old seedlings of cv. 'Tamnut 74' were inoculated with peanut mottle virus (PMV) or left untreated, and 4 weeks later sprayed with Cercospora arachidicola (2×10^4 conidia/ml) and placed in a polyethylene enclosure in a growth chamber (22-30C, 100% RH). After 3 weeks; lesions/leaflet, conidia/leaflet, necrotic area/leaflet, conidial density, and percent necrotic area were determined on 4 leaflets for each of 12 PMV infected and 12 virus-free plants. Conidia/leaflet, an important parameter in evaluating peanut resistance to C. arachidicola (Phytopathology 73:556-558), was reduced significantly (P=0.05) on PMV infected plants compared to virus-free plants in each of 4 experiments. Lesions/leaflet, necrotic area/leaflet and percentage necrosis were similarly reduced in 3 of 4 experiments and conidial density was reduced in 2 of 4 experiments. Evaluation of peanut entries for resistance to C. arachidicola may result in erroneous resistant selections if plants are infected with PMV.

A708

FIELD RESISTANCE OF OATS TO PUCCINIA GRAMINIS MEASURED IN YIELD AND SEED WEIGHT REDUCTION. M. D. Simons and P. G. Rothman. ARS, USDA. Dept. of Plant Pathology, ISU, Ames, IA 50011, and Cereal Rust Lab., University of Minn., St. Paul, MN 55108.

To locate potentially useful sources of field resistance to oat stem rust race NA 27, we tested 150 lines of oats in central Iowa in 1982. Data were expressed as indexes calculated by dividing the value of a rusted plot by the value of the corresponding rust-free control plot. Infection was severe, and all lines appeared susceptible. However, variation for quantitative response to infection was statistically significant. In 1983, 240 visibly susceptible lines were grown. Mean yield and seed weight were reduced about 50%. Statistically significant variation again occurred, with 20 and 46 lines exceeding the overall mean indexes for yield and seed weight, respectively. Heritability values were 30% and 58% for yield and seed weight indexes, respectively. There was a weak negative relationship between inherent yielding ability and resistance to stem rust.

A709

COTTON LEAF RESPONSES TO WOUNDING AND TREATMENT WITH CELL-FREE EXTRACTS OF ASPERGILLUS FLAVUS. H. J. Zeringue, E. J. Conkerton, and D. C. Chapital. Southern Regional Research Center, USDA, ARS, 1100 Robert E. Lee Blvd., New Orleans, LA 70124

The initial true leaves of one-month post emergence, Acala SJ-2 cotton plants used for response studies. Wounding initiated accumulation of ferulic acid on the third/fourth day after treatment whereas wounding and application of hot water-soluble, cell-free extracts of Aspergillus flavus mycelia induced production of scopoletin one day after treatment. Elevated levels of scopoletin were not detected in leaves that were only wounded and enhanced quantities of ferulic acid were not observed in leaf tissue after exposure to wounding/fungal extract treatment. The results indicate delineation of a wound response from a wound/fungal extract reaction. Apparently the synthesis of lignin from phenylalanine via the shikimate pathway to cinnamic acid is directed to scopoletin in the presence of the fungal extract.

A710

RELATION OF RESISTANCE TO COLLETOTRICHUM TRIFOLII IN ALFALFA TO AGRONOMIC TRAITS. S. J. Allen, J. L. Caddel, G. L. Barnes, and C. M. Taliaferro, Departments of Plant Pathology and Agronomy, Oklahoma State University, Stillwater 74078.

Effects of resistance in alfalfa to Colletotrichum trifolii Bain & Essary were studied using four bioindicator pairs developed by USDA alfalfa researchers at Beltsville, MD. (Beltsville 1-An4 and Glacier; Beltsville 2-An4 and Saranac; Beltsville 3-An4 and Vernal; and Arc and Team). Percent stand (5 yr), forage yield (4 yr), numbers of stems and plants/m.; weight of shoots and roots/m. (1 yr) were recorded. Significant differences in forage yield and percent stand were observed within semi-dormant pairs (Arc over Team and Beltsville 2-An4 over Saranac). Yield component compensation due to increased crown size in stands of dormant pairs (Beltsville 1-An4 and Glacier; and Beltsville 3-An4 and Vernal) apparently negated advantages of resistance to C. trifolii.

A711

RESISTANCE IN HORDEUM SPONTANEUM TO THREE RACES OF PUCCINIA HORDEI. Linda M. Treeful and Roy D. Wilcoxson, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

High-level resistance to leaf rust (Puccinia hordei) is generally absent in U.S. cultivated barley, but Hordeum spontaneum, a wild barley from Israel, has been reported to have resistance. Seedlings and heading adult plants of H. spontaneum were inoculated with race 8 of P. hordei, the most prevalent race. Seedlings of 29 lines were rated after 15 days. Twenty lines were resistant, six were susceptible and three had a mixture of resistant and susceptible plants. Adult plants were rated after 13 days. Eighteen lines were resistant, seven were susceptible, and four had a mixture of resistant and susceptible plants. Several lines had seedling and adult plant resistance. Similar results were obtained when adult plants were inoculated with races 13 and 19. In the future, resistance from these collections may be incorporated into resistant cultivars. Susceptible lines will be examined for slow rusting characteristics.

A712

DIRECT MEASUREMENT OF SOYBEAN TAP-ROOT TOLERANCE TO PHYTOPHTHORA MEGASPERMA F. SP. GLYCINEA. A. F. Olah and A. F. Schmitthenner. Dept. of Plant Pathology, The Ohio State University/OARDC, Wooster, OH 44691.

A simple growth chamber test for measuring soybean tap-root tol-

erance to Phytophthora megasperma f. sp. glycinea has been developed. This procedure uses modified Leath-Kendall slant-boards (Phytopathology 68:826-831) and can be completed in 14 days. After tap-root inoculation with zoospores or mycelial brei, mean rot and tolerance level of soybean cultivars are determined by measuring tap-root rot. Rot differences within a cultivar, attributed to residual genetic heterozygosity for the quantitative genes responsible for root tolerance, can be demonstrated. After measuring rot, individual plants can be grown to maturity or cloned for other uses.

A713

CHANGES IN PERCENT SMUTTED PLANTS CAUSED BY MULTIPLE INFECTIONS. B. J. Christ and C. O. Person. Department of Botany, University of British Columbia, Vancouver, B. C. V6T 2B1.

A considerable amount of variation for phenotype of smutted heads exists within a single genotype of Ustilago hordei. This variation can be accounted for by environment, physiological events and multiple infections. Multiple infection of individual seeds is known to occur and can possibly alter the amount of smut observed. A theoretical examination of multiple infections was made using data of U. avenae where the homozygous virulent, heterozygous and homozygous avirulent gave averages of 80%, 8% and 0% smutted plants, respectively. One, 2 and 3 infections per seed with a mixed population of the 3 genotypes gave expected values of 24%, 42% and 56% smutted plants, respectively. With a heterogeneous population of smut, multiple infections can play a role in the variability of disease expression within and among plants.

A714

DISINFECTION OF SEED-SURFACES WITH SODIUM HYPOCHLORITE. D. B. Sauer and R. Burroughs, USDA, ARS, U.S. Grain Marketing Res. Lab, and Dept. of Grain Sci., KSU, Manhattan, KS 66502.

Aspergillus glaucus grew from up to 100% of heavily surface-contaminated wheat kernels when put on agar after standard treatments with sodium hypochlorite (NaOCl); the wheat was known to be free from viable internal fungi. Aspergillus spp. spores were killed almost instantaneously by 1-5% solution of NaOCl, so the problem appeared to be lack of contact between spores and NaOCl, because of air bubbles, cracks, surface hairs, or debris on seed surfaces. Rinsing seeds in ethanol before washing in NaOCl improved effectiveness, especially with wheat, but rinsing in wetting agents did not. Reducing the pH of NaOCl solutions from 11-12 to about 8 increased effectiveness, but such solutions were unstable. Some literature reports on growth of storage fungi in grain may be inaccurate because of failure to eliminate surface contaminants.

A715

GROWTH OF ASPERGILLUS AND PENICILLIUM SPECIES FOLLOWING MIXED INOCULATION IN STORED GRAIN. D. B. Sauer, USDA, ARS, U.S. Grain Marketing Res. Lab, Manhattan, KS 66502

Corn was inoculated with various Aspergillus and Penicillium species and stored in environments with 80 to 90% relative humidity (RH) to provide a range of moisture conditions. Aspergillus glaucus and A. restrictus grew at 80% RH regardless of temperatures from 10 to 35°C, whereas A. candidus growth was observed at that humidity at 20° and 25° but not at 30°C. A. ochraceus and Penicillium citrinum infected the corn at or slightly below 85% RH but A. niger and A. flavus required an RH above 85%. When adequate moisture and temperature prevailed, A. candidus caused the most damage in terms of germination reduction, moisture increase, ergosterol content and visible mold on the kernels.

A716

OCCURRENCE OF FUSARIUM MONILIFORME ON CORN ASSOCIATED WITH EQUINE LEUCOENCEPHALOMALACIA. Ronald F. Vesonder, J. J. Ellis, J. Haliburton, W. B. Buck, and J. F. Tuite, Northern Regional Research Center, ARS, USDA, Peoria, IL 61604; Texas A&M, Amarillo, TX 79116; University of Illinois, Urbana, IL 61801; Purdue University, West Lafayette, IN 47907.

Mycological examination of corn associated with equine leucoencephalomalacia (ELEM) by plating surface-sterilized kernels on agar surfaces revealed 100% Fusarium moniliforme in the samples and 50% Penicillium sp. Twenty-seven F. moniliforme strains were isolated from ELEM-associated corn grown in Oklahoma, Illinois, or Indiana. The strains were cultured in the laboratory on corn at 25° C for 14 days. The fermented corn, dried in a forced-air oven at 76° C for 24 hours, was analyzed for T-2 toxin, diacetoxyscirpenol, vomitoxin and zearalenone; none were detected. It was then fed to 1-day-old ducklings to determine if other toxins might be present. Acute mortality occurred in ducklings fed corn fermented with 18 of the 27 F. moniliforme strains; ducklings that died had slightly swollen, diffusely reddened livers, and little body fat.

A717

PLUM LEAF SCALD BACTERIA: SURVIVAL THROUGH WINTER. C. J. Chang and C. Yonce. Department of Plant Pathology, Georgia Station, University of Georgia, Experiment, GA 30212, and USDA Southeastern Fruit and Tree Nut Lab., Byron, GA 31008.

Direct isolation of xylem-limited bacteria (XLB) on CS20 agar medium was used to assess bacterial survival. Thirty to forty twigs (0.5-1.0 cm X 5-8 cm) were randomly sampled from each of the three plum leaf scald trees from November 1982 to December 1983. Sap obtained from squeezing each surface-sterilized twig with pliers was immediately placed on CS20 agar plates. Plates, enclosed in plastic bags and incubated at 30 ± 1 C, were examined microscopically (30-50 X) every week after inoculation for colonies. Bacteria were isolated each month from Nov. 1982 to Dec. 1983 except May 1983 for two trees and each month from July 1983 to Dec. 1983 for the other tree. Results indicate that XLB can survive through the winter at Byron, GA and suggest that the XLB may be transmitted to healthy trees anytime vectors are present.

A718

ISOLATION AND CHARACTERIZATION OF TN5 INDUCED MUTANTS OF PSEUDOMONAS SYRINGAE SUBSP. SYRINGAE ALTERED IN SYRINGOTOXIN PRODUCTION. M. K. Morgan and A. K. Chatterjee, Dept. of Plant Pathology, Kansas State University, Manhattan, KS 66506.

A syringotoxin (ST) producing strain of P. syringae was subjected to Tn5 transposon mutagenesis using the plasmid vector pSUP1011. Upon screening 2193 insertion mutants for ST production, several classes of prototrophic toxin mutants were identified. Seven clones produced no detectable levels of ST. Two apparent transport mutants secreted less than 5% of wild type levels and showed growth inhibition on media that stimulates toxin production. One mutant produced 5-10% of normal toxin levels, and 5 mutants with a probable cell wall alteration produced about 10% of wild type levels. Genomic DNA from non-toxigenic mutants was digested with Eco RI, electrophoresed, and probed with Tn5 in Southern blot analyses. These mutants appeared to contain single Tn5 insertions into one of two chromosomal Eco RI fragments.

A719

EVALUATION OF COPPER OXYCHLORIDE SPRAYS TO CONTROL BACTERIOSIS DISEASE OF MEXICAN LIME TREES IN COLIMA, MEXICO. J. J. Stapleton, USDA-ARS, and V. M. Medina U., INIA, CAE Tecoman, Apartado Postal #88, Tecoman, Colima, Mexico.

An emergency spray program consisting of periodic applications of copper oxychloride (2.5 g AI/l) on developing shoots has been initiated to control citrus bacteriosis disease (CBD), thought to be caused by Xanthomonas sp., of foliage and twigs of Mexican lime trees in Colima, Mexico. To test the efficacy of this program, field experiments were done in two heavily infected commercial groves. Seven treatments varying the timing, frequency, and total dosage of spray applications were tested on developing flushes. Infected leaves (33-86%), number of lesions/leaf (69-92%) and number of lesions/infected leaf (29-70%) were usually reduced after 2 to 4 applications 5 days apart. Even with 4 applications over a 20-day period, however, at least 7% of leaves were infected when assayed 3 wk after the final spray. This spray program may not effectively control CBD of Mexican lime trees except when disease severity is very low.

A720

EVALUATION OF CELLULAR PROTEIN PROFILES FOR IDENTIFICATION OF CORYNEBACTERIUM MICHIGANENSE SSP. NEBRASKENSE AND C.M. SSP. TESSELLARIUS FROM INFECTED PLANTS. B. JOSHI and A.K. Vidaver, Dept. of Pl. Path., Univ. of Nebraska, Lincoln, NE 68583-0722.

A comparative study was made of total cell protein extracts from bacteria grown on NBY agar and bacteria isolated from greenhouse-infected plants. Bacteria were extracted from >5 gm infected tissues and purified on 10-60% discontinuous sucrose density gradients. The yellow-orange pigmented bacteria could be retrieved principally in the 50-60% interphase of the gradient. Cellular proteins were extracted with lysozyme:SDS:phenol. The extracts were subjected to SDS-PAGE on

gradient slab gels and silver stained. Protein patterns of C.m. ssp. nebraskense (Cmn), causative agent of Goss's bacterial wilt and blight of corn, grown in vitro on NBY agar and from infected plants were indistinguishable. In contrast, similar procedures showed only a few proteins in common between C.m. ssp. tessellarius (Cmt), causative agent of bacterial mosaic of wheat, from wheat plants and NBY agar. Thus, unambiguous identification is feasible for Cmn, but not for Cmt.

A721

GROWTH OF CORYNEBACTERIUM MICHIGANENSE SSP. NEBRASKENSE on corn (ZEA MAYS L.) CALLUS TISSUE. T.R. Rocheford, A.K. Vidaver, and C.O. Gardner, Dept. of Agronomy and Dept. of Plant Pathology, Univ. of Nebraska, Lincoln, NE 68583.

Callus was generated from the first internode of corn seedlings and subcultured on modified Murashige and Skoog medium. Two corn hybrids were used: Mo17 x B73 and A632 x A619, which are resistant and susceptible, respectively, to Goss's bacterial wilt and blight in the field. Callus pieces were abraded with sterile microforceps and then inoculated. A micropipette was used to place a 5 µl droplet containing 5 CFU of Corynebacterium michiganense ssp. nebraskense (Cmn) onto the wounded callus pieces. At various times after inoculation, callus pieces were sampled and homogenized in phosphate buffer. Homogenates were diluted serially and plated onto a selective medium (CNS). Populations of Cmn increased on both genotypes of callus but were higher on susceptible than resistant callus at most sampling times. This technique offers promise in screening for resistance to Goss's bacterial wilt and blight of corn.

A722

SEPARATION AND ACTIVITY OF EXTRACELLULAR PECTOLYTIC ENZYMES OF ERWINIA CAROTOVORA. R. S. Livingston, E. A. Maher, and A. Kelman, Department of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706.

When incubated in air potato tubers were more resistant to maceration by Erwinia carotovora pv carotovora (Ecc) and pectolytic filtrates of the bacterium than when incubated anaerobically. Partially purified preparations with PG and PL activity from culture filtrates of Ecc co-chromatographed on CM-Sephadex and P-60. The pectic enzyme fraction was eluted from the CM-Sephadex column then bound to an Agarose (Bio-Gel A 0.5 m) column in 25 mM Tris pH 8.5. Two PL's and one PG were separated when the Agarose column was eluted using a gradient of 0-80 mM NaCl. Tissue maceration by purified endo-PL, the major extracellular pectic enzyme, was significantly greater in tubers incubated anaerobically than in those incubated aerobically. Thus, the relative resistance of tuber tissue in air to Ecc may be dependent on mechanisms that block tissue maceration by endo-PL.

A723

THE USE OF INFECTIVITY TITRATIONS TO COMPARE RELATIVE RESISTANCE OF TOBACCO TO PSEUDOMONAS SYRINGAE PV. TABACI. Kim Knoche and R. W. Fulton, Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706

The relative resistance of three cultivars of tobacco to Race 0 and Race 1 of Pseudomonas syringae pv. tabaci was compared. Dilutions of the bacteria were infiltrated into month-old tobacco with a hypodermic syringe. Eight days later the number of necrotic responses (percent response) for each dilution was recorded and the median effective dose was calculated for each race on each cultivar. These were used to compare the relative resistance of each cultivar to the race of bacteria. Havana 142 was susceptible to Race 0 and to Race 1. Havana 503 was susceptible to Race 1 but resistant to Race 0. By this method line 8A2S4-9 was susceptible to Race 1 and resistant to Race 0; however, in field inoculations line 8A2S4-9 was resistant to both races.

A724

INFLUENCE OF WOUNDING AND TIME IN STORAGE ON SUSCEPTIBILITY OF RUSSET BURBANK POTATOES TO ERWINIA CAROTOVORA (Ec). E. A. Maher and A. Kelman, Department of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706.

Potatoes sampled at intervals and bruised with a pendulum bruiser developed soft rot lesions at bruise sites following 48 h mist chamber incubation. Soft rot severity and percent infection increased when bruise sites were inoculated immediately with >10^4 CFU/site yet decreased when bruised tubers were held at 20 C and 92% R.H. for 72 h prior to inoculation and mist chamber incubation. Percent infection and degree of maceration were significantly higher at bruise sites than at shallow horizontal cuts made in the potato periderm. Susceptibility declined during the first 15 weeks when tubers were assayed by injection and anaerobic incubation. This pattern was not observed in tubers assayed by bruising and mist chamber incubation. Type of injury apparently influences the wound response in potato tissue and thereby affects soft rot incidence and severity.

A725

Diagnostic Procedures for Plant Diseases Caused by Erwinia carotovora and Pseudomonas solanacearum. J. C. Trolinger, R. K. Jones, and B.C. Raju, Technical Division, Yoder Bros., Inc., Alva, FL 33920 and Dept. of Plant Pathology, N. Carolina State University, Raleigh, NC 27650

Diagnosis is the first step in the study of plant disease. With the increased demand for the production of economic crops comes the need for greater expediency and accuracy in plant disease diagnoses. A plant disease diagnostic lab must use reliable techniques in order to operate. Techniques involving various selective media have been utilized in the effective diagnoses of bacterial diseases. Procedures useful in the diagnosis of soft rot (Erwinia carotovora) and bacterial wilt (Pseudomonas solanacearum) will be presented.

A726

FLUORESCENT-ANTIBODY DETECTION AND ENUMERATION OF THE RATOON STUNTING DISEASE BACTERIUM. M. J. Davis. University of Florida, REC, Ft. Lauderdale, FL 33314.

A fluorescent-antibody, direct-count technique was developed for detection and enumeration of Clavibacter xyli subsp. xyli. The fluorochrome-antibody conjugate was prepared with fluorescein isothiocyanate (FITC) and immunoglobulin G (IgG) specific for C. xyli purified by affinity chromatography. Sap was extracted by centrifugation of sugarcane internodes. Equal volumes (0.1 ml each) of sap and diluted (1:40) FITC-IgG conjugate were mixed, incubated for 30 min, and then diluted to 2-5 ml with buffer. Fluorescent-labelled bacteria in each preparation were then collected by filtration on the surface of a 13-mm Nucleopore membrane filter (0.2 µm) previously stained with Sudan black B. The bacteria were observed with epifluorescence microscopy (x1000). Bacterial counts were found to follow a Poisson distribution and one cell/microscope field was equivalent to ca. 4 x 10^4 cells/ml of sap.

A727

FATTY ACID PROFILES OF BACTERIA CAUSING RATOON STUNTING DISEASE (RSD) OF SUGARCANE AND BERMUDAGRASS STUNTING DISEASE (BSD). A. G. Gillaspie, Jr., USDA-ARS, Beltsville, MD 20705; M. Sasser, U. of DE, Newark, DE 19717-1303; M. J. Davis, U. of FL, Ft. Lauderdale, FL 33314.

Whole cell fatty acid extracts from Clavibacter xyli subsp. xyli (RSD pathogen) and from C. xyli subsp. cynodontis (BSD pathogen) grown in RSD broth and C. michiganese subsp. michiganese grown on trypticase-soy agar or RSD broth were analyzed by gas chromatography. The extracts contained 17-24%, 26-31%, and 50-57% 15:0 anteiso, 5-14%, 8-13%, and 8-10% 16:0 iso, and 62-72%, 54-60%, and 31-35% 17:0 anteiso acids for C.x.x., C.x.c. and C.m.m., respectively. In addition, C.m.m. contained 3-6% 15:1 anteiso and 2-3% 16:0 acids. These bacteria could be differentiated based on the ratio of 15:0 anteiso to 17:0 anteiso acids (approx. 1:3, 1:2, and 5:3 for C.x.x., C.x.c., and C.m.m., respectively) and on the presence/absence of 15:1 anteiso and 16:0 acids.

A728

SURVIVAL OF ERWINIA AMYLOVORA ON NON-HOST FLOWERS OF SWEET CHERRY. Sherman V. Thomson, Department of Biology, Utah State University, Logan, Utah 84322

Epiphytic colonization by Erwinia amylovora of flowers of fire blight hosts such as pear and apple occurs on the stigmatic surfaces of pistils. Multiplication occurs on apparently healthy stigmas, and populations frequently reach 10^6 to 10^7 cells per flower. However, there generally is no symptom development on these colonized host flowers. Erwinia amylovora survived on flowers of non-host sweet cherry flowers for up to 17 days in the field, but populations did not exceed the original inoculum levels. No symptoms were observed. Survival was only slightly better on the pistils when compared to survival

on other flower parts. These data suggest that the stigmas of host flowers are conducive for growth of E. amylovora whereas non-host cherry flowers allow survival but do not permit significant multiplication.

A729

AN INDICATOR DYE FOR XANTHOMONAS CAMPESTRIS PV. TRANSLUCENS. D.C. Sands, G. Mizrak and V. Hall, Plant Pathology Dept., Montana State University, Bozeman, MT 59717.

Most plant associated bacteria are unable to utilize lactose as a carbon source. All strains of the bacterial leaf streak pathogen (Xanthomonas campestris pv. translucens) can utilize lactose. Tests of the bacteria in culture indicated that they produce B-galactosidase, even in the absence of lactose, as indicated by chromogenic hydrolysis of 5-bromo-4-chloro-3-indolyl B-D-gluco-pyranoside (20 mg/l). The presence of the bacterium on or in barley seed can be visualized by overnight incubation with this colorless stain in 0.05 M potassium phosphate buffer, resulting in a blue reaction.

A730

POPULATION DYNAMICS AND SYRINGOMYCIN BIOASSY TO EVALUATE RESISTANCE OF SPRING WHEAT CULTIVARS TO PSEUDOMONAS SYRINGAE PV. SYRINGAE. W. W. Shane and J. S. Baumer. Department of Plant Pathology, University of Minnesota, St. Paul 55108.

Resistance of wheat lines to bacterial blight (BB), caused by Pseudomonas syringae pv. syringae, was evaluated. Leaves of wheat in the boot stage were infiltrated with bacterial suspensions at 5 X 10E6 cfu/ml. Inoculated plants were incubated at 18-20 C under mist after macroscopic signs of watersoaking dissipated. Symptom and bacterial population development revealed differences among the 18 wheat lines tested. Symptoms correlated closely with bacterial populations after 3 to 5 days incubation: chlorosis, grey-green necrosis and watersoaking corresponded to populations >10E6 cfu/cm2 leaf tissue, moderate chlorosis alone with titers of 10E5 to 10E6, and faint or no symptoms with lower titers. Wheat lines were approximately equal in sensitivities to syringomycin solutions applied to leaf surfaces--thus toxin bioassay appears to be an unsatisfactory means to evaluate the resistance of wheat to BB.

A731

ADAPTION OF SEEDBORNE BACTERIAL PATHOGENS TO LOW WATER POTENTIALS in vitro. P. Graham and B. Kennedy. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul, MN 55108.

Pseudomonas syringae pv. glycinea race 2 was grown in glucose-salt medium amended with NaCl to -6x,-13x,-21x,-27x, and -38x10^8 mPa. Initial populations were 10^4 CFU. Comparisons were made between nonadapted cultures (grown at -6x10^8 mPa) and adapted cultures (grown at -27x10^8 mPa) in terms of a) slope of log phase growth (CFU/hr) and b) time (hr) to attain 4x10^7 CFU. No differences occurred in growth patterns between cultures at -6x or -13x10^8 mPa. At -27x10^8 mPa, slopes of nonadapted and adapted cultures were 3.8x10^6 and 5.6x10^6. At -38x10^8 mPa, growth was observed only in adapted culture with a slope of 1.2x10^6. At -27x10^8 mPa, times to attain 4x10^7 CFU for nonadapted and adapted cultures were 37.1 and 28.7 hr. At -38x10^8 mPa, time of adapted culture was 167 hr. Similar patterns were observed when medium was amended with sucrose. These growth patterns also were observed with Ps. syringae pv. phaseolicola.

A732

EPIDEMIOLOGY AND CONTROL OF BACTERIAL CANKER OF PAPAYA CAUSED BY AN ERWINIA SPECIES IN ST. CROIX, U. S. VIRGIN ISLANDS. R. Webb CVIAES, St. Croix, USVI 00850

A species of Erwinia was determined to be the cause of systemic, firm, water-soaked cankers on the stems, and angular, water-soaked lesions on the leaves of papaya trees in St. Croix. Trees affected with this disease usually topple soon after stem cankers are observed. The pathogen does not survive longer than 2 weeks in the soil, but may survive indefinitely in leaf lesions or cankers of affected trees and on the leaves of suitable non-hosts. A duration of free moisture, except for the amount needed for dispersal and/or deposition, is not necessary for infection nor is it required for pathogen survival on leaf surfaces. Attempts to control the disease with antibiotics, commercial bactericides, and an antagonistic bacterium were unsuccessful. However, a high degree of resistance was observed in a number of land varieties from St. Croix and the Eastern Caribbean. Commercial varieties from Hawaii, Puerto Rico, Costa Rica, and Jamaica were highly susceptible to the canker disease.

A733

ANTIBIOTIC PRODUCTION IN ERWINIA HERBICOLA STRAIN C9-1. C. Ishimaru and E. J. Klos, Department of Botany and Plant Pathology, Michigan State Univ., East Lansing, MI 48824-1312

Erwinia herbicola strain C9-1, isolated from a Michigan apple orchard, produces compounds with antimicrobial activity in vitro. On completely defined and buffered medium some gram positive and gram negative bacteria are inhibited by culture filtrates of E. herbicola C9-1. Two compounds with different antimicrobial activity can be isolated from these supernatants by reversed phase HPLC. Antibiotics produced by E. herbicola are called herbicolins whereas their bacteriocins are called herbicolacins. The antimicrobial activities of E. herbicola C9-1 purified from culture filtrates are therefore called herbicolins designating their antibiotic nature. One of these purified herbicolins is inhibited by the addition of L-histidine, L-histidine dipeptides and histidinol into the overlay medium. Another herbicolin is inhibited by some as yet unknown constituent(s) of yeast extract. Other E. herbicola isolates are being screened for potential antibiotic production.

A734

INFLUENCE OF TIMING OF APPLICATION ON CHEMICAL CONTROL OF BACTERIAL SPECK OF TOMATOES. D.J. Jardine and C.T. Stephens, Department of Botany and Plant Pathology, Michigan State Univ., E. Lansing, MI 48824

Bacterial speck (Pseudomonas syringae pv. tomato) continues to be a serious disease problem in Michigan tomatoes. Recommended spray schedules of 7-day intervals with fixed coppers have provided little control. Greenhouse experiments were conducted to determine the effect of timing of application of selected chemicals on artificially inoculated plants. Streptomycin sulfate, oxytetracycline and a copper-maneb complex were applied at various times prior to or after inoculation with P. tomato. Only streptomycin provided significant control and then only if applied within 2 days before or 1 day after inoculation. Sprayed plants exposed to simulated rain conditions showed increased disease susceptibility as compared to sprayed plants alone.

A735

USE OF TWO-DIMENSIONAL ELECTROPHORESIS TO IDENTIFY SOFT-ROTTING BACTERIA. H. E. Moline, USDA, ARS, Beltsville, MD 20705

Two dimensional polyacrylamide gel electrophoresis (2-D PAGE) was used to differentiate a number of Erwinia and Pseudomonas strains. Forty erwinias including E. carotovora, E. atroseptica, E. chrysanthemi, E. rhapontici, E. amylovora, E. herbicola, and unknown soft-rotting strains were compared to Pseudomonas fluorescens strains. Profiles of acidic ribosomal enriched proteins allowed differentiation of all strains. The technique has proven that soft-rotting bacterial strains can be readily differentiated and that E. carotovora and E. atroseptica are consistently distinct enough to separate them into two species and further subgroup strains within a species. 2-D PAGE should prove helpful to better characterize other heterogeneous genera such as E. herbicola.

A736

THERMOSENSITIVE CHARACTERS ASSOCIATED WITH VIRULENCE IN AGROBACTERIUM TUMEFACIENS. CHAREST, Pierre J. et Patrice DION. Département de phytologie, Université Laval, Québec, Qc, Canada G1K 7P4

Plants of Kalanchoe fedtschenkoi have been inoculated with A. tumefaciens C58CI (pTi B6S3) and incubated at 20°C and 27°C. Tumors and adventitious roots developed at similar rates at these two temperatures. However secondary shoot formation which is a characteristic feature of Kalanchoe fedtschenkoi inoculated with octopine strains - occured only at 20°C. Two lines of evidence show that thermosensitivity of secondary shoot formation is at least partly determined by the Ti plasmid. First, with the nopaline strains T-37, C58 and 213B secondary shoot formation was not thermosensitive. Second, chemical in vitro mutagenesis of pTi B6S3, and subsequent transformation into C58CI, yielded a transformant that elicited faster secondary shoot formation than the control at 20°C.

A737

CHANGES IN LEAF SURFACE CHARACTERISTICS INFLUENCE THE MEAN, VARIANCE, AND NUCLEATION FREQUENCY OF EPIPHYTIC ICE NUCLEATION ACTIVE BACTERIAL POPULATIONS. D. M. Haefele and S. E. Lindow. Dept. of Plant Pathology, University of California, Berkeley, CA 94720.

Physical, chemical, and genetic changes in leaf surface characters tended to raise mean populations of epiphytic Pseudomonas syringae pv. syringae per leaf and lowered population variance and nucleation frequency. Abrasion without visible injury increased populations of epiphytic ice nucleation active bacteria (EINAB) from 5.576 to 6.705 mean log CFU gram^{-1} fr. wt. Plants treated with the herbicide EPTAM (Stauffer), known to disrupt epicuticular wax deposition, and untreated controls had EINAB populations of 7.451 and 5.804 mean log CFU gram^{-1} fr. wt., respectively. Mean populations (mean log CFU gram^{-1} fr. wt.) and variance for EINAB on glossy mutants of corn (having less or altered epicuticular wax) and near isogenic controls were: x = 5.861, var = 0.088 and x = 5.166, var = 0.284, respectively. Nucleation frequencies were lowered by as much as one order of magnitude on plants with altered leaf surfaces.

A738

IMPLICATION OF PHOSPHOLIPASE IN A BACTERIALLY INDUCED HYPERSENSITIVE REACTION IN TOBACCO. M. A. Roy and M. Sasser, University of Delaware, Newark, DE 19711.

Several proven inhibitors of phospholipase were used to probe the role of the enzyme in the hypersensitive response (HR) to an incompatible bacterial pathogen. Compounds were infiltrated into leaves of tobacco 'MD 201' along with inocula of Pseudomonas syringae pv. syringae 61. Inhibitors which prevented subsequent confluent necrosis included butacaine, chlorpromazine, mepacrine and U10029 (Upjohn). That these compounds are active at micromolar concentrations suggests a possible role for phospholipase in events leading to the HR. Studies conducted in vitro determined that HR prevention by the compounds was not due to inhibitory effects on bacterial growth.

A739

COMPUTER ASSISTED IDENTIFICATION OF BACTERIA BASED ON FATTY ACID ANALYSIS. J. M. Sasser, D. J. Fieldhouse, Dept. of Plant and Soil Science, University of Delaware, Newark, DE 19717-1303 and C. N. Carter, Hewlett-Packard, Avondale, PA 19311.

A computer generated 'library' of bacterial fatty acid profiles has been acquired through gas chromatographic assay of whole cell extracts of thousands of strains of Agrobacterium, Corynebacterium, Erwinia, Pseudomonas and Xanthomonas. One computer program names the fatty acid peaks; another then searches the 'library' for similarities, allowing tentative identification of an unknown culture within 70 minutes.
The fatty acid profiles have been found to be complex, stable and may be highly diagnostic at the species level and often at the pathovar level.

A740

POPULATIONS OF XANTHOMONAS CAMPESTRIS PV. VESICATORIA ON LEAVES ACCOMPANYING NITROGEN AND POTASSIUM FERTILIZATION TO TOMATOES. R. G. McGuire and J. B. Jones, GCREC-Bradenton, FL 34203

Bacterial spot, caused by Xanthomonas campestris pv. vesicatoria (Xcv), is a serious problem on leaves and fruit of tomato. Resistant cultivars are not available, but fertilizer applications may promote some control. Transplants in plastic-mulched beds received combinations of N at 4 or 8 lbs and K at 4, 8 or 16 lbs/100' row applied through a drip irrigation system. An Xcv mutant, resistant to rifampicin and streptomycin, was sprayed onto foliage at 10^8 cfu/ml, and populations were monitored weekly by culturing leaf washings on nutrient agar supplemented with rifampicin and streptomycin. Populations averaged 10^5 cfu/g leaf through day 8, rose to 7 x 10^5 by day 15, then slowly declined to 3 x 10^4 by day 43. Although K applications alone did not affect populations, a significantly lower colony count accompanied the higher rate of N. The interaction of the two elements was more significant, with reduced populations accompanying increased K at the higher N rate.

A741

Systemicity of Agrobacterium radiobacter pv. tumefaciens and low frequency of galling on chrysanthemum cuttings. J. B. Jones and B. C. Raju. GCREC-Bradenton, FL 34203.

In one study, Surf and Circus chrysanthemum plants were inoculated by a strain of Agrobacterium radiobacter pv. tumefaciens (ART). After 1, 2 and 4 weeks, cuttings were taken from inoculated plants aseptically. Two to 3 mm thick chips were cut from the base of each cutting. The chips were assayed for ART. The cuttings were grown for 6-8 weeks to check for gall development. ART was isolated from 1 of 59 chips. Galls developed at the cut surface of 3 of 57 plants after 6-8 weeks, whereas, 4 of 58 rooted cuttings developed galls. In a second study, 5 ART strains were inoculated separately at the base of Circus chrysanthemum plants. Only 2 of 50 inoculated plants developed galls at the site of the cutting. Also, only 2 of 50 cuttings developed galls. The bacterium was observed to be systemic. Systemic detection of the bacterium was low in frequency and had little effect on galling of cuttings or the cut surface of the stock plant.

A742

Evaluation of a selective medium for the isolation of Xanthomonas campestris pv. vesicatoria from seeds and foliage. R. G. McGuire and J. B. Jones. GCREC-Bradenton, FL 34203

Isolation and identification of Xanthomonas campestris pv. vesicatoria (Xcv), the pathogen inciting bacterial spot of tomatoes and peppers, from diseased tissue is frequently difficult. Although the yellow, mucoid appearance of the colony is very suggestive of Xanthomonas, Erwinia herbicola is similarly colored and present with other saprophytes on most leaves. A combination of antibiotics can be added to a solid medium containing Tween, a fatty acid ester, to reduce saprophytic organisms and distinguish Xcv by pigment intensification and lypolytic activity. The basal medium contains (per L of DW) 10 g of Bacto peptone, 10 g of KBr, 0.25 g of CaCl, and 15 g of agar. After autoclaving, 10 ml of Tween 80 are added with the antibiotics cephalexin, 5-fluorouracil, and tobramycin at 25, 6, and 0.4 mg/L, respectively. Cycloheximide at 75 mg/L is added to control additional fungi. All 30 strains of Xcv tested grew on this medium and were intensely yellow; 60% were lypolytic, whereas other yellow bacterial species did not grow.

A743

EFFECT OF FLOODING AND FUNGICIDE TREATMENTS ON SEVERITY OF PHYTOPHTHORA ROOT ROT OF TAXUS. John R. Hartman and William P. Clinton, Department of Plant Pathology and Robert E. McNiel, Department of Horticulture and Landscape Architecture, University of Kentucky, Lexington, KY 40546.

Rooted Taxus media ('Hicksii') cuttings were subjected in greenhouse tests to the following H_2O flooding regimes: 8 hr, weekly; 24 hr, weekly; 48 hr, biweekly; continuous flooding; and no flooding with watering as needed in well-drained soil. Each regime was applied to Taxus growing in soil with and without Phytophthora cinnamomi. All flooding treatments significantly enhanced chlorosis and necrosis of foliage, stunting, and root decay in pathogen-containing soil. Although root browning and foliar discoloration in the absence of the pathogen were enhanced significantly by flooding, only continuous flooding reduced growth and in each case, root and shoot symptoms were less severe than when the pathogen was present. Metalaxyl and ethazole treatments reduced root rot symptoms for 4 months compared to propamocarb and no fungicide when pathogen-infested Taxus soils were flooded 48 hr biweekly.

A744

RAPID DETECTION OF XANTHOMONAS CAMPESTRIS PV. PELARGONII IN PELARGONIUM X HORTORUM. Tuinier, J.E. and C.T. Stephens. Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824

Bacterial wilt of geranium caused by Xanthomonas campestris pv. pelargonii (Brown)Starr and Burkh. is a serious problem to the geranium industry. The bacterium can survive in infested symptomless stock plants which can serve as inoculum sources for spread throughout the greenhouse. A method of detection was developed to allow rapid identification of the pathogen on stock plants being introduced into the greenhouse. Antiserum was prepared in rabbits injected with live bacterial antigen. The antiserum was then used in an enzyme-linked immunosorbent assay (ELISA). Positive results were obtained only from infected geranium plants while none of the check plants gave a false positive result. Numerous other plant pathogenic and saprophytic bacteria were also tested and of those that tested positive, none are normally found associated with geranium culture.

A745

INCREASED PYTHIUM ULTIMUM MORTALITY ON 'RINGO SCARLET' GERANIUMS TREATED WITH SILVER THIOSULPHATE. M.K. Hausbeck, C.T. Stephens*,

and R.D. Heins, Departments of Horticulture and *Botany & Plant Pathology, Michigan State University, East Lansing, MI 48824

Silver thiosulphate (STS) prevents petal abscission in seed geraniums. Increased mortality due to crown and root rot caused by Pythium ultimum was observed after STS foliar treatment. Geraniums not treated with STS but grown in Pythium-infested media appeared healthy except for significantly reduced plant size. When grown in Pythium-infested media, Pythium was always reisolated from the root tissue of plants treated and not treated with STS. The cultivar 'Ringo Scarlet' grown in Pythium infested soil had a higher death rate when sprayed with STS. Mortality due to Pythium in STS treated plants was reduced by using appropriate fungicides. Studies suggest that foliar sprays of STS increased the decline in plants already infected with Pythium.

A746

CONTROL OF DAMPING-OFF PATHOGENS IN SOILLESS CONTAINER MEDIA. C. T. Stephens and T. C. Stebbins, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824-1312

Control of damping-off pathogens with commonly used greenhouse fungicides was variable and dependent on the growing medium. Damping-off of impatiens seedlings, due to Rhizoctonia solani or Pythium ultimum, was readily controlled in peat, composted pine bark and composted hardwood bark media drenched with the more efficacious fungicides. Damping-off control was least effective in processed pine bark. Overall, metalaxyl outperformed several other fungicides in controlling Pythium damping-off in this study. PCNB was slightly more effective than ethazole plus thiophanate methyl and benomyl at low rates but all provided excellent control of Rhizoctonia damping-off. There was less Pythium and Rhizoctonia damping-off in untreated composted hardwood bark than in other media tested. The addition of fungicides did not adversely affect the suppressive characteristics of this medium.

A747

SUSCEPTIBILITY OF DIFFERENTLY AGED CRABAPPLE AND HAWTHORN LEAVES TO GYMNOSPORANGIUM RUSTS. Dan Neely, Botany and Plant Pathology Illinois Natural History Survey, Champaign, IL 61820.

Ornamental crabapple and hawthorn are indeterminate in growth habit and can produce more than 20 leaves per shoot per year. In east-central Illinois leaf emergence occurs from late April into July. Phenological and epidemiological observations over 12 years indicate that host/pathogen/environmental factors favoring severe natural infection of Malus coronaria, M. pumila, and Crataegus oxyacantha usually occur once, seldom twice, each season. Severe natural infection occurred primarily on young leaves: 41 and 42% of Malus- and 33 and 27% of Crataegus-infected leaves were 4-10 and 11-17 days old, respectively. Hawthorn leaves over 24 days old remained susceptible while crabapple leaves 18 days old seldom exhibited lesions. An average of 5.4 adjacent leaves on Crataegus and 2.9 on Malus were severely infected at each inoculation. Due to leaf maturity, application of fungicides later than June 15 to control Gymnosporangium rusts on woody ornamentals is seldom warranted in Illinois.

A748

THE EXTENT OF NORTHERN CORN LEAF BLIGHT AND HELMINTHOSPORIUM TURCICUM RACE-TYPES IN PENNSYLVANIA. W. Bair and J. E. Ayers, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

A survey was conducted during August and September, 1983 in the corn growing regions of Pennsylvania to determine the incidence and severity of northern corn leaf blight (NCLB) in the state. An additional objective was to determine the frequency of race-types (1 or 2) of the pathogen, H. turcicum. NCLB was detected only in the central, eastern, and southern portions of the state; 52 of 103 fields inspected contained NCLB. Disease severity was generally low, and the effects on yield probably were minimal due to the drought in July and August. Disease severity varied on a field-to-field basis rather than on a regional basis. All 52 of the H. turcicum isolates obtained were race 2. Isolates of H. turcicum race 1 may not have been found because a large portion of the corn hybrids grown in Pennsylvania contain the Ht gene.

A749

EFFECT OF SOME FUNGI ON SEED QUALITY OF SOYBEAN. I. J. Gupta and A. F. Schmitthenner, Dept. of Plant Pathology, The Ohio State University, Columbus, OH 43210 and the Ohio Agricultural Res. & Devel. Center, Wooster, OH 44691.

Soybean cultivar Wells was inoculated with Diaporthe phaseoli var. caulivora (Dpc), and species of Phomopsis (Pho), Fusarium (Fu), Alternaria (Alt), Aspergillus (Asp), Chaetomium (Cha) or Epicoccum (Epi) by injecting ca 50 ul of spore-mycelial slurry of 10,000 c.f.u./ml into the distal locule of pods. Seeds from proximal locules of inoculated pods were evaluated for infection, germination, sand bench emergence, cold test performance and accelerated aging performance to assess seed quality. The pod injection method was very effective in producing more than 50% seed infection by Fus, Dpc, Pho, Alt and Asp. These fungi significantly reduced seed germination. In accelerated aging tests Fu reduced seed quality most, followed by Alt, Dpc and Pho. In cold tests Pho reduced seed quality most, followed by Dpc, Alt, Fus and Asp. Cha and Epi did not affect seed quality in any test.

A750

PHYTOPHTHORA SPP. IN CHINA. H.H. Ho[1], Y.N. Yu[2], W.Y. Zhuang[2], Z.R. Liang[2], J.Y. Lu[3], and L.Y. Gong[3]. Dept. Biology, State Univ. New York, New Paltz, N.Y. 12561[1], Inst. Microbiol., Academia Sinica, Beijing, China[2] and Dept. Plant Protection, Nanjing Agric. College, Nanjing, China[3].

Isolates from diseased plant materials and soil samples in China included homothallic species of Phytophthora, e.g., P. boehmeriae on cotton, P. cactorum on apple and P. citricola on citrus, as well as heterothallic species, e.g., P. capsici on pepper, P. cinnamomi on black locust, P. citrophthora on Hevea rubber, P. colocasiae on taro, P. drechsleri on cucurbits, P. infestans on potato, "P. melonis" on cucumber, P. nicotianae on tobacco, P. palmivora on Allium tuberosum, "P. palmivora" MP4 on black pepper and "P. sinensis" on cucumber. P. nicotianae was the most abundant species with the widest host range. Of note is the presence of A1, A2 and homothallic strains of P. cinnamomi in Nanjing area.

A751

THE INFLUENCE OF MATURITY OF 'GOLDEN DELICIOUS' APPLES ON THE EFFECT OF POSTHARVEST CALCIUM TREATMENT ON DECAY. W. S. Conway and C. E. Sams, USDA, ARS, HSI, Beltsville, MD 20705

Apples were harvested from the same block of trees at three harvest intervals to obtain fruits of three different maturities. Fruits were infiltrated at harvest with solutions of $CaCl_2$. After 4 months storage (OC), the three lots of fruits were inoculated with Penicillium expansum. Fruits from the first harvest period infiltrated with an 8% $CaCl_2$ solution had twice as much calcium in the flesh as nontreated fruits, 25% less decay, and little injury. Fruits from the second harvest period or prime harvest, after being infiltrated with an 8% $CaCl_2$ solution had five times the flesh calcium concentration of nontreated fruits, 57% less decay, and exhibited surface injury. Infiltration of an 8% $CaCl_2$ solution into the lot of fruits from the third harvest resulted in a sevenfold increase in the flesh calcium concentration over the nontreated fruits, 67% less decay, but exhibited injury which extended into the cortex. Maturity of apples at time of treatment can have a significant effect on calcium uptake, which in turn affects the level of decay control

A752

VARIATIONS IN PATHOGENICITY WITHIN SORGHUM ISOLATES OF COLLETOTRICHUM GRAMINICOLA. M.E.K. Ali and H.L. Warren, Dept. of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907

Nine sorghum isolates of Colletotrichum graminicola from different states where sorghum is grown were tested for pathogenicity on 6 sorghum lines under field conditions. These sorghum lines were reported to have varying levels of resistance to an isolate of C. graminicola from Indiana. Inoculations were made with a pressurized sprayer. A 5 ml conidial suspension ($5x10^6$ or $3x10^6$ condia/ml) was sprayed into the plant whorl 55 and 62 days after planting, respectively. The plants were rated for disease severity, using a 1-5 scale index, 90, 108 and 115 days after planting. The differential responses exhibited by the sorghum lines 954130, IS 8361 and 954062 to the isolates, made it possible to separate the 9 isolates into 5 physiological races.

A753

DIFFERENTIATION OF ENDOTHIA AND CRYPHONECTRIA SPECIES BY POLY-ACRYLAMIDE GEL ELECTROPHORESIS. J. A. Micales and R. J.

Stipes. Department of Plant Pathology, Physiology and Weed Science, Virginia Polytechnic Institute and State University, Blacksburg, VA 24061.

Chemotaxonomic studies of Endothia and putative relatives were conducted. Soluble protein extracts of 60 isolates, representing 12 species of Endothia and Cryphonectria, were compared by polyacrylamide gel electrophoresis. The banding patterns were highly species specific except among non-vouchered isolates of E. radicalis (= C. radicalis) which were extremely variable; cultural characteristics of these isolates were also diverse. The protein profiles of isolates of E. eugeniae (= C. eugeniae) closely resembled those of C. cubensis and would suggest the combination of these species. Isolate CBS 505.63, previously identified as E. havanensis (= C. havanensis), produced banding patterns similar to those of the E. eugeniae - C. cubensis group. These findings reveal that disparities may exist between stock culture listings and vouchered isolates.

A754

METAMORPHOSIS OF PERONOSPORA HYOSCYAMI F.SP. TABACINA PROTOPLASM. W.E. McKeen, A.M. Svircev and J.W. Berry. Plant Sciences Dept., University of Western Ontario, London, Canada, N6A 5B7.

Two distinct types of protoplasm occur in P. hyoscyami f.sp. tabacina. Actively growing protoplasm is in one state and inactive protoplasm in the other state. The inactive protoplasm occurs in the hyphal "knot", sporangiophore, sporangium and oospore, while the active protoplasm is present in the germinating spore, germ tube, intercellular mycelium and haustoria. The inactive protoplasm is very dense, contains much fine ground plasma, and organelles are camouflaged. The active protoplasm has large electron-transparent spaces and contains well-defined organelles. The ultrastructure and probably the molecular structure is different in each state.

A755

POPULATION DYNAMICS OF FUNGI IN PEACH GUMMOSIS CANKERS. K. O. Britton and F. F. Hendrix, Dept. of Plant Pathology, Univ. of Georgia, Athens, GA 30602.

Peach twigs naturally infected with the gummosis fungi Botryosphaeria dothidea, B. obtusa, and B. rhodina were sampled at monthly intervals for 18 mos. Series of 1-inch segments distal to each of 20 cankers were plated to ascertain patterns of twig colonization. Most twigs contained all three species in apparently random patterns for each twig. There were, however, seasonal fluctuations in the percentage of twig segments yielding Botryosphaeria, as well as species shifts within this population. Botryosphaeria levels peak early in summer, when all three species are present. At this time, B. dothidea is dominant. Populations of B. rhodina also increase in summer. Botryosphaeria colonization is lowest in the fall. Increases from Jan to Apr are due to the presence of B. obtusa, which represents 90% of the population during these months. Populations of B. obtusa never completely disappear from the twigs, but those of B. dothidea and B. rhodina apparently do so in Jan.

A756

USING DRY INOCULUM IN THE FIELD FOR TESTING BEANS FOR RESISTANCE TO ANGULAR LEAF SPOT. **D. A. Inglis, D. J. Hagedorn, and R. E. Rand, Department of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706.**

Inoculum of Isariopsis griseola (IG) for field screening generally consists of conidia in liquid suspension. However, advantages for using dry inoculum include ease of transport, advanced preparation and long-term storage. To improve techniques for using dry inoculum of IG, infected bean leaves were dried at 26-28 C, pulverized in a Wiley Mill, sized in a No. 18 mesh sieve to ≤1 mm and stored at 5-10 C. Dry inoculum at $3.0x10^6$ & $4.0x10^7$ conidia/g was dusted onto plants in the field following irrigation and application of a fungicide sticker. Data collected at three dates showed that treatments with dry inoculum had significantly higher diseased leaf ratings, greater defoliation, and greater yield reduction when compared to a treatment inoculated with a suspension of conidia. Dry inoculum may have greater versatility in the field when suboptimal environmental conditions for disease prevail.

A757

PATHOGENIC CAPACITY OF VERTICILLIUM ALBO-ATRUM PROTOPLASTS. K. C. Blits and A. L. Morehart, Dept. of Plant Science, Univ. of Delaware, Newark, DE 19717-1303.

Verticillium albo-atrum protoplasts were obtained by incubating 24-hour-old hyphae with lytic enzymes isolated from soil microorganisms. Cell wall lysis was complete when protoplasts exhibited no fluorescence in Calcofluor White ST solution (.1% v/v). Greenhouse-grown seedlings of 3 resistant and 2 susceptible tomato varieties were inoculated by root dipping for 10 min. in protoplasts or conidia, with sterile water and protoplast osmoticum (0.7M $MgSO_4 \cdot 7H_2O$ in phosphate buffer) as controls. Leaf chlorosis and wilting occurred 6 days after inoculation with protoplasts only on susceptible varieties. In vivo lysis by plants of invading Verticillium mycelium has been associated with disease remission, yet typical vascular wilt symptoms were observed for both protoplast and conidia treatments during a 4-week period, and Verticillium was consistently recovered from stem and leaf tissue plated on Czapek-Dox medium. Protoplasts subcultured on filtered xylem sap extracted from yellow-poplar formed an extensive mycelium within 10 days.

A758

NEGATIVE GEOTROPISM IN VENTURIA INAEQUALIS. David M. Gadoury and William E. MacHardy, Department of Botany and Plant Pathology, University of New Hampshire, Durham 03824.

Scabbed apple leaves were overwintered in an orchard with either the adaxial or abaxial surface exposed. In mid-February, prior to ascus formation, leaves from each group were collected and incubated in the dark at 10 C and 90% RH for 30-40 days with the adaxial surface facing up, the adaxial surface facing down, or the leaves were inverted at daily or weekly intervals. Leaf disks were then embedded, sectioned, and examined microscopically. The neck and ostiole of the ascocarps was always directed towards the surface that had faced upward during winter, except when the leaves were inverted initially upon incubation or at daily or weekly intervals. Ascocarps in the inverted leaves frequently developed both adaxial and abaxial ostioles as observed by James et al (Mycologia 73:564-566). This response is a result of negative geotropy and may provide a mechanism for conservation of inoculum in disturbed leaf litter.

A759

SOAKING AS A METHOD OF PREPARING SAMPLES FOR AN ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA) FOR Bipolaris sorokiniana. R.H. Vargo and J.S. Baumer. Dept. of Plant Pathology, Univ. of Minnesota, St. Paul 55108.

In an indirect ELISA developed with mycelium of B. sorokiniana, infected spring wheat leaves provided little or no response over that of healthy leaves when ground in 0.05 M carbonate buffer, pH 9.6. Good responses were obtained if individual intact leaves were soaked for 24 hr in 1 ml 0.1 M acetate buffer with 0.02% NaN_3 (w/v). Each leaf was rolled and inserted into a 1.5 ml polypropylene centrifuge tube with lid. The leaf was removed along with trapped fluid and the remaining solution used in the coating step without disturbing sediment. Bruising or tearing infected leaves reduced the antigenic response. Soak and coat pHs below 6 gave the best results and reduced interference from Alternaria alternata. The reaction time was halved to 90 min by adding 100 mg $MgCl_2 \cdot 6H_2O$/L substrate buffer. The soak method reduced labor, worked with spikelets, and was successful in a new ELISA for A. solani on potato.

A760

A GLOMERELLA AND COLLETOTRICHUM SP. FROM SOYBEANS. J. B. Manandhar, G. L. Hartman, and J. B. Sinclair, Dept. of Plant Pathology, Univ. of Illinois, 1102 S. Goodwin, Urbana, IL 61801.

Several Colletotrichum spp. were isolated from field-grown soybean stems in 1983. One isolate formed Glomerella perithecia on potato-dextrose agar with ascospores 13.9 to 19.7 (14.5) x 5.2 to 8.7 (7.3) um and conidia 14.1 to 19.2 (16.3) x 5.1 to 7.0 (6) um. Four Colletotrichum culture types were derived from single ascospores based upon conidial production in an acervulus or pycnidium, presence or absence of setae in acervulus, pigmentation, and pycnidium size in culture. Perithecia formed when two pycnidial types or pycnidial and acervular types were mated, but not when two acervular types were mated. Dark, small pycnidial types generally were self fertile, large pycnidial types and acervular types with setae types being less so. Acervular types without setae were all self sterile.

A761

EARLY INFECTION EVENTS OF Bipolaris oryzae ON WILD RICE. L. M.

Schickli and J. A. Percich. Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Wild rice (Zizania palustris L.) cultivars K2 and Johnson, and bird's nest ferns (Asplenium nidus) were inoculated with Bipolaris oryzae conidia and incubated for 8, 12, 18, 24 and 48 hr at 28-30 C and 96-100% RH. Conidia were evaluated for the number of germ tubes, germ tube length, hyphal branching, appressoria formation, and lesion development. At 8 hr the percent germination of conidia was significantly different between the bird's nest fern and the wild rice cultivars. Seven events were identified on wild rice: germination, germ tube elongation, formation of a mucilaginous sheath, hyphal branching, stomatal or direct penetration, inter-and intracellular hyphal growth, and emergence of hyphae through the cuticle and stomates. Stomatal penetration occurred without appressoria formation. Direct penetrations were more frequent than stomatal penetrations. The bird's nest fern was not infected.

A762

APPLICATION OF SCANNING ELECTRON MICROSCOPY TO PARAFFIN-EMBEDDED PLANT TISSUES TO STUDY INVASIVE PROCESSES OF PLANT-PATHOGENIC FUNGI. D. A. Gaudet and E. G. Kokko, Agriculture Canada Research Station, Lethbridge, Alberta, Canada T1J 4B1.

Scanning electron microscopy (SEM) applied to paraffin-embedded tissue sections is compared with the traditional techniques of light microscopy (LM) and surface SEM for the study of invasion by a plant-pathogenic fungus. SEM of paraffin-embedded sections of wheat leaves infected by Coprinus psychromorbidus consistently yielded high-quality micrographs showing three-dimensional views of both internal and external disease development processes. When the orientation of the specimen in the SEM is manipulated, the specimen can be viewed from different perspectives. The technique is simple and inexpensive and combines the advantage of great depth of focus and high resolution of the SEM with the single preparatory techniques employed for light microscopy.

A763

A NEW CANE CANKER OF THORNLESS BLACKBERRY CAUSED BY BOTRYOSPHAERIA DOTHIDEA. J. L. Maas and F. A. Uecker, ARS-USDA Beltsville Agricultural Research Center, Beltsville, MD 20705

A new and destructive stem (cane) canker disease of thornless blackberry (Rubus spp. cultivars) is described from the eastern United States. The anamorph of Botryosphaeria dothidea is shown to be the causal agent. The fungus pathogen is characterized to distinguish it from other fungi that cause similar-appearing canker diseases. The disease is first evident as reddish-brown lesions at nodes along second-year (fruiting) canes, which enlarge, killing the lateral bud or branch and girdle the cane. Portions of canes above well-developed cankers wilt and die. Most damage is evident during the period of fruit maturation; fruits on affected portions of canes dehydrate and become mummified. An indication of low-level field resistance to the disease has been observed among several cultivars evaluated. Two methods, subjective and objective, were used to quantify field resistance and to compare field resistance evaluation techniques.

A764

SURVIVAL OF THICK-WALLED AND THIN-WALLED OOSPORES AND SPORANGIA OF PYTHIUM ULTIMUM EXPOSED TO SOIL FUNGICIDES. T. E. Stasz and S. P. Martin, College of Agriculture, University of Hawaii at Hilo, Hilo, Hawaii 96720.

Spores were treated with fungicide suspensions for 24 hr and rinsed. Oospores were treated either before or after conversion in nonsterile soil extract from the thick-walled, constitutively dormant condition to the thin-walled, exogenously dormant condition. Sporangia and thin-walled oospores were similarly sensitive to etridiazol, fenaminosulf, captan, thiram, and maneb and exposure to soil drench concentrations greatly reduced their subsequent germination in dilute potato dextrose agar syrup. Thick-walled oospores, however, tolerated fungicide concentrations as high as 10,000 ppm and most treated oospores subsequently became thin-walled and germinated normally. Thick-walled oospores, which can survive in soil for years and which are not detected by soil plating techniques, may not be eradicated effectively by soil drenches with fungicides.

A765

CHEMICAL CONTROL OF SNOW MOLD ON WINTER WHEAT IN ALASKA.

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Sclerotinia borealis, a sclerotial Low Temperature Basidiomycetes (Coprinus spp. ?) and most recently Gerlachia nivalis (Fusarium nivale) are snow molds seriously affecting winter survival of wheat in Alaska. For a period of 3 years from fall of 1980, field trials were conducted at the University of Alaska Experimental Farm to study the efficacy of fungicides in controlling the snow mold complex on winter wheat. Benomyl, dithane M-45 and terraclor were each tested at 3 rates. Five cultivars of early maturing, hard red winter wheat were used in studing the varietal responses to fungicide treatments. These fungicides were evaluated by assessment of the rate of seedling survival and the yield and quality of the seeds produced (1000 kernal weight). Among the fungicides tested, terraclor was consistently superior to others. Significant differences were also noticed in the responses of cultivars to fungicide treatments.

A766

EFFICACY OF PROPICONAZOLE (TILT) AGAINST PHYMATOTHRICHUM OMNIVORUM ON COTTON. J. T. Mathieson, Texas Agricultural Experiment Station, Vernon, TX, 76384, and S. D. Lyda, Dept. of Plant Plathology, Texas A&M University, College Station, TX 77843.

Propiconazole had an ED 50 of 1 ng/ml against Phymatotrichum omnivorum in vitro. The most effective control in greenhouse studies of P. omnivorum was achieved with a 2.5% granular formulation of the fungicide applied at the rate of 2.5 kg a.i./ha. The application resulted in a 59.3% reduction of disease incidence. Cotton grown in field plots treated with propiconazole at rates ranging from 0.1-1.5 kg a.i./ha manifested a 22-66% decrease in disease incidence compared to cotton grown in non-treated plots. The emulsifiable concentrate and granular formulations of propiconazole applied on the seed in the furrow caused severe stunting of cotton seedlings. Stunting was prevented by applying the chemical 5.0 cm from the seed at rates up to 1.25 kg a.i./ha.

A767

CONTROLLING THE RHIZOCTONIA DISEASE OF POTATO WITH SEEDPIECE TREATMENTS. **J.R.Davis and L.H. Sorensen, University of Idaho Research & Extension Center, Aberdeen, ID 83210.**

Results provide evidence that seedborne Rhizoctonia solani is an important source of inoculum in Idaho potato fields. With the control of seedborne R. solani inocula on the 'Butte' cv (by 1.8% a.i. formaldehyde dip for 5 min), the rhizoctonia disease of potato was significantly reduced (P = 0.05) in 9 out of 16 fields. The incidence of R. solani stem lesions on the 'Russet Burbank' (RB) cv was also reduced (P = 0.05) when potato seed was treated with a variety of other fungicides [pentachloronitrobenzene (UniRoyal), DPX965-75 (duPont), Bay NTN 19701 (Mobay), and Thiophanate (Gustafson)]. Although disease control did not significantly influence yield, effects on tuber appearance were evident. Treatments that provided the most effective control on RB cv (either Thiophanate or Bay NTN 19701--applied as a 2.5% a.i. dust) consistently (P = 0.05) reduced the % of malformed potatoes and the incidence of tuber scurf.

A768

EFFECT OF CHEMICALS ON TELIOSPORE GERMINATION OF KARNAL BUNT, NEOVOSSIA INDICA. E. J. Warham and J. M. Prescott, Centro Internacional de Mejoramiento de Maiz y Trigo, Londres 40, Apdo. Postal 6-641, 06600 Mexico, D.F. Mexico.

Thirty-one chemicals (systemics, heavy metals and organic compounds recommended for bunts and smuts) were screened for their efficacy as seed treatments for Neovossia indica. Of these compounds, mercury, pentachloronitrobenzene, and the heavy metals (copper, zinc and manganese) effectively inhibited Karnal bunt teliospore germination. Longevity of the effect of the chemical compounds was also determined by testing the germination of teliospores at 24 hours, 1 week, 1 month, 2 months, 4 months and 6 months after application. To determine whether a seed treatment influenced seed germination, simultaneous seed germination tests were also conducted. Chemicals that showed effective inhibition of Neovossia indica teliospores were retested using a range of application rates.

A769

LEAF-PRINTING: AN EFFICIENT METHOD TO RAPIDLY SCREEN SAMPLES OF CLADOSPORIUM FULVUM FOR TOLERANCE TO FUNGICIDES. V. Miao and V.J. Higgins. Department of Botany, University of Toronto, Toronto, Ontario M5S 1A1.

The leaf printing method is a variation of the replica plating techniques used in bacterial studies. A tomato leaf supporting sporulation of the fungus Cladosporium fulvum is gently pressed into selective media, e.g. rose bengal agar supplemented with 0.25 ug/ml benomyl or 10 ug/ml cycloheximide, such that a mirror image spore print of the leaf remains in the agar; prints can be scored for growth of fungicide tolerant mutants after incubation at room temperature for three days. As one leaflet can be used for serial prints, tolerance to more than one fungicide can be screened from the same diseased tissue, and comparison of replicate prints permits identification of individuals with single or multiple tolerance. This method permits objective assessment of a large sample of spores within a short time.

A770

THE RELATIONSHIP BETWEEN OPP RESIDUES, FRUIT CONDITION, AND DECAY RATES OF CITRUS. Hesh J. Kaplan and Cathleen M. Smithwick, Pennwalt Corporation, Decco Tiltbelt Division, 1713 S. California Ave., Monrovia, California 91016-0120

The postharvest fungicide o-phenylphenol (OPP) has been used in citrus packinghouses as a sanitizer and decay control agent for nearly two decades. The efficacy of the treatment and desired residue levels have been disputed since the antifungal properties and penetration of OPP into the intact rind have not been adequately quantified. Some California shippers have contended that minimum residue levels are mandatory to prevent decay during transit and marketing. This paper presents the results of a series of tests conducted under controlled conditions to quantify the relationships between fruit condition, injury, OPP residues, and the incidence of green mold in lemons and oranges inoculated with Penicillium digitatum Sacc.

A771

EVALUATION OF FUNGICIDES IN GINSENG GARDENS. M. K. Rahimian and J. E. Mitchell. Dept. of Plant Pathology, University of Wisconsin-Madison, Madison, Wi 53706

Evaluation of the effect of fungicide treatments on ginseng is complicated by the fact that plants in the beds are not uniformly distributed. Two experimental gardens each about .08 h were established in Marathon and Richland Counties. Permanent sampling sites, 30 cm^2, were marked in beds so that the same plants could be rated on several dates. Several fungicides were applied with a hand sprayer at a rate of 2.5 l/25 sq m weekly during the growing season to control seedling and leaf blight caused by Alternaria spp. The amount of disease was determined as percentage of infected plants, affected leaf area and seedling survival at each sampling site biweekly. Maneb, mancozeb and iprodione at rates of 1.8, 1.8 and 1.1 kg ai/ha, respectively, reduced the percent of infected plants and percent of leaf area infected in both garden locations. In Richland County, stands were significantly 30% and 27% higher in plots treated with iprodione and mancozeb, respectively, than in nontreated plots. In Marathon County, fungicide treatments did not improve stands.