

ABSTRACTS OF PAPERS

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ABSTRACTS

1

IN SITU OBSERVATION OF VA MYCORRHIZAL INFECTION

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VA mycorrhizal resting spores germinate on water agar and produce a network of exploratory hyphae. Hyphae branch in response to nearby root tips, make contact, form appressoria, and penetrate the root surface. Seedlings of *Nicotiana tabacum*, *Brassica campestris* or *B. napus*, and spores of *Glomus mosseae* or *Gigaspora gigantea* were placed in proximity on slides thinly coated with water agar. Slides were covered with dialysis membrane and incubated on filter paper in 15 cm petri plates under cool white lights. Plates were inclined 30° with a pool of Hepper's nutrient solution in the bottom. Mycorrhizae developed on the slide when hyphae encountered elongating roots, and could be monitored at 12-30x without opening the plates. Slides were removed for high power examination after 2-3 weeks. After living mycorrhizae were observed at 100-400x, the slide was fixed in glutaraldehyde and OsO₄. Excised segments were prepared for SEM or embedded and sectioned. Mycorrhizae stained black with Os and were easily located.

2

SPORULATION OF *GIGASPORA MARGARITA* ON ROOT CULTURES OF TOMATO. M. A. Miller-Wideman and L. S. Watrud. Monsanto Agricultural Products Co., 800 N. Lindbergh, St. Louis, MO 63167.

Use of pre-germinated spores of *Gigaspora margarita* on agar root cultures of tomato routinely resulted in successful mycorrhizal infections on more than 50% of the plates which were inoculated. Penetration into the root, vesicle formation, arbuscule production and new spore formation were reproducibly observed and are described.

3

PHELLINUS ROBINEAE STEM DECAY OF ROBINIA PSEUDOACACIA IN OKLAHOMA PLANTINGS. Jerry W. Riffle, Al Myatt, and Roger Davis. Rocky Mountain Forest and Range Exp. Stn., Forestry Sciences Laboratory, Univ. Nebr., Lincoln, 68583; Oklahoma Forestry Division, Route 1, Box 44, Washington, Okla. 73093; and Oklahoma Forestry Division, State Capitol Building, Oklahoma City, Okla. 73195, respectively.

Incidence of *Phellinus robineae* stem decay of black locust in Oklahoma plantings was determined by examination of 14,172 trees in 123 plantings of six ages in five major land resource areas (MLRA) from April 1981 to January 1982. Infected trees were found in 47% of the plantings. Incidence of infected trees among the five MLRA's ranged from 0 to 7%, and was highest in the Central Rolling Red Plains (MLRA 78). Incidence of infected trees in plantings of age 10, 15, 20, 24, 30, and 40 years was 0.2, 0.7, 3.6, 4.7, 7.1, and 22.2% respectively. From data obtained we estimate that 11% of the living black locust age 20 years and over in Oklahoma plantings are infected with *P. robineae*.

4

INFECTION OF NORTH AMERICAN AND ITALIAN STRAINS OF *ENDOTHIA PARASITICA* WITH MIXTURES OF NORTH AMERICAN AND ITALIAN VIRUS-LIKE CYTOPLASMIC HYPOVIRULENCE AGENTS. J. E. Elliston, The Connecticut Agricultural Experiment Station, 123 Huntington St.,

P. O. Box 1106, New Haven, CT 06504.

Two North American and two Italian normal strains of *Endothia parasitica*, EP-155 and EP-523, and EP-408 and EP-421, respectively, were each infected with each of two North American and two Italian virus-like cytoplasmic hypovirulence (CH) agents, H_{M1} and H_{M2}, and H_{I1} and H_{I2}, respectively. All possible double infections with these agents were then established in each strain by allowing pairs of singly infected forms to interact on agar. This demonstrates that mixed infections can be established easily in this fungus.

5

ABNORMAL CANKERS ON AMERICAN CHESTNUT TREES AND HYPOVIRULENT STRAINS OF *ENDOTHIA PARASITICA* IN MICHIGAN. D. W. Fulbright, W. H. Weidlich and J. H. Hart, Department of Botany & Plant Pathology, Michigan State University, East Lansing, MI 48824

Hypovirulent (H) strains of *Endothia parasitica* were isolated from abnormal cankers in 5 stands of chestnut trees (*Castanea dentata*) in Michigan. Abnormal cankers were also observed on *C. dentata* in at least 3 other locations. Our observations suggest that normal cankers were replaced by abnormal cankers in the first generation of coppice sucker shoots. Most single spore isolates from a severely debilitated H mass isolate (GHU4) and produced large cankers on excised dormant chestnut wood; however, four randomly selected single spore isolates from GHU4 contained double-stranded ribonucleic acid (dsRNA). Growth of GHU4 on PDA with 10 µg cycloheximide/ml and 100 µg chloramphenicol/ml, cured this mass isolate of all detectable dsRNA. Single spores from another H strain (GH2, less debilitated than GHU4) produced cultures that were more debilitated than the parent isolate. Single spore isolates collected from these cultures produced only H isolates.

6

WETWOOD PROPERTIES INHIBITORY TO DECAY FUNGI. J. J. Worrall and J. R. Parmeter, Jr., Dept. Plant Pathology, University of California, Berkeley, CA 94720.

Observations of natural decay columns and *in vitro* growth assays suggest that wetwood of white fir is inhibitory to *Fomes annosus*. Redox potentials and oxygen diffusion rates indicate hypoxic conditions in wetwood, and *F. annosus* and other decay fungi are sensitive *in vitro* to such oxygen availabilities. Although C_x cellulase of *F. annosus* is unaffected by O₂, laccase activity is very oxygen dependent, indicating that wood decay may be even more sensitive to O₂ availability than growth on laboratory media. In addition, low molecular weight organic acids are present in wetwood in concentrations which inhibit or prevent fungal growth. The data indicate that properties of intact wetwood may substantially restrict or prevent colonization and decay by *F. annosus* and that individual trees vary greatly in properties influencing fungal growth.

7

DEVELOPMENT OF ROOT GALLS ON *PHAEOLUS SCHWEINITZII* INFECTED DOUGLAS-FIR (*PSEUDOTSUGA MENZIESII*). S. H. Dubreuil and N. E. Martin. USDA Forest Service, Forest Pest Management, P. O. Box 7669, Missoula, MT 59807 and Intermountain Forest and Range Experiment Station, 1221 S. Main St., Moscow, ID 83843.

Development of galls in *Phaeolus schweinitzii*-infected roots was studied in 1-2 year old and in older roots. Infections originating in adventitious root tips or tips of 1-2 year

old roots caused cambium disruption. This damage resulted in numerous inactive branch primordia (adventitious buds) on 1-2 year old roots and on root galls of older roots. Cambium damage in older roots resulted in two or three rows of reaction parenchyma followed by proliferation of distorted tracheids and disorganized, multiseriate rays. Hyphae from lateral root infections did not extend into cambium from root heartwood and no swelling resulted. The irregular form of gall xylem may result in decreasing conduction efficiency and eventual death of root portions acropetal to galls.

8

ATYPICAL HYPHAE IN RESISTANT SLASH PINES INOCULATED WITH *CRONARTIUM QUERCUM* F. SP. *FUSIFORME*. C. H. Walkinshaw Southern For. Expt. Stn., Box 2008 GMF, Gulfport, MS 39503 and F. F. Jewell, Sr., Louisiana Tech. Univ., Ruston, LA 71272.

Hyphae of *Cronartium quercum* f. sp. *fusiforme* were observed in cambial and cortical tissues of resistant slash pines (*Pinus elliotii* var. *elliottii*) which were inoculated in the greenhouse. Initial growth of hyphae in resistant pines was rapid for 9-14 days. Degradation of these rust hyphae was evident at 21 days after inoculation and nearly complete by 60 days. Large variations in numbers of haustoria were found for different slash pine families. Haustoria were not observed in some pines. Nuclei and cytoplasm of host cells near degenerating hyphae stained intensely suggesting an accelerated metabolism. Such areas appeared necrotic by 60 days after inoculation. Other observations on inoculated shortleaf pines (*P. echinata*) showed similar but more rapid hyphal degradation. This suggests that slash and shortleaf pines have one type of resistance in common.

9

OCCURRENCE, SYMPTOMS AND INTERACTIONS OF *PHAEOLUS SCHWEINITZII* AND ASSOCIATED FUNGI CAUSING DECAY AND MORTALITY OF CONIFERS. S. H. Dubreuil and N. E. Martin. USDA Forest Service, Forest Pest Management, P. O. Box 7669, Missoula, MT 59807 and Intermountain Forest and Range Experiment Station, 1221 S. Main St., Moscow, ID 83843.

A root map of 23 trees ranging from 18 to 120 years in age is presented. *Phaeolus schweinitzii* infections originating in root tips were extensive in Douglas-fir. Grand fir was less affected and ponderosa pine, western larch, western hemlock and western white pine were scarcely affected. *Armillaria mellea* was frequently found in *P. schweinitzii*-induced galls on Douglas-fir in which acropetal root tissue had died leaving open wounds. Adventitious roots were produced from root gall tissue of Douglas-fir. Infected 1-2 year-old roots had numerous adventitious buds which may be associated with gall formation. Green stain in stem heartwood caused by an unidentified fungus with green hyphae was always associated with resin-soaking or decay from *P. schweinitzii* infections.

10

USING HIGH PERFORMANCE LIQUID CHROMATOGRAPHY FOR ANALYZING PLANT GROWTH SUBSTANCES IN BLACK SPRUCE INFECTED WITH *ARCEUTHOBium PUSILLUM*. W.H. Livingston and M.L. Brenner, Department of Plant Pathology and Department of Horticulture and Landscape Architecture, University of Minnesota, St. Paul, MN 55108.

Plant growth substances can be purified from black spruce tissue, healthy or infected with *Arceuthobium pusillum*, using reverse phase high performance liquid chromatography (HPLC). The first system consisted of two preparative columns, PRP-1 (a porous polystyrene gel) and C₁₈. The mobile phase for the first column was a pH/ethanol gradient and for the second column was an ethanol gradient. Three fractions were collected from this system: zeatin and zeatin riboside, IAA, ABA. The ABA fraction was methylated and subsequently quantified using gas chromatography with an electron-capture detector. The other two fractions were further purified using HPLC with a 7 μ C₁₈ column and acetonitrile mobile phase. Analytical HPLC consisting of an anion exchange column with fluorescence and electrochemical detectors was used to quantify IAA. Zeatin and zeatin riboside were quantified using a cation exchange column and UV detectors.

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HISTOPATHOLOGY OF VERTICILLADIELLA WAGENERII IN DOUGLAS-FIR. P.F. Hessburg and E.M. Hansen. Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, Oregon 97331.

Verticilladiella wagnerii causes a lethal root disease in Douglas-fir and in other members of the Pinaceae. Like related sap-stain fungi, *V. wagnerii* imparts a dark stain to infected Douglas-fir sapwood, the result of amber-colored hyphal walls, an ensheathing hyphal sheath, and discolored host cells adjacent to hyphae. Sapwood

colonization was effected through a serpentine to helicoid movement of multi-branched hyphae in the lumens of axial tracheids. Most radial and circumferential growth between axial tracheids was accomplished through bordered pit-pairs on their radial walls. Limited radial growth along ray tracheids gave rise to the crescent-shaped pattern of staining. Observations of prepared specimens from naturally and artificially infected trees using phase-contrast microscopy revealed no instance of parenchyma cell colonization in the xylem nor in any cell type of the secondary phloem. In both insect and non-insect-mediated inter-tree transmission of *V. wagnerii*, wounds appear to be prerequisite to entrance and exit of inoculum through bark and vascular cambium of roots.

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A COMPARATIVE STUDY OF DECAY AND CANKER FORMATION BY *Phellinus PINI* ON WHITE AND BALSAM FIR. Robert A. Blanchette, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Phellinus pini var. *cancriformans*, the cause of extensive cankers on *Abies concolor* in Oregon and California, and *P. pini* var. *pini*, the cause of small cankers on *A. balsamea* in Minnesota, initiated a nonspecific host response in phloem and xylem. Phloem cells occluded around infected areas but the fungus was not confined. Barrier zones in xylem consisted of numerous tangential series of resin ducts and parenchyma cells. These barrier zones, often several within one annual ring, did not effectively compartmentalize the fungus. *P. pini* var. *cancriformans* caused a typical white rot decay of heartwood (removing lignin and cellulose) in white fir; whereas, *P. pini* var. *pini* selectively delignified wood of balsam fir.

13

A LIGHT AND ELECTRON MICROSCOPE STUDY OF THE GROWTH AND DEVELOPMENT OF BLUE STAIN FUNGUS IN SAPWOOD OF LODGEPOLE PINE. R. G. Ballard, M.A. Walsh, W.A. Cole, Biology Department, UMC 45, Utah State University, Logan, Utah 84322.

In Western North America, mountain pine beetle infestations take a tremendous toll of coniferous species, especially lodgepole pine. The beetles carry into the attacked trees a blue stain fungus complex which is responsible for rapid drying of bark and outer sapwood. The primary fungal colonizer, *Leptographium* sp., grows radially via sapwood rays and colonizes this tissue extensively before moving out into the surrounding tracheids. Fungal colonization of tracheids does not appear to be extensive in the early phase of colonization of the sapwood, while disruption of transpiration stream is total. This report describes the growth and development of the fungal pathogens in the rays and tracheids in the sapwood of lodgepole pines.

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SYMPTOMOLOGY OF TREES WITH LEPTOGRAPHIUM PINE WILT ON CAPE COD. Highley, L., K. Rane and T. A. Tattar, Shade Tree Lab, Dept. of Plant Pathology, U. of Mass., Amherst, MA 01003

Leptographium pine wilt appears to be responsible for widespread death in Scots pine (*P. sylvestris*) and Japanese black pine (*P. thunbergii*) on Cape Cod, MA. Japanese black pines attacked by the insect vector, the black turpentine beetle (*Dendroctonus terebrans*), averaged 25 years in age and were growing vigorously. Number of aboveground beetle attacks were few and blue stain in the xylem was confined to the lower bole. Scots pines averaged 50 years in age and exhibited small incremental growth and thinning crowns. The vector attacked these trees in large numbers and the blue stain was more extensive. Presence of secondary insects, engraver beetles (*Scolytidae*) and pine sawyers (*Cerambycidae*) was observed in Scots pine. Resin flow of unattacked Scots pines was heavy. Attacked trees with foliar symptoms exhibited very little resin flow. Inoculation of stressed Scots pines with *Leptographium* sp. in the absence of the vector resulted in death. The fungus was readily recovered from these trees.

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EFFECTS OF BACTERIA ISOLATED FROM CHLOROTIC WHITE SPRUCE ON ROOT GROWTH OF BLACK AND WHITE SPRUCE SEEDLINGS AND CUTTINGS. S. Navratil and E. Mireku. School of Forestry, Lakehead Univ., Thunder Bay, Ontario P7B 5E1.

Bacteria of the genera *Enterobacter*, *Serratia*, *Pseudomonas* and *Pasteurella* (?) were isolated from the fine roots, bark and xylem of large roots and stems, foliage and twigs of chlorotic 10 to 12-year-old white spruce (*Picea glauca*) from Northern Ontario. Bacterial concentrations varied from 5.5×10^2 to 6.0×10^5 cells per gram of dry weight tissue.

Pathogenicity was tested by dipping young black (*P. mariana*) and white spruce germinants and green cuttings into suspensions of bacteria. Several isolates, singly or in combination, reduced hypocotyl elongation and root growth in the black, but not in the white spruce seedlings. Inocula applied to green cuttings of both species reduced rooting frequency and the number and length of developing roots. These effects were predominantly associated with the *Enterobacter* spp. isolates, part. *E. agglomerans*, and one unidentified isolate.

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PINUS ELLIOTTII AND PINUS TAEDA SAWTIMBER VOLUME LOSS CAUSED BY CRONARTIUM QUERCUM F. SP. FUSIFORME. R. S. Webb, School of Forest Resources and Conservation, University of Florida, Gainesville, FL 32611.

Fusiform rust, caused by *Cronartium quercum* Miyabe ex Shirai f. sp. *fusiforme* Burds. et Snow, causes serious economic loss annually among slash (*Pinus elliottii* Engelm.) and loblolly (*P. taeda* L.) pines in the southeastern U.S. Losses among colonized sawtimber-sized loblolly and slash pines are due primarily to degradation of product value from stem colonization. To quantify sawtimber volume losses, over 450 loblolly and slash pines exhibiting stem symptoms of fusiform rust were analyzed using a Barr-Stroud dendrometer. Volumes of symptomatic and asymptomatic stem portions were computed using the STX procedure for trees in 1-in dbh classes from 10- to 18+ in. Volume losses were similar among loblolly and slash pine sawtimber and decreased proportionately as diameter increased. The most critical economic loss occurred where 50% of the stem was girdled in the first 8 ft of the butt log.

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BASIDIOMYCETES ASSOCIATED WITH DECAY AND DISCOLORATION OF BLACK WALNUT. John H. Hart, Terry L. Kamps, Barbara J. Dyko. Depts. of Botany & Plant Pathology and Forestry, Mich. State Univ., E. Lansing, MI 48824 and Karen K. Nakasone, Forest Products Lab, US Forest Service, Madison, WI 53705.

Isolations were made from trunk defects (primarily behind branch stubs) in *Juglans nigra* trees collected in Michigan, Wisconsin and Kansas. Wood chips were removed from various zones around the defects and placed on 2% malt agar with or without benomyl. *Schizophyllum commune* and *Hypochnicium vellereum* were the basidiomycetes most frequently recovered. Both fungi were associated with wounds, were isolated from sapwood and heartwood, and were associated with discolored but not visibly decayed wood. Other hymenomycetes identified and their niche were *Coriolus versicolor* (dead branch stubs), *Phellinus gilvus* (dead sapwood), *Peniophora cinerea* (dead sapwood), *Hericium coralloides* (decayed heartwood) and an unknown basidiomycete (advanced white rot). *Hyphodontia sambuci*, *Coprinus micaceus* and *Cylindrobasidium albulum* were each isolated once as well as various non-hymenomycetes and bacteria.

18
FOREST DISEASE SURVEY IN DELAWARE. J. C. Adams, A. L. Morehart and A. E. Levine, Dept. of Plant Science, Univ. of Delaware, Newark, DE 19711.

The incidence and severity of major fungal and nematode pathogens of commercial forest trees in Delaware were evaluated from 1979 to 1982. This survey was the first of its kind in Delaware. Soil-borne nematodes and soil fertility were not major factors in diseases of forest trees. *Nectria galligena* caused the greatest damage on hardwoods, infecting 10% of the standing timber growing in plots. The predominant fungal pathogens on coniferous species were *Cronartium quercum* f. sp. *quercum* and *Heterobasidion annosum*. Incidence of the latter pathogen was as high as 88% in thinned stands of *Pinus taeda*.

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FIELD AND GREENHOUSE DISEASES OF PROSOPIS (MESQUITE) M.S. Lesney and P. Felker. Caesar Kleberg Wildlife Research Institute, Texas A&I University, Kingsville, TX 78363.

Canker and tip dieback diseases have been found in field and greenhouse plantings of *Prosopis* being examined for energy production in South Texas. Several fungal species were associated with the field cankers, most notably a *Botryodiplodia* and a *Pestalotia*. In the greenhouse, a tip dieback as well as a canker disease that began at pruning cuts on the stock plants was seen. *Pestalotia* was most commonly isolated from these plants. Isolated cultures of the *Botryodiplodia* and the *Pestalotia*, together with several other often associated fungi, (including *Alternaria* and *Fusarium*) were tested for pathogenicity on young mesquite plants. Only *Botryodiplodia* and *Pestalotia* produced cankers under the conditions tested when

spores were applied using wound inoculation techniques. *Pestalotia* required high humidity and proved much less damaging compared to *Botryodiplodia*, which closely mimicked field symptomatology.

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RELATIONSHIP BETWEEN MELANOSE INCIDENCE AND DEAD WOOD IN TEXAS GRAPEFRUIT. R.M. Davis, Texas A&I University Citrus Center, Weslaco, TX 78596

The relationship between the incidence of melanose on fruit and the amount of dead wood in Ruby Red grapefruit trees ranging from 4 to 29 years of age was studied. The amount of dead wood was estimated by measuring sizes and lengths of dead twigs and branches. Highly significant, positive linear correlations were found between: tree age and melanose incidence; tree age and amount of dead wood; and dead wood and melanose incidence. Trees 6 years old and younger did not possess enough dead wood to justify application of protective fungicides. Dead terminal twigs were an important source of inoculum of *Diaporthe citri* since they constituted a significant part of the total amount of dead wood. Large dead branches near the centers of trees were not important sources of inoculum since few fruit were produced in that area. Significantly more dead wood was found in the north and west quadrants of the trees than in the south and east quadrants.

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HISTOPATHOLOGY OF COTTON BOLLS INFECTED WITH COLLETOTRICHUM CAPSICI. R. G. Roberts & J. P. Snow, Dept. Plant Path. & Crop Physiol., La. State Univ. Agric. Expt. Sta., Baton Rouge, LA 70803.

Cotton bolls were inoculated with conidia from a sporulating culture of *Colletotrichum capsici* (Syd.) Butler & Bisby. Bolls up to 20 days-old showed a hypersensitive-type reaction which involved severe plasmolysis, collapse of anticlinal walls and nuclear disintegration in epidermal and subepidermal cells. Bolls 30 days-old or older were susceptible to infection, with rapidly spreading blue-black lesions appearing 3-7 days after inoculation. At 3 days after inoculation subcuticular hyphae were associated with swellings in the epidermal cell walls. Subcuticular hyphae penetrated between epidermal cells by 5 days, followed by rapid intercellular and intracellular growth of hyphae in pericarp tissue. Noninfected cell walls became swollen and difficult to stain. Stromatic development of the fungus was initiated beneath the cuticle and in the swollen remains of the pericarp epidermal cell walls by hyphae which aggregated into a prosenchymatous to pseudoparenchymatous tissue. Sporulation occurred before or after the rupture of the cuticle.

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USE OF FLUORESCENCE MICROSCOPY FOR COMPARISON OF FUSARIUM OXYSPORUM CHLAMYDOSPORE GERMINATION IN CONDUCTIVE AND SUPPRESSIVE SOIL. Fran M. Scher and Ralph Baker, Dept. of Botany and Plant Pathology, Colorado State University, Fort Collins, Colorado, 80523.

Fusarium oxysporum f. sp. *lini* hyphae and conidia were mixed into *Fusarium*-suppressive and -conductive soil. In some treatments, the chelates EDDHA, EDTA, DTPA, or their iron complexes, were added at 100-1000 µg/g soil. The soil was alternately wetted and air-dried to allow chlamydospore formation. Chlamydospores in the soil were stimulated to germinate by addition of 0.1% glucose and asparagine, and, after 16 hr, were stained with a 0.3% aqueous Calcofluor solution. Microscopic observation was made, 3 hr after staining, with 400 nm light supplied by an epifluorescent illuminator. Chlamydospore germination and germ tube elongation were significantly less in suppressive than in conductive soil. Addition of iron chelates had no effect on chlamydospore germination or germ tube elongation in either soil.

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EFFECT OF STERILIZATION PROCEDURE AND TISSUE STORAGE ON RECOVERY OF FUNGI FROM LEAFSPOTS. D. D. Brunk and A. R. Chase, Dept. of Plant Pathology, University of Fla., Gainesville, 32611.

Isolation of the causal organism is often crucial for accurate diagnosis of fungal leafspot diseases. Experiments were performed to test the effect of sterilization procedure and tissue storage on recovery of *Corynespora* from zebra plant, *Cylindrocladium* from leatherleaf fern, and *Exserohilum* and *Phaeotrichoconis* from areca palm. Factorial CRD experiments consisted of 4 sterilant concentrations (0, 0.05, 0.5, 2.5% NaOCl), 3 exposure times (1, 3, 10 min), and two air pressures (ambient, evacuated). Other experiments included three tissue storage conditions: 1) freshly collected; 2) lesions stored 1 wk after excision; and 3) tissue stored 1 wk before lesion excision. With all diseases saprophyte recovery

was decreased and causal organism recovery was either unaffected or increased as sterilant concentration increased. Neither air pressure nor exposure time significantly affected organism recovery. *Cylindrocladium* recovery from leatherleaf fern was lower from excised, stored lesions than from other storage conditions. *Corynespora* recovery from zebra plant was the same for all storage conditions.

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THE USE OF FREEZE-ETCH OR FREEZE-SUBSTITUTION IN THE STUDY OF RUST FUNGAL/HOST INTERACTIONS. D.E. Harder and K. Mendgen. Agriculture Canada, 195 Daffoe Road, Winnipeg, R3T 2M9, Canada; and Universität Konstanz, 7750 Konstanz, Fed. Rep. Germany.

The standardized methods of chemical processing of tissue for electron microscopy may result in considerable structural artifact and incomplete histochemical data. Preliminary studies using ultrarapid cryofixation with subsequent freeze-etching or freeze-substitution were conducted on the bean rust and oat crown rust fungal/host interactions to determine the feasibility of applying these techniques to circumvent some of the above problems. The key objective was to minimize ice crystal damage to the tissue, but at the same time to avoid chemical fixation and cryoprotection with glycerol. By limiting sample size dimensions, freezing as rapidly as possible using liquid propane, and cryoprotection with polyvinylpyrrolidone, marked improvement in the preservation of some structural components was obtained. The extrahaustorial membrane, particularly around young haustoria, is smooth rather than undulating, and the extrahaustorial matrix stains more uniformly and more intensely.

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VARIATIONS AMONG ISOLATES OF *CORTICIUM FUCIFORME* CAUSING RED THREAD DISEASE OF TURFGRASSES. J. V. Cahill, N. R. O'Neill, and P. H. Dernoeden. USDA, Field Crops Laboratory, Beltsville, MD 20705 (1st and 2nd authors) and Agronomy Department, University of Maryland, College Park, MD 20742.

Isolates of *Corticium fuciforme* (Berk) Wakef., were obtained from diseased leaves of five grass species during 1980-81. The fungi fell into two anastomosis groups: Those exhibiting rapid growth at 26 to 31 C, binucleate vegetative cells, and clamp connections, and those exhibiting multinucleate cells and no clamp connections. Typical red thread symptoms were produced after inoculation with eight isolates from the two groups. Physiological and morphological differences between the two groups of fungi indicate that red thread is caused by at least two different fungi. The taxonomy of *Corticium fuciforme*, however, is imperfectly understood. The fungi were tentatively identified as *Athelia fuciformis* (Wakef.), binucleate, and *Laetisaria fuciformis* (McAlp) Burds., multinucleate.

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DETECTION OF *DRECHSLERA GRAMINEA* ON THE BASIS OF DIFFERENCES IN STEROL COMPOSITION OF INFESTED AND NON-INFESTED BARLEY SEED. T. R. Gordon and R. K. Webster, Department of Plant Pathology, University of California, Davis 95616.

A laboratory assay for the presence of *Drechslera graminea* (cause of barley stripe) in seed lots of barley (*Hordeum vulgare*) has been developed. The procedure is based on the ultra violet absorbance spectrum of the sterol fraction extracted from *D. graminea* infested seed. This spectrum is nearly identical to that of purified ergosterol, a common fungal sterol. The spectrum is evident in extracts of seed lots with as little as 1% of the seed infested. A comparable extract from healthy seed does not have an ultra violet absorbance spectrum resembling ergosterol. The method appears promising as a means of rapidly evaluating barley seed lots for infestation by the stripe pathogen.

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THE EFFECT OF PYRAMIDING "DEFEATED" WHEAT POWDERY MILDEW RESISTANCE GENES ON COMPONENTS OF "SLOW MILDEWING." R. R. Nelson, W. L. Pedersen, and D. R. MacKenzie, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802

Four near-isogenic lines, each possessing a different powdery mildew resistance gene, were intercrossed, selfed, and selected to obtain seed that was heterozygous or homozygous for two resistance genes. One four-gene heterozygous pyramid was obtained by crossing two heterozygous two-gene pyramids. When tested for components of "slow-mildewing," one homozygous pyramid, Pm3c x Pm4, significantly reduced the number of lesions per leaf, while another pyramid, Pm4 x MA, reduced sporulation per lesion. All homozygous pyramids were more effective at reducing both lesion

numbers and sporulation than the corresponding heterozygous pyramids. The four-gene heterozygous pyramid was more effective than the two heterozygous two-gene pyramids in reducing lesion number and sporulation.

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EXPRESSION OF LEAF RUST RESISTANCE IN *TRITICUM AESTIVUM* 'ATLAS 66'. L. E. Browder and M. G. Eversmeyer, USDA-ARS, Dept. of Plant Pathology, Kansas State Univ., Manhattan, KS 66506.

The wheat cultivar Atlas 66 is known to have a gene which conditions a specific reaction to *Puccinia recondita* Rob. ex. Desm. The effect of this gene is not expressed in commonly used seedling tests, but is expressed as a "fleck" reaction in field tests of adult plants where avirulent parasite genotypes prevail. We tested plants of Atlas 66 and Wichita in the 2-3 leaf stage and 4-5 leaf stage with 12 *P. recondita* cultures after clipping the primary leaf. Resistance in Atlas 66 to 10 of 12 cultures was expressed as a marked increase in latent period (LP), as compared to Wichita, with little or no difference in final infection type. A slight LP increase in Atlas 66 was observed with the other 2 cultures. The LP difference increased with plant age. Although these data do not unequivocally relate the increased LP effect and the "fleck" reaction at later growth stages and/or other environments, increased LP may be an effect of specific host:parasite interaction and thus not an adequate marker of general resistance.

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HISTOLOGY OF *PUCCINIA RECONDITA* INFECTION ON SLOW- AND FAST-RUSTING WHEAT CULTIVARS. T.S. Lee & G. Shaner, Dept. of Botany & Plant Pathology, Purdue Univ., W. Lafayette, IN 47907

The histology of *P. recondita* infection was investigated to determine if the longer latent period on slow-rusting wheats compared to fast-rusting wheats was the result of slow fungal growth in early stages of infection. Fungal penetration and colony development 24-216 hr after inoculation of flag leaves on fast-rusting (Suwon 92, and Morocco), and slow-rusting (Suwon 85, SW72469-6, L574-1, and CI 13227) wheats were observed with a fluorescence microscope on tissue treated with an optical brightener. Among these wheat cultivars, the frequency of substomatal vesicles was the same. However, colonies were smaller and had fewer haustoria in slow-rusting cultivars than in fast-rusting cultivars. On both fast- and slow-rusting cultivars, urediniospore formation began when there were 175-200 haustoria per colony, at which time colony area was about 0.12 mm². The longer time required for colonies to reach this incipient sporulation stage on slow-rusting cultivars would explain their longer latent period.

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FACTORS CONTRIBUTING TO VARIABILITY IN *SCLEROTIUM ROLFII*. Z. K. Punja and R. G. Grogan, Department of Plant Pathology, University of California, Davis, CA 95616.

Seventy-two isolates of *Sclerotium rolfii* from 19 hosts and 15 areas varied in growth rate, sclerotial production and size, and frequency of clamp formation. Forty-six isolates formed the basidial state (*Athelia rolfii*) on PDA containing 2% activated charcoal. Variation within single-basidiospore progeny (Sb) from each of five field isolates suggested that they were heterokaryotic. Sixty-four percent of the Sb isolates fruited, indicating that these isolates were homothallic. When field isolates were crossed in all combinations, aversion (barrage) zones were noted in 86% of the crosses. This phenomenon was used to establish compatibility (Cm) groups. Between isolates from any two Cm-groups, aversion was associated with killing of hyphae; isolates from the same group did not avert when crossed. Aversion is based on dissimilarity of compatibility factors and may prevent cytoplasmic exchange. Of all possible crosses between Sb isolates, 86% showed aversion; this prevented heterokaryon formation in most but not all cases.

31

AROMATIC NITRILES AND RELATED FLAVOR COMPOUNDS: STIMULATORS OF GERMINATION OF UREDOSPORES OF *PUCCINIA HELIANTHI* AND *UROMYCES VIGNAE*. R.C. French and W.M. Dowler, USDA, ARS, Plant Disease Research Laboratory, P.O. Box 1209, Frederick, Maryland 21701.

Uredospores of sunflower rust and cowpea rust were stimulated by a variety of aldehydes, ketones, alcohols, nitriles, and esters, ranging from volatile oils to non-volatile, water soluble substances. β -Ionone, reported active on other species, was active on these, and was used as a standard for comparing compounds. With *P. helianthi*, 16 of the 25 active aromatic compounds were nitriles. Benzonitrile was very effective and stimulated uredospore germination in pustules of diseased plants in

dew chambers. 3-Cyanophenol (water soluble), could be sprayed on rusted plants to induce the same response. With *U. vignae*, 5 of the 7 active aromatic compounds were nitriles. Mixtures of 4-CN-benzaldehyde and methyl 3,5 dihydroxybenzoate were synergistic and induced some germination in pustules of rusted cowpea plants in dew chambers. Some aromatic nitriles are effective in stimulating uredospore germination, and may be useful for inducing precocious germination.

32

FUNGAL GROWTH AND SPORULATION INHIBITION BY SOYBEAN SEED EXUDATES. Wayne Gade, Jean Gade and B.W. Kennedy, Department of Plant Pathology, University of Minnesota, and College of St. Catherine's, Chemistry Department, St. Paul, MN 55108.

Dialyzed samples of soybean seed (*Glycine max*) exudates and purified soybean lectins were spotted on cultures of *Aspergillus amstelodami*, *A. repens*, *A. chevaleri*, *A. restrictus* and *A. niger* and *Fusarium solani* grown in petri dishes. Growth and sporulation of the aspergilli were inhibited, especially when cultures were grown under conditions that promoted abundant sporulation. There was less inhibition under conditions that favored rapid growth and sporulation, such as darkness and malt-yeast-extract + sucrose media. Binding of purified soybean lectin to specific *Aspergillus* structures were demonstrated by using fluorescent-labeled lectin. We conclude that there is an association between sporulation and inhibition by lectins and exudates.

33

β -GALACTOFURANOSIDASE ACTIVITY OF HELMINTHOSPORIUM SPECIES CONVERTS H. SACCHARI TOXIN TO TOXOIDS. R. S. Livingston and R. P. Scheffer, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824

H. sacchari produces a host-selective toxin and several related compounds (toxoids or "noxins") which can counteract the effects of toxin on sugarcane tissue. In culture, toxin concentration peaked at 3 weeks, followed by declining titers for the next 3 weeks. Toxoid concentration increased as the toxin titer fell, suggesting that toxin may be converted into toxoids by removal of galactose units. An enzyme which cleaves the β , 1-5-linked galactofuranose units in the toxin molecule was isolated from cultures; the enzyme converts toxin to the toxoids. *H. turcicum*, *H. carbonum*, *H. victoriae*, and *H. maydis* each produced much greater β -galactofuranosidase activity than did *H. sacchari*. Low production by *H. sacchari* suggests a possible role of the enzyme in accumulation of toxin. *Fusarium oxysporum* and *Cladosporium cucumerinum* produced little or no β -galactofuranosidase activity.

34

CYTOPLASMIC RESPONSES TO FUNGAL ATTACK: A FIBRILLAR SYSTEM RELATED TO ORGANELLE TRANSPORT. *M.G. Smart, *N. S. Allen, *J. R. Aist, and *H. W. Israel, *Department of Plant Pathology, Cornell University, Ithaca, NY 14853, and *Department of Biological Sciences, Dartmouth College, Hanover, NH 03755.

The cytoplasmic aggregate response results from a redirection and localized disruption of organelle transport. Since transport in other plant cells is related to a system of F-actin fibrils, we tried to visualize such fibrils in barley coleoptile cells using the AVEC microscopic technique [Allen Video-Enhanced Contrast; Allen, R. D. and N. S. Allen. 1982. J. Micros. (in press)]. Cells of the inner epidermis had a system of relatively thick fibrils along which organelles were transported rapidly for distances of more than 250 μ m. These cables appeared taut, exhibited a slow, lateral drift, and were weakly birefringent. There was also a network of finer, rapidly oscillating, flexible fibrils, along which organelles were transported over shorter distances. A summary video demonstration will be presented.

35

TRANSGRESSIVE SEGREGATION FOR RESISTANCE TO COMMON ROOT ROT IN DURUM WHEAT. R. W. Stack, Plant Pathology Department, North Dakota State University, Fargo, ND 58105

Two durum cultivars, 'Wakooma' and 'Edmore' of very different parentage, have shown better than average resistance to common root rot caused by *Helminthosporium sativum* in extensive field trials. Using these cultivars as parents, hybrid seed was produced in the greenhouse. We tested the root rot response of the F_1 plants and of 268 F_2 -derived lines in randomized, replicated field experiments. The root rot response of the F_1 plants was the same as the more resistant parent (Wakooma). The distribution of the 268 F_2 lines was not normal ($P = .01$) and skewed to-

ward susceptibility. The mean of the F_2 population was significantly less resistant than the mean of the parent lines ($P = .0001$). There were, however, 32 F_2 lines which were more resistant than either parent. Eleven of these showed gains in resistance of 10-28% over either parent which indicated that transgressive segregation for resistance had occurred.

36

INDUCTION AND ISOLATION OF AUXOTROPHIC AND DRUG-RESISTANT MUTANTS OF THE COMMON WHEAT-BUNT FUNGUS, *TILLETIA CARIES*. A.C.L. Churchill and D. Mills, Dept. of Botany and Plant Pathology, Oregon State Univ., Corvallis, OR 97331.

Genetic studies of *Tilletia caries* have been hampered by the lack of genetic markers. Protocols for the induction of auxotrophic and drug-resistance mutations from secondary sporidia of *T. caries* were developed. Log-phase sporidia of two monokaryotic, compatible strains of race T-1 were mutagenized with nitrosoguanidine. Two "non-leaky" nutritional mutants were isolated and characterized; one required adenine, the other uracil. Five "leaky" nutritional mutants were also isolated. One was identified as a glycine auxotroph, and the other four had mutations in pathways for general nitrogen metabolism. A preliminary mating of compatible, "non-leaky" auxotrophic strains on minimal medium produced mycelial growth presumed to be the result of complementation. Cycloheximide-resistant strains were also isolated following the selection of spontaneous and ethyl methanesulfonate-induced mutations.

37

INHERITANCE OF RESISTANCE TO RACE 5 OF *CERCOSPORA SOJINA* IN TWO CULTIVARS OF SOYBEAN. D. V. Phillips and H. R. Boerma, Dept. of Plant Pathology, Georgia Experiment Station, Experiment, GA 30212 and Dept. of Agronomy, University of Georgia, Athens, GA 30602.

The soybean cultivars Davis and Lincoln were resistant to race 5 of *Cercospora soja* Hara, but differed in reaction to inoculation. Davis often developed very small (less than 1 mm) lesions or flecks while Lincoln seldom developed any lesions. In contrast, the susceptible cultivars Blackhawk and Hood developed numerous, large spreading lesions. Crosses were made among these four cultivars and the progeny from the F_1 , F_2 , and F_3 generations were inoculated with race 5 in a greenhouse. The results demonstrated that resistance in Davis was conditioned by a single dominant gene and that resistance in Lincoln was conditioned by another dominant gene at a different locus. There was no evidence of cytoplasmic or maternal effects on the expression of resistance in either cultivar. The genes in Lincoln and Davis can be used to develop cultivars with two different sources of resistance to race 5 of *C. soja*.

38

TRANSFER OF CYTOPLASMIC DETERMINANTS BETWEEN VEGETATIVE COMPATIBILITY GROUPS IN *ENDOTHIA PARASITICA*. Sandra L. Anagnos-takis, CT Agricultural Experiment Station, Box 1106, New Haven, CT 06504.

Endothia parasitica, the chestnut blight pathogen, has a heterogenic system of vegetative compatibility (v-c) that can restrict transfer of cytoplasmic determinants. When inoculated close together (≤ 2 mm) on Difco potato dextrose agar (PDA), most strains in different v-c groups form barrage zones that are easily seen where their mycelia meet. Determinants for hypovirulence (H), which are cytoplasmic, can sometimes be transferred between strains forming these barrages after several days of contact. Some v-c groups interact weakly, forming only faint barrage lines where their mycelia meet. These line-formers were paired on sterile cellophane over PDA that contained methionine (100 mg/l) and biotin (1 mg/l). One member of each pair contained Italian H determinants (confer-ring "white" morphology). The H determinants were rapidly transferred between strains that only form faint barrage lines.

39

INSTABILITY OF THE R PLASMID pJB4JI AND TRANSPOSITION OF Tn5 IN *ERWINIA CAROTOVORA* PV. *CAROTOVORA*. R. T. Zink, D. A. Feese, and A. K. Chatterjee, Department of Plant Pathology, Kansas State University, Manhattan, KS 66506.

In crosses between *Escherichia coli* (2942/pJB4JI) and *E. carotovora* pv. *carotovora* (DeBoar strain 71, serotype III; hereafter DB71), kanamycin (Km)-resistant transconjugants of DB71 were obtained at an approximate frequency of 1.6×10^{-4} per input donor cell. In these crosses, Km^r and Gm (gentamycin)-resistant transconjugants were not detected suggesting an instability of the R plasmid and the transposition of Tn5 in the

strain DB71. Km^r clones of DB71 were invariably Gm^S ; a proportion of $Km^r Gm^S$ clones were either auxotrophic (Cys^- , Ile^- , Tyr^- or Ura^-) or defective in catabolism of succinate, sucrose or xylose. The R plasmid (pJB4J1) DNA was detected in lysates of the donor *E. coli* strain but not in Km^r transconjugants of DB71. The $Tn5$ -insertion mutants, like the parent strain, were pectolytic, proteolytic, macerated potato tuber tissue, and were susceptible to bacteriophages infecting the parent, DB71.

40

IN VIVO CONSTRUCTION OF AN F-PRIME PLASMID CARRYING THE DRUG-RESISTANT TRANSPOSOME $Tn5$ AND THE USE OF THE PLASMID IN MOBILIZING THE CHROMOSOME OF *ERWINIA CHRYSANTHEMI*. A. K. Chatterjee, J. S. Ziegler, and K. K. Thurn, Department of Plant Pathology, Kansas State University, Manhattan, Kansas 66506.

Having noted a random chromosomal insertion of $Tn5$ in *E. chrysanthemi* (EC16), we determined the chromosome mobilizing ability of a conjugative plasmid carrying $Tn5$. We first constructed $F'_{ts} lac^{+}::Tn10::Tn5$, by introducing $F'_{ts} lac^{+}::Tn10$ into an *E. chrysanthemi* strain carrying a chromosomal $Tn5$ insertion and by selecting for the cotransfer of lac^{+} , tet^{r} , kan^{r} . Lac^{+} , Tet^{r} , Km^{r} transconjugants were purified and tested for the cotransfer and cosegregation of all three markers. The genetic data indicated that $Tn5$ transposed into the parent plasmid. $F'_{ts} lac^{+}::Tn10::Tn5$ was then introduced into strains carrying chromosomal $Tn5$ insertions. The stable Lac^{+} , Tet^{r} , Km^{r} clones mobilized asn^{+} , gta^{+} , leu^{+} , orn^{+} , pur^{+} , $pyrA^{+}$ and ser^{+} . The frequency of marker transfer varied with the site of chromosomal $Tn5$ insertion in the donor, indicating polarity in the chromosome mobilization.

41

$Tn5$ INDUCED MUTATIONS ALTER EXTRACELLULAR ENZYME PRODUCTION IN *ERWINIA CHRYSANTHEMI*. K. K. Thurn, D. J. Tyrell and A. K. Chatterjee, Department of Plant Pathology, Kansas State University, Manhattan, KS 66506.

We recently observed that $Tn5$ transposed into various sites on the *E. chrysanthemi* (EC16) chromosome causing mutations in biosynthetic and catabolic genes. We subsequently isolated kanamycin-resistant ($Tn5$ -insertion) clones with altered pectate lyase (PL) or protease (PRT) activity. PL-defective mutants, although invariably reduced in both PL and PG (polygalacturonase) activity, were normal with respect to PRT and PLC (phospholipase C). Based upon the distribution of PG and PL and the quantities of OGL (oligogalacturonate lyase, an intracellular enzyme), PL-mutants examined thus far fell into one of three classes: I. transport-defective for PG, normal OGL; II. transport-defective for PL and PG, reduced OGL; and III. normal transport for PG and PL, reduced OGL. $Tn5$ -induced PRT deficient mutants produced normal quantities of other extracellular enzymes. Potato tuber tissue maceration was much reduced only with class II PL-mutants.

42

ISOLATION OF THE PLASMID-ORIGIN OF REPLICATION IN A PLANT PATHOGENIC PSEUDOMONAD. A. R. Poplawsky, L. J. Szabo, and D. Mills. Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR. 97331

Strain LR700 of *Pseudomonas syringae* pv. *phaseolicola* contains a 147 kbp cryptic plasmid, pMC7105. The plasmid has been shown to be integrated into the bacterial chromosome in strain LR719. Occasional excisive recombination has generated 15 excision plasmids ranging in size from 33 to 250 kbp. Restriction endonuclease analysis has revealed that only one region from pMC7105 is common to all the excision plasmids. This 23 kbp region is expected to contain the plasmid origin of replication. Three Bam HI restriction fragments comprising pEX8080, a 34.8 kbp excision plasmid, have been cloned into pBR322 and are being cloned into other vectors which also fail to replicate in pseudomonads. We are attempting to identify the region encompassing the origin of replication of pMC7105 by testing for stable maintenance of these chimeric plasmids after transformation or conjugation into strains of pseudomonads.

43

CONTROL OF STEM RUST OF KENTUCKY BLUEGRASS BY MANCOZEB AND TRIADIMEFON. J.A. Houfek and J.E. Watkins. Dept. of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722.

The protective fungicide mancozeb (Fore® 80W) and systemic fungicide triadimefon (Bayleton® 50WP) were applied to Touchdown Kentucky bluegrass as 7 or 3-day preinoculation or 1 or 5-day postinoculation treatments. Plants were inoculated by dusting

the second leaf with a urediospore/talc mixture. Treated plants were sampled 6, 10, 14, and 18 days postinoculation for comparison of treatment efficacy. Both fungicides adequately controlled stem rust but efficacy was dependent on time of application. Stem rust levels for all Bayleton treatments were significantly lower than the untreated control. Rust severity with Fore was significantly lower than the control at all application dates except 5 days postinoculation. Additionally, percent leaf area infected with Bayleton was consistently lower than with Fore at corresponding application dates for all sampling periods.

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AN IN VITRO TECHNIQUE FOR PROFUSE SPORULATION OF *DRECHSLERA TRITICI-REPENTIS*. P.J. Raymond and W.W. Bockus. Dept. of Plant Pathology, Kansas State Univ., Manhattan, KS 66506

Tan spot, caused by *Pyrenophora trichostoma* was the second most important wheat disease in Kansas for the past 5 yr. Inoculations with the conidial stage *Drechslera tritici-repentis* (Dt) are used to identify sources of resistance. The following sporulation technique was less laborious than the standard procedure, produced higher numbers of conidia, and spore suspensions contained lower amounts of mycelial fragments. Molten ¼ strength PDA was poured into slanted 100-mm petri dishes to form a 70 mm long agar wedge tapering from 8 mm to 0 mm thick. After agar solidification, the dishes were laid flat and 7 ml V-8 agar (15% V-8, 3.0 g $CaCO_3$ and 15 g agar/l) added. A mycelial plug of Dt was placed on the outer edge of the PDA side of the plate and incubated at 21 C under continuous light. When Dt grew 20 mm onto the V-8 agar, the dish was placed at 16 C for 24 hr under a 12:12 light:dark cycle to induce sporulation. Up to 4 daily harvests of conidia were possible from each plate.

45

LEPTOSPHAERIA NODORUM AND *MYCOSPHAERELLA GRAMINICOLA* IN NORTH AMERICA. A. L. Scharen and F. R. Sanderson, USDA, ARS, Plant Pathology Dept., Montana State Univ., Bozeman, MT 59717 and DSIR, Crop Research Division, Christchurch, New Zealand, respectively.

Leptosphaeria nodorum and *Mycosphaerella graminicola* have not been reported in North America. Perithecia of the two genera were found in wheat leaf debris from Hays, Kansas. *Mycosphaerella perithecia* have been reported in wheat and other plant species in Washington State. During 1980-81, a search was made for perithecia in leaves from wheat plants inoculated with *Septoria nodorum* and *S. tritici* at Bozeman, Montana. Perithecia of *L. nodorum* were found in relative abundance in leaf remnants that had remained in the field during the winter. Single ascospore cultures produced pycnidia of *Septoria nodorum*. *Mycosphaerella* ascospores in culture have not produced pycnidia of *S. tritici*. Further studies may reveal the epidemiological importance of ascospores of *L. nodorum* and *M. graminicola* as primary inoculum of glume blotch and speckled leaf blotch in North America.

46

OCCURRENCE OF POWDERY MILDEW (*LEVEILLULA TAURICA*) ON TOMATO TRANSPLANT STOCK. Wayne B. Jones and Sherman V. Thomson, Dept. of Biology, Utah State University, Logan, Utah 84322.

In 1980 and 1981 disease symptoms of *Leveillula taurica* (Lev.) Arn. first developed in Utah tomato fields on tomato transplant stock brought in from the Moapa Valley in southern Nevada; symptoms occurred later in fields planted with locally grown transplants. In 1981 symptomless transplants from Nevada were planted with transplants raised locally and disease development was monitored. Disease symptoms occurred first on Nevada transplants 29 days after planting in Utah and approximately 3 weeks later on the Utah-grown transplants. Plants from Nevada and Utah were equally infected 7 weeks after planting and no differences in severity or yield were detected at the end of the season. Transplants not used in the field experiment were maintained for 4 weeks in a greenhouse. Mildew lesions were noted within 21 days on Nevada transplants while no symptoms were detected on locally grown transplants. Field inspections of tomatoes in Nevada in September 1981 revealed no diseased plants, however, tomatoes in a nearby greenhouse were severely infected.

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Analysis of spore dispersal gradients of *Botrytis cinerea* and gray mold disease gradients in snap beans. K. B. Johnson and M. L. Powelson, Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331

Spore dispersal gradients and blossom and pod disease gradients from point inoculum sources of *Botrytis cinerea* were measured over time in two snap bean field experiments. Laboratory grown inoculum was placed at ground level in a 30 x 30 cm square at bloom initiation and removed at full bloom. Dispersal of

inoculum, assessed by quantifying spores washed from bean foliage, was limited to within 3 m from the inoculum source during bloom. At harvest, the spore populations on plants were 20 to 30 times higher than populations at full bloom suggesting production of secondary inoculum. At two sampling times during bloom, incidence of *B. cinerea* on senescing blossoms averaged 70% at a distance of 0.9 m from the inoculum source but less than 2% at distances greater than 4 m. In one experiment, the incidence of pod rot at harvest averaged 7.2% at 0.9 m from the inoculum source but only 1.3% at a distance of 4.5 m. Spore dispersal gradients analyzed by regression of the form $\log Y = a + b \log X$ showed significant flattening at harvest compared to full bloom, whereas gradients for pod rot incidence at harvest did not flatten when compared to blossom incidence at full bloom. Blossom infections early in the bloom period from local inoculum sources are important in disease development.

48

GERMINATION OF *PERONOSPORA DESTRUCTOR* SPORANGIA AFTER EXPOSURE TO DIFFERENT RELATIVE HUMIDITIES AND TEMPERATURES. E. Bashir and D. E. Aylor, The Connecticut Agricultural Experiment Station, Box 1106, New Haven, Connecticut 06504 USA.

P. destructor sporangia were germinated after exposure to diverse conditions of temperature, RH and UV irradiation. Sporangia, collected from infected greenhouse-grown onion plants, were exposed to 76, 53 and 33% RH at 10, 20, 25 and 35°C. At 10 and 20°C, the sporangia were resistant to dryness and the percentage that germinated was only moderately decreased, even after 72 h of exposure. The percentage that germinated at 35°C was halved after 2 h exposure to either 51 or 75% RH, and was reduced by 90% after only 30 min at 33% RH. After dry, detached sporangia were irradiated with short-wave (254 nm) UV light (410 µW/cm² for 12 min), none germinated. Germination was partially restored by exposure to visible light. Irradiation with long-wave (365 nm) UV light (1100 µW/cm² for 30 min) did not affect germination. The results suggest that dispersed sporangia of *P. destructor* should normally survive the extreme conditions found in the field.

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FURTHER STUDIES ON PATTERN OF VIRUS INFECTION IN STRAWBERRY FRUITING FIELDS OF CENTRAL CALIFORNIA. Annamaria Pisi, Barbara A. Rogers and A. H. Gold, Dept. Plant Pathology, University of California, Berkeley, CA 94720.

In our continuing investigation of the virus spread in commercial strawberry fruiting fields in the Watsonville area of California, we were able to detect infection by only four viruses: Strawberry Mild Yellow Edge, Mottle, Crinkle and Tobacco Streak (Necrotic Shock). Tobacco streak virus occurs at low incidence, spreads slowly from plant to plant and was found only in one commercial variety. The patterns of spread of virus infection in the field plot studies were essentially uniform. Virus movement in most cases requires plants in actual contact and the distance between beds appears to be a barrier rarely breached. Our studies indicated that during the fall months immediately after transplanting, spread of infection was negligible. The highest rate of spread was during the late winter and early spring months. In the late spring and warm summer months spread was not appreciable. Differences in the rate of virus spread in different fields may be attributed to variations in the local environment.

50

DISPERSAL OF UREDINIOSPORES WITHIN A WHEAT CANOPY. M. G. Eversmeyer and C. L. Kramer, USDA, ARS, Dept. of Plant Pathology, and Division of Biology, Kansas State University, Manhattan, Kansas 66506.

Rapid development of a wheat leaf rust epidemic occurs primarily as a result of secondary spread of inoculum. In order to determine the spread of urediniospores from primary foci, rod samplers and artificial leaves were placed at 25 cm, 50 cm, and 1 m heights within an Indar protected wheat canopy. Twenty-six sets of samplers were placed at 1 m intervals in a grid surrounding a 0.6 m² primary infection focus. Lateral spread of spores downwind of the focus was greater when wind movement was perpendicular to the wheat rows and number of spores trapped at each meter away from the focus decreased by 50%. When wind movement was parallel to the wheat rows, lateral spread was less pronounced and number of spores trapped each meter away from the focus decreased by only 30%.

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INTERACTION AMONG GUAYULE, VERTICILLIUM DAHLIAE, AND NON-PATHOGENIC BACTERIA. M.W. Olsen and I.J. Misaghi. Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

The potential of certain non-pathogenic bacteria to promote growth of guayule and to reduce the incidence of Verticillium wilt is being studied. Several isolates of bacteria, collected

from field soils and guayule rhizospheres, promoted growth of greenhouse-grown guayule when introduced into the soil. Percent increase in growth of 12-week-old guayule seedlings ranged from 17 to 56, nine weeks after inoculation. Development of Verticillium wilt was delayed by at least two weeks in plants treated with fluorescent pseudomonads by root dipping two weeks prior to inoculation with *V. dahliae*. However, responses of plants to dual inoculations were not consistent. Studies are underway to determine if the increased growth of plants treated with bacteria helps improve establishment and survival of seedlings transplanted into the field.

52

THE EFFECT OF FOLIAR AND SOIL MAGNESIUM APPLICATION ON BACTERIAL LEAF SPOT OF PEPPERS. J. B. Jones, S. S. Woltz and J. P. Jones. Assistant Plant Pathology, respectively, Univ. of Florida, AREC-Bradenton, Fla. 33508.

'Early Cal Wonder' pepper plants were grown in Myakka fine sand amended with calcium carbonate or dolomite. Plants in five boxes of each liming treatment were sprayed weekly with MgCl₂ (2.4 g/l). Inoculation consisted of infiltration of *Xanthomonas campestris* pv. *vesicatoria* (XV) at 8 x 10³ CFU/ml in leaves (one leaf per plant) in each treatment in early October and later in November the same plants were mist inoculated with XV at 10⁹ CFU/ml. With both inoculation techniques disease was most severe on plants grown in dolomite that were sprayed with Mg. Non sprayed plants grown in dolomite amended soil or Mg sprayed plants grown in CaCO₃ amended soil had the lowest disease severity. Tissue analysis revealed that magnesium levels were positively correlated with disease development.

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ULTRASTRUCTURAL REACTIONS OF DIFFERENT COTTON LINES TO BACTERIA AND STARCH GRAINS. A. Almousawi, P. Richardson, and M. Essenberg. Okla. State Univ., Stillwater, OK 74078; W. Johnson, Langston Univ., Langston, OK 73050 and Okla. State Univ., Stillwater, OK 74078.

The cotton line Im 216, universally resistant to the cotton pathogen *Xanthomonas campestris* pv. *malvacearum* (Xcm), forms envelopes around live Xcm, dead Xcm, and live *X. campestris* pv. *campestris* (Xcc), a heterologous pathogen. The cotton line Ac 44, universally susceptible to Xcm, did not envelop live Xcm, but did envelop Xcm killed by heat, rifamycin or UV light. Ac 44 also enveloped live Xcc, live gram (+) saprophytic *Micrococcus lysodeikticus*, and starch grains. OK 1.2, a line with two genes for resistance, and OK 2.3, with one resistance gene of the three in Im 216, were examined. Both were found to form envelopments with inoculum levels of 5 x 10⁷ cells/ml, the envelopments broke down at 2 days in OK 2.3 and at 3 days in OK 1.2. With 10⁵ cells/ml of inoculum, Im 216 was found to form loose envelopments of less dense fibrillar material and larger colony size than with higher than 10⁸ cells/ml.

54

THE MICROBICIDAL ACTIVITY OF HYDROQUINONE OXIDATION CATALYZED BY PEROXIDASE. Joseph S. Beckman and James N. Siedow. Duke Univ., Durham, N.C. 27706

The enzymatic oxidation of phenolics to yield toxic quinones has long been recognized as a possible defense against disease. However, Peroxidase also releases phenolic free radicals into solution, which may be more toxic than the final quinone products. Survival of *Pseudomonas fluorescens* and *Erwinia amylovora* was determined after exposure to benzoquinone (BQ) or the combination of hydroquinone, peroxidase and H₂O₂ (Hq-P). The time course of killing with BQ showed a lag of several minutes that was proportional to concentration. The lag is shorter in the Hq-P reaction. The lag was apparently due to reaction with multiple sites being required to "kill" the bacterium. Neither anaerobic conditions nor the addition of the chelator DTPA had significant effects on the lethality of either reaction, indicating that reactive oxygen species are not required for the observed toxicity. (This work was supported by NIH grant GM26095).

55

ELECTROPHORETIC PROTEIN PROFILES OF TOTAL CELL ENVELOPES OF XYLEM-LIMITED PLANT PATHOGENIC RICKETTSIA-LIKE BACTERIA (RLB). C. J. Chang and N. W. Schaad, Dept. of Plant Pathology, Univ. of Georgia, Georgia Station, Experiment, GA 30212.

The total cell envelope proteins of Pierce's disease (PD), phony peach disease (PP), plum leaf scald (PLS), elm leaf scorch (ELS)

and periwinkle wilt (PW) organisms were compared using SDS-polyacrylamide-gel electrophoresis (SDS-PAGE). Total envelopes isolation and electrophoresis were carried out as described (W. E. Kuriger and N. W. Schaad, In Rickettsiae and Rickettsial Diseases, Eds., W. Burgdorfer and R. L. Anaker pp. 483-492, Academic Press, 1981, New York). Profiles of the five RLB showed overall similarity but were easily differentiated from one another. All the strains tested, except PW, contained major polypeptides (MP) of 63.5, 53.5, 43.5, 35.5, and 21.7 Kdal. PD strains had additional MP of 18.3 and 14.6 and PD and ELS strains showed a MP of 58.0 Kdal. Profiles of PP and PLS strains had additional MP of 48.0 and 30.5 Kdal and PP had a MP of 57.0 when PLS had one of 58.0 Kdal. The two PW strains had MP of 61.0, 50.5, and 41.2 Kdal.

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EFFECT OF TEMPERATURE ON THE POPULATION DYNAMICS OF *PSEUDOMONAS SYRINGAE* PV. *SYRINGAE* IN PEACH TREES. Elke Endert and D. F. Ritchie, Dept. of Plant Pathology, North Carolina State University, Raleigh 27650.

One-yr-old dormant peach trees (*Prunus persica* cv. 'Clayton') were subjected to 20/14C day/night acclimation in environmental chambers. Trees were puncture inoculated with *P. syringae* pv. *syringae* after 1 wk, and 24 hr later chilled to +5, -5, or -10C for 6 hr and thereafter maintained at 14/10C d/n temperatures. Internal populations were assayed at predetermined distances from the inoculation site. Exposure to -10C resulted in the most rapid initial increase of bacteria, although no visible tissue injury was induced by the chilling. Maximum colonization of tissue was followed by development of sunken, discolored cankers and subsequent gumming. A similar experiment using trees whose dormancy had been fulfilled resulted in 42 and 88% injury to the blossom buds exposed to -5 and -10C, respectively. Bacterial population increase and symptom expression were slower than occurred in the first experiment. In both experiments, the bacteria remained within 2 cm of the inoculation site.

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Selective medium for *Xanthomonas campestris* pv. *translucens*. H. K. Kim, M. Sasser* and D. C. Sands. Dept. of Plant Pathology, Montana State Univ., Bozeman, MT 59717. *Dept. of Plant Science, Univ. of Delaware, Newark, DE 19711.

Xanthomonas campestris pv. *translucens* is a pathogen of cereal grains attacking wheat, barley, rye, oats and other grasses. Heretofore there was no selective medium that could be used in studying the ecology and epidemiology of this pathogen. We have developed a medium designated KM-1, containing: 10g of lactose, 4g of D(+) trehalose, 0.2g of thiobarbituric acid, 0.03g of yeast extract, 1g of NH_4Cl , 0.8g each of K_2HPO_4 and KN_3PO_4 and 15g of Difco Bacto-agar per liter of distilled water. Before adding agar, the ingredients are dissolved completely and pH is adjusted to 6.6 with N NaOH. After autoclaving, the medium is cooled to 50C and 100 mg cycloheximide (dissolved in 95% ETOH), 1 mg ampicillin and 8 mg tobramycin (in 50% ETOH), are added. Most common soil and plant-associated bacteria do not grow on KM-1. This medium exhibited high selectivity with soil samples and barley leaf debris. The plating efficiency ranged from 0.91 to 2.13 for strains of *X. campestris* pv. *translucens* compared to the non-selective Wilbrink's medium.

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CHARACTERISTICS OF STRAINS OF *PSEUDOMONAS SOLANACEARUM* FROM CHINA. L. Y. He, Chinese Acad. Agr. Sc., Beijing, China, L. Sequeira, and A. Kelman, Dept. of Plant Pathology, Univ. of Wisconsin-Madison, Madison, WI 53706.

Twenty-nine strains of *Pseudomonas solanacearum* isolated from 14 cultivated and wild host plants in different locations in China were compared in terms of physiological characteristics in culture and pathogenicity to six differential host plants. Based on Hayward's classification scheme, two strains were placed in biotype II, 14 in biotype III, and 10 in biotype IV. Three strains from mulberry, however, differed from established biotypes because of their ability to produce acid from three disaccharides and mannitol, and their failure to oxidize dulcitol and sorbitol. These strains have been placed in a new biotype, biotype V. Also, these strains were characterized by low virulence on potato (cv. Russet Burbank) and eggplant (cv. Black Beauty). All other strains belonged to races 1 or 3, which are highly virulent on these host plants.

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INJURY TO POTATO TISSUE BY PROTEASE OF *PSEUDOMONAS FLUORESCENS* (BIOTYPE A). J.-S. Wang and A. Kelman, Dept. of Plant Pathology, University of Wisconsin-Madison, WI 53706.

Tuber slices were macerated rapidly by strain W51 of *Pseudomonas fluorescens* (Biotype A). Cell walls in infected tissue were degraded and cells were killed in 4-6 hr. Culture filtrates of W51 grown on a range of substrates were positive for protease but negative for cellulase. Assays for pectic lyase and polygalacturonase at different pH values and with and without Ca^{++} were also negative. An extract from infected tissue had high protease activity with casein as a substrate. Hydroxyproline was released from potato tuber cell walls by culture filtrates. Partially purified enzyme preparations killed potato cells and macerated potato tuber tissue. Three different fractions with proteolytic activity on casein were separated by ion exchange chromatography. Of the three fractions eluted only one was capable of injuring potato tuber tissue. Levels of protease activity were significantly higher in filtrates of W51 than in filtrates of other fluorescent pseudomonads from soil or strains of *Erwinia carotovora*.

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DIVERSITY OF EPIPHYTIC BACTERIAL COMMUNITIES ON BEAN (*PHASEOLUS VULGARIS*) LEAVES AND PODS BASED ON NUTRIENT UTILIZATION. C.E. Morris and D.I. Rouse. Dept. Plant Path., UW, Madison, WI 53706

To assess the diversity of epiphytic bacteria on leaves and pods of young to mature bean plants from the field, washings were plated on tryptic soy and King's B media. Fifty isolates representing all types of bacteria growing on plates were randomly selected from washings of each of three leaves and three pods at each sampling date. Isolates were replica-plated on a series of minimal media supplemented with single carbon or single nitrogen sources. Thirteen carbohydrates and organic acids were tested as carbon sources. Nine amino acids were tested as nitrogen and as carbon sources. There were differences in nutrient utilization by communities on young leaves as compared with those on mature leaves. On young leaves the majority of epiphytes utilized only a few carbon and nitrogen sources. Most of these sources were amino acids. On mature leaves the majority of epiphytes utilized a wide variety of the nutrients tested. The types of nutrients utilized by epiphytes on pods were different from those utilized by epiphytes on leaves.

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RELATIONSHIP OF THE XYLEM-LIMITED BACTERIA CAUSING PERIWINKLE WILT AND PIERCE'S DISEASES. M.J. Davis, Univ. Fla., IFAS, AREC, Ft. Lauderdale, FL 33314; B.C. Raju, Yoder Bros., P.O. Box 68, Alva, FL 33920; R.H. Bransky and R.F. Lee, Univ. Fla., Lake Alfred, FL 33850; R.E. McCoy and R.C. Norris, Univ. Fla., IFAS, AREC, Ft. Lauderdale, FL 33314. The periwinkle wilt (PW)-associated bacterium was isolated on BCYE and PW media. Both periwinkle and grapevine were inoculated by needle puncture with four isolates of the PW bacterium (PWB) and with five strains of the Pierce's disease bacterium (PDB) from grapevine, almond, and alfalfa. Some PWB and PDB strains infected both hosts as confirmed by electron microscopy and reisolation. All PWB isolates incited PW symptoms and were reisolated from periwinkle, and all PDB strains incited PD symptoms and were reisolated from grapevine. PWB did not incite discernible symptoms in grapevine. Only slight chlorosis of mature leaves but not wilting was associated with PDB in periwinkle. PWB was not isolated and grew poorly in subculture on PD2 medium unlike PDB. ELISA indicated that PWB was serologically distinguishable from PDB.

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ISOLATION OF BACTERIA FROM THREE ELM SPECIES AND MULBERRY EXHIBITING LEAF SCORCH. S.J. Kostka, J.L. Sherald*, and T.A. Tattar. Dept. of Plant Pathology, Univ. of Mass., Amherst, MA 01003 and *National Park Service, USDI, Washington, DC 20242

Leaf scorch symptoms typically associated with fastidious, xylem-limited bacteria (XLB) were observed in *Ulmus pumila* (Siberian elm) and *U. americana* cv 'Augustine Ascending' in Washington, DC and New Orleans, LA and in *U. glabra* (Scots elm) and *Morus* sp. (mulberry) in Wash., DC. Mulberry exhibited symptoms in seedling to mature trees. Elms were established trees (>15 cm in dia. at 1.4 m). Bacteria were detected in stems of symptomatic trees using vacuum extraction and phase contrast microscopy and by incubating wood chips in a modified PD-2 broth at 28 C. Turbidity was evident after 7-9 days. All isolates (subcultured on PD-4) were Gram negative, catalase positive and immunofluorescent positive against antisera to the elm scorch bacterium or Pierce's disease bacterium. Symptomless* trees were free of XLB. The data demonstrate an increased host range for XLB and common hosts in two geographically and environmentally distinct regions.

THE EFFECT OF LIGHT AND TEMPERATURE ON THE GENERATION TIME, ADSORPTION, AND YIELD OF THE CYANOPHAGE AS-1. G. B. Olson and P. R. Desjardins. Department of Plant Pathology, University of California, Riverside, CA 92521.

Different light combinations and temperatures were separately tested on the one-step growth curve of the AS-1 *Synechococcus leopoliensis* (Anacystis nidulans, IU 625) system [Cyanobacteria]. Analysis of light spectra of different light sources at 30°C showed a linear relationship between virus adsorption (percent) and red light intensity and between virus yield and the red/far-red ratio. Environmental parameters could be manipulated to vary adsorption and yield separately. Temperature, under a single light regime, dramatically varied the length of the lytic cycle, but virus adsorption (percent) and virus yield were relatively constant. An inverse log-linear relationship existed between total generation time and temperature over the range tested. These data support the contention that adsorption is a separate process from virus yield. Light quality directly affects virus adsorption, multiplication, and release. Temperature primarily affects the rate at which the reactions occur.

ENVIRONMENTAL MODIFICATION OF AVOCADO SUNBLITCH VIROID SYMPTOMATOLOGY IN HASS AVOCADO. G. B. Olson, R. J. Drake, and P. R. Desjardins. Department of Plant Pathology, University of California, Riverside, CA 92521.

No herbaceous host is known for Avocado Sunblotch Viroid (ASBV). Symptom development in avocado requires 45 days (rarely) to 2 years. A quick *in vivo* test system would aid ASBV research. Hass seedlings were inoculated with a nucleic acid extract from ASBV leaf tissue by stem slashings and cut back. After the axial buds broke, the seedlings were placed in an environmental chamber at elevated temperature (32°C/29°C) under varying light conditions. Symptoms 30 days after inoculation were atypical for sunblotch, but similar to Potato Spindle Tuber Viroid and Citrus Exocortis Viroid in herbaceous hosts. Preliminary results show a correlation between concentration of viroid inoculum and symptom expression as well as differences in leaf number, epinasty, and leaf curl between controls and ASBV seedlings. Early symptoms have been detected within 22 days post inoculation. This technique may be applicable to other slow developing virus or viroid diseases in perennial crops.

AN UNUSUAL STRAIN OF CITRUS EXOCORTIS VIROID. J. G. UTERMOHLEN AND J. S. SEMANCIK. DEPARTMENT OF PLANT PATHOLOGY, UNIVERSITY OF CALIFORNIA, RIVERSIDE, CA 92521.

Citrus exocortis viroid (CEV) has been characterized as an infectious, single-stranded, circular RNA with a molecular weight of 119,000. A field isolate was characterized as CEV-moderate by symptomatology produced in *Eurog citron* Arizona 861. This isolate has been transmitted to *Eurog citron* by budding or tissue grafts of infected tissue. Severe symptomatic tissue can be obtained from CEV-moderate infected plants. Nucleic acid extracts from infected tissue, independent of symptom severity, failed to produce CEV-like symptoms when mechanically inoculated into either "Rutger" tomato or *Gynura aurantiaca*. The 2M LiCl soluble fraction of these nucleic acid extracts did not contain detectable levels of the viroid RNA when analyzed on polyacrylamide gels. However, the 2M LiCl precipitable fractions from the same extracts did contain RNA sequences which hybridized to 32p labeled CEV. The level of hybridization was equal to that observed for similar extracts from CEV-infected *Eurog citron*.

ELISA, RIA, AND ISEM: A COMPARISON OF THEIR ABILITY TO DETECT BLUEBERRY SHOESTRING VIRUS (BBSSV). J. M. Gillett, K. M. Morimoto, D. C. Ramsdell, W. G. Chaney, K. K. Baker and W. J. Esselman. Department of Botany and Plant Pathology, Michigan State University, East Lansing, Michigan 48824.

Three assays, enzyme-linked immunosorbent assay (ELISA), solid phase radioimmunosorbent assay (RIA), and immunosorbent electron microscopy (ISEM) were compared for their ability to detect BBSSV in purified form and in the BBSSV aphid vector, *Illinoia pepperi*. Using purified BBSSV, ELISA detected a minimum of 5.0ng BBSSV, RIA 0.5ng, and ISEM less than 0.15ng. When purified BBSSV was added to non-viruliferous *I. pepperi* extracts, ELISA detected BBSSV at a minimum concentration of 5.0ng, RIA 0.5ng, and ISEM 20ng. When aphids were fed for 24 hr on sachets of purified BBSSV, all three assays reliably detected the presence of BBSSV. However, when aphids were fed for 24 hr

on BBSSV-infected blueberry leaves, RIA detected 47/53 aphids as viruliferous, ELISA 10/53, and ISEM rarely detected any. While ISEM is most sensitive for detecting purified BBSSV, RIA is most sensitive for detecting BBSSV in aphids.

TIME COURSE OF BLUEBERRY SHOESTRING VIRUS (BBSSV) ACQUISITION BY THE APHID VECTOR *ILLINOIA PEPPERI* AS DETERMINED BY ¹²⁵I-LABELING TECHNIQUES. K. M. Morimoto, J. M. Gillett, W. G. Chaney, D. C. Ramsdell. Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824.

Blueberry shoestring virus (BBSSV) was detected by radioimmunoassay (RIA) in single aphids fed on BBSSV-infected tissue or on sachets containing purified BBSSV or ¹²⁵I-labeled BBSSV. Purified BBSSV and anti-BBSSV rabbit gamma globulin were iodinated with ¹²⁵I using the chloramine-T method. The presence of virus within individual aphids was determined with a double antibody sandwich method of RIA or aphids were directly counted where ¹²⁵I-labeled BBSSV was used. Nonviruliferous aphids were allowed acquisition access times of 10 min, 1, 6, 12, 24, 48, and 72 hr. The number of aphids containing detectable virus reached a maximum at 24 hr. The aphids acquired greater quantities of virus when allowed to feed on the sachet versus the infected tissue. Virus content was found to vary considerably between individual aphids which had fed on tissue or BBSSV-containing sachets.

LOCALIZATION OF BLUEBERRY SHOESTRING VIRUS (BBSSV) IN THE BLUEBERRY APHID, *ILLINOIA PEPPERI*. Maureen A. Petersen, Donald C. Ramsdell, Karen K. Baker and W. G. Chaney. Dept. of Botany and Plant Pathology, Michigan State Univ., E. Lansing, MI 48824

Scanning electron microscopy autoradiography (SEM AR), using backscattered electrons, was used to localize ¹²⁵I-labeled BBSSV in its vector *Illinoia pepperi*. Aphids were fed ¹²⁵I-BBSSV, or various control solutions, and processed for SEM AR. Aphids were examined on an SEM using backscattered electrons. After a 6 hr feeding time the radiolabeled virus was detected in the thorax, in the posterior thorax at 12 hr, in the stomach at 24 hr, and in the posterior regions of the abdomen at 72 hr. Controls showed only nonspecific background radiation. Variations in feeding behavior occasionally resulted in inconsistencies in location of radioactive label. These data demonstrate that BBSSV acquired by *I. pepperi* travels through the alimentary canal. The method of SEM AR, with appropriate controls, can be a valuable tool in studying the movement of viruses through their vectors.

A SIMPLE SYSTEM FOR GEL ELECTROPHORESIS OF INTACT VIRUSES. Leslie C. Lane and Bharati Joshi, Dept. of Plant Pathology, Univ. of Nebraska, Lincoln, NE 68583-0722.

Intact virus particles, including those of tobacco mosaic virus (TMV, rod-shaped, length 300 nm) and wheat streak mosaic virus (WSMV, filamentous, length 700 nm) can be electrophoresed in horizontal agarose slabs, containing as little as 0.3% agarose, poured on GelBond (Marine Colloids). Treating samples with specific antisera prior to electrophoresis prevents specific antigens (viruses) from migrating into gels. Viruses can be stained with Crowles stain (protein) or ethidium bromide (nucleic acids). Nucleic acids inside TMV and WSMV stain only after disrupting the viruses by brief exposure to alkali. Nucleic acids inside many spherical plant viruses stain directly without capsid disruption. Submicrogram amounts of virus are easily detected. The technique is useful for diagnosis, comparison of virus strains and for quantitative analysis of virus concentrations.

SOUTHERN BEAN MOSAIC VIRUS STRAIN DERIVED BY PREFERENTIAL SYSTEMIC MOVEMENT. M. H. McGovern and C. W. Kuhn, Dept. of Plant Path., Univ. of Georgia, Athens, GA 30602.

A new strain of southern bean mosaic virus (SBMV), designated NCP, was derived when resistant cowpea plants were inoculated with the cowpea strain (CP) and maintained at 21 and 24 C. NCP was serologically different from CP and three other strains of SBMV. Strain CP caused necrotic local lesions on cowpea cultivar Clay and PI 399419 while NCP caused no symptoms on Clay and local chlorosis and systemic mosaic on PI 399419. Virus accumulation was similar in PI 399419 but NCP was about 10 times greater in Clay than CP. Systemic movement and subsequent replication, particularly in PI 399419, appeared to be responsible for the derivation of NCP. While little or no CP moved

systemically at 21 and 24 C, NCP occurred frequently in uninoculated leaves, even when the original CP inoculum was transferred serially via local lesions. We speculate that the NCP genome induces a protein which aids or is required for systemic movement.

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TMV INDUCES ENATIONS ON NICOTIANA TOMENTOSA. James L. White, Prairie Regional Laboratory, National Research Council of Canada, Saskatoon, Saskatchewan, S7N 0W9.

TMV-infected *N. tomentosa* develop enations on systemically infected leaves. These outgrowths are dark green, leaf-like and vary in size and shape extending downward as much as 0.5 cm. To determine if the enations were virus free, disinfested enation or yellow-green infected tissues were placed in Linsmaier and Skoog (LS) medium containing IAA [0.6 mg/L] and N-dimethylallyl-adenine (DiMeAd) [2 mg/L]. Calli were transferred to LS medium containing IAA [0.1 mg/L] and DiMeAd [10 mg/L] to induce shoots and later to LS medium lacking hormones to induce roots. About 70% of the plantlets regenerated from yellow-green areas and about 1/3 of the plants regenerated from yellow-green areas were virus free. Virus-free plants regenerated from enations were inoculated with TMV. Enations developed on approximately 75% of these plants while only 6% of the plants from seeds (from Tobacco Research Lab., Oxford, N.C.) formed enations. Whether the high frequency enation forming plants transmit this characteristic through seeds is being determined.

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CHARACTERIZATION OF ARAUJIA MOSAIC VIRUS (POTYVIRUS GROUP) BY IN VITRO TRANSLATION ANALYSES. E. Hiebert and R. Charudattan Dept. of Plant Pathology, Univ. of Florida Gainesville, FL 32611.

Araujia mosaic virus (AjMV) was tentatively described as a new member of the potyvirus group (Phytopathology 70:909-913). In vitro translation analyses were used to further compare AjMV with other potyviruses. Isolated AjMV RNA was translated in a rabbit reticulocyte lysate system. The products were analyzed by immunoprecipitation and SDS-PAGE. Two products, estimated MW 48,000 (48k) and 53k, similar in size and serological reaction to tobacco etch virus (TEV) nuclear inclusions and capsid protein were identified among the AjMV RNA translation products. Antiserum to TEV capsid protein reacted with the presumed AjMV capsid protein while antiserum to TEV pinwheel inclusions did not react with any of the AjMV translation products. The proposed genetic map for AjMV is: 5' end -81k protein-48k protein-42k protein-70k pinwheel inclusion protein-53k protein-31k capsid protein-3' end. These results show that three genetic products of AjMV are serologically related to products of TEV.

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ANALYSES OF THE CAPSID, NUCLEAR AND PINWHEEL INCLUSION PROTEINS OF FOUR PHENOTYPICALLY DISTINCT STRAINS OF TOBACCO ETCH VIRUS. R. G. Christie, E. Hiebert, Univ. of Florida Gainesville FL., and W. G. Dougherty NCSU, Raleigh, N.C.

The structural and three nonstructural proteins of four phenotypically distinct strains of tobacco etch virus were analysed. The four strains were phenotypically similar but were distinguished as follows; 1) "typical", 2) nonaphid transmissible, 3) "severe" (severe symptoms on tobacco, and 4) oxnard (forms unique bipyramidal nuclear inclusions and infects *N. tabacum* V-20). The strains were similar in virus particle length measurements and in SDS-immunodiffusion tests. Partial proteolytic digestion of the capsid proteins with *S. aureus* V-8 protease and analysis by SDS-PAGE revealed identical peptide patterns. The cytoplasmic inclusion proteins of the four strains appeared identical in serological tests and in peptide digests. Serological tests did not reveal differences in the nuclear inclusion proteins of the four strains. At the level examined, differences which might be correlated with the observed viral phenotypes were not detected in four viral proteins representing 60% of the viral genome.

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NUCLEOLAR AND EXTRANUCLEOLAR PERICHRROMATIN GRANULES INDUCED BY EUPHORBIA MOSAIC VIRUS. K. S. Kim and E. M. Martin, Dept. of Plant Pathology, University of Arkansas, Fayetteville, AR 72701

Cells of *Datura stramonium* infected with Euphorbia mosaic virus, a member of the geminivirus group, exhibited unusual accumulations of electron-dense granules in the nucleoplasm. The granules, 50-70 nm in diameter, were grouped in clusters and occupied much of the peri- and interchromatin areas. The granules originated from either perinuclear regions or from fibrillar centers of the nucleoli, and migrated into the interchromatin regions. In the processes of typical nuclear changes due to

virus infection, the granules were observed before the appearance of virus particles and were often associated with fibrillar bodies, another characteristic inclusion induced by the virus. They were seldom observed at late stages of infection indicating they were not degenerating products of nuclear components. A cytochemical study indicated that the granules contain RNA suggesting that they are perichromatin granules formed by the disturbance of nucleolar and extranucleolar RNA metabolism.

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PRODUCTION OF HYBRID CELL LINES SECRETING VIRUS SPECIFIC MONOCLONAL ANTIBODIES TO THREE ILARVIRUSES AND ALFALFA MOSAIC VIRUS. E. L. Halk, H. T. Hsu, J. Aebig, and K. Chang. American Type Culture Collection, Rockville, MD 20852.

Hybrid cell lines (hybridomas) were produced by somatic cell fusion between mouse myeloma cell lines NS1/1 or P3X63Ag8.653 and spleen cells from Balb/c mice immunized with a mixture of purified tobacco streak (TSV), prunus necrotic ringspot (NRSV), apple mosaic (ApMV) and alfalfa mosaic (AMV) viruses. Hybridomas secreting antibodies specific for the viral antigens were detected by indirect ELISA. Eighteen stable hybridoma lines were selected by 2-3 cycles of single cell cloning. Antibody producing hybridomas were directed against TSV (7 lines), AMV (2 lines), NRSV (5 lines), ApMV (3 lines) and both NRSV and ApMV (1 line). Monoclonal antibodies were produced from cells in culture or from ascites tumors in mice injected with hybridoma cells. ELISA antibody titers (reciprocal) of cell culture media ranged from 10^1 - 10^3 whereas ascites titers ranges from 10^4 - 10^7 . These immortal cell lines will be able to provide a continuous supply of antibody for the future.

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CLONING OF A DNA COPY OF TOBACCO ETCH VIRUS RNA. William G. Dougherty and John C. Sorenson*, Departments of Plant Pathology and Genetics*, North Carolina State University, Raleigh 27650.

Tobacco etch virus (TEV), a member of the potyvirus group, has a single stranded RNA genome with a mol wt of 3.2×10^6 . To provide specific probes for transcriptional and translational analyses complementary DNA (cDNA) sequences of TEV RNA have been cloned. In an oligo-dT primed reaction, cDNA was synthesized using reverse transcriptase. RNA was removed by alkali digest and a short poly-C tail added to the 3'-OH terminus of the cDNA using terminal transferase. The cDNA was made double stranded using the Klenow fragment of DNA polymerase I in an oligo-dG primed reaction. Eco RI and Sal I 'linkers' were ligated to the cDNA and the insert digested with the restriction enzymes Eco RI and Sal I. The cDNA insert was ligated into the plasmid pBR322 which had been digested with the same restriction endonucleases. Recombinant plasmids with cDNA inserts ranging from 3500 base-pairs to 100 basepairs were generated. A restriction map was generated and discrete viral RNAs were detected in infected tobacco seedlings in 'Northern' hybridization experiments.

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ENZYME CONJUGATED PROTEIN A: A UNIVERSAL REAGENT FOR INDIRECT ENZYME-LINKED IMMUNOSORBENT ASSAYS (ELISA). H. T. Hsu, J. Aebig, ATCC, Rockville, MD 20825; E. L. Civerolo, C. Helkie, USDA, Beltsville, Maryland 20705.

Protein A (PA) binds specifically to the F_c portion of IgG molecules. Antigens can be detected by the reaction between PA-enzyme conjugates and specific antibodies immobilized in microtiter plates. The F_c portion of the coating IgG can be removed by pepsin digestion precluding binding of the PA-conjugate to the coating antibody. The resulting $F(ab')_2$ retains antigen-binding specificity. The PA-enzyme conjugate can serve as a universal reagent in indirect ELISA, replacing individual antibody-enzyme conjugates for each pathogen to be assayed. Samples of plant extracts containing viral and bacterial pathogens, dilute specific antisera, PA-enzyme, and appropriate substrate were successively incubated in $F(ab')_2$ -sensitized plates. The system has been successfully used to detect tobacco ringspot, tomato ringspot apple mosaic, prune dwarf, prunus necrotic ringspot, and potato X viruses, and strains of *Xanthomonas campestris* pv. *citri* in plant tissue extracts.

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LOCALIZATION OF BARLEY STRIPE MOSAIC VIRUS (BSMV) IN ULTRATHIN SECTIONS WITH COLLOIDAL GOLD-LABELLED IgG. Na-Sheng Lin and W.G. Langenberg, Dept. of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722.

An immunocytochemical technique for the ultrastructural localization of viral protein has been used successfully by direct

post-embedding staining with gold-labelled antibody. Ultra-thin sections of glutaraldehyde-fixed and Lowicryl- or Araldite Epoxy-embedded tissues were etched with alcoholic sodium hydroxide to uncover the partially masked antigenic sites. Without etching or after osmic acid post-fixation, staining intensity was low. Staining was also less specific and more difficult to achieve with other plastics. High intensity of specific staining was obtained in sections of BSMV-infected wheat tissue. Non-specific staining could be reduced by adding normal serum to the staining solution. Barley stripe mosaic virus protein accumulated in the cytoplasm and also in the nucleus of systemically infected wheat cells. Systemically infected root tips were a better source for the early detection of BSMV than infected leaves.

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CHLOROPLAST RNA AND PROTEINS DECREASE AS WHEAT STREAK AND BARLEY STRIPE MOSAIC VIRUSES MULTIPLY IN EXPANDING, SYSTEMICALLY INFECTED LEAVES. James White and Myron Brakke, Prairie Regional Laboratory, Saskatoon, Sask. S7N 0W9, and USDA-ARS, Dept. of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722.

Wheat streak mosaic (WSMV) and barley stripe mosaic (BSMV) viruses accumulated predominantly during expansion of systemically infected wheat or barley leaves. Viral RNA polymerases reached peak activity in expanding, systemically infected leaves, confirming the rapid virus multiplication in these leaves. Chloroplast ribosomal RNA, but not cytoplasmic, was reduced in infected as compared to uninfected leaves. Analysis of proteins from systemically infected leaves by SDS gel electrophoresis showed that certain proteins were present in decreased relative amounts as a result of infection by either virus. One of these was the large subunit of ribulosebiphosphate carboxylase, and the others were tentatively identified as chloroplast related. Another set of proteins, probably viral coded, were present in increased relative amounts, were mostly different for each virus, and included viral coat proteins.

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CUCUMBER MOSAIC VIRUS DISEASE-INDUCED CRYSTALLINE INCLUSION APPEARANCE, ENLARGEMENT, AND DISPERSAL IN SWEET PEPPERS. G.W. Moorman. University of Massachusetts, Suburban Experiment Station, 240 Beaver Street, Waltham, MA 02254.

Crystalline inclusions induced in 'Yolo Wonder' sweet peppers by cucumber mosaic virus were found concentrated in the epidermis covering the abaxial surface of veins of inoculated leaves using the azure A staining technique of Christie and Edvardson (1977, Fla. Ag. Exp. Sta. Monograph 9). Crystals were sparse elsewhere in the leaf. Two types of crystals were observed. Hexagons, always smaller than the cell nucleus, remained separate from one another. Angular plates, often larger than nuclei, varied in maximum size from 25 to 110 μm^2 (longest length X width taken perpendicular to length measurement). Angular plates were singular or aggregated into asterisk-like shapes. These plates appeared 3-5 days after inoculation and enlarged and dispersed within 20 days at 32°C. At 21°C, 5-7 days were required for appearance and 30 days for enlargement and dispersal (20 hr. day length).

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INFLUENCE OF VIRUS INFECTION OF HOST PLANTS ON APHID VECTOR MORPHOLOGY. F. E. Gildow. Plant Pathology Department, University of California, Berkeley, CA 94720.

Cereal grain aphids, *Sitobion avenae* and *Rhopalosiphum padi*, were more likely to mature as winged adults (alatae) when reared on oats or barley infected with BYDV, than were aphids reared on healthy plants. When 400 *S. avenae* were reared at 15C with a 16 hr photoperiod, the percent aphids maturing as alatae on BYDV-infected oats or healthy oats was 70% and 24%, respectively. Similar results were obtained with *R. padi* reared on several susceptible or tolerant oat and barley varieties. Early instar nymph of *R. padi* collected in the field from BYDV-infected or healthy barley were reared to maturation on healthy oats and alatae counted. The percent aphids maturing as alatae from BYDV-infected or healthy plants was 87% of 907 aphids, and 24% of 400 aphids, respectively. Although different plant species did influence wing development of *Myzus persicae*, luteovirus infection of host plants had no effect.

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OCCURRENCE, SPREAD, AND EVALUATIONS OF DENT CORN HYBRIDS AND IN-BRED LINES FOR REACTION TO CORN LETHAL NECROSIS IN NEBRASKA. Ben Doupnik, Jr., Les Lane, and David S. Wycsong. Univ. of Nebr. Clay Center, 68933, and Lincoln, 68583.

Corn lethal necrosis (CLN), a disease of maize caused by the synergistic interaction of maize chlorotic mottle virus (MCMV) and maize dwarf mosaic virus-strain B (MDMV-B) was found in Nebraska in 1977 in one field. In 1978 CLN was epiphytotic in 3 counties and was found in 2 additional counties in 1980. Since little data was available on the field reactions of dent corn to CLN, evaluations were made from 1979-1981. Plots were located in fields having previous histories of CLN and inoculated by rubbing the upper 2-3 leaves at the 7-9 leaf stage with a buffered suspension of sap expressed from fresh corn leaves previously inoculated with MCMV and MDMV-B in the greenhouse. Final disease intensity ratings were made in August on a scale of 1-5 (1 = no disease symptoms; 5 = dead plants). Of the 296 hybrids and 213 inbred lines tested, only 46 and 8, respectively, had disease intensity ratings of 2.5 or less. The inbreds were AR252, B64, B68, Ex315, ND376, Oh91613, Oh91678, and Pa405.

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SPECIFICITY OF ACTINOMYCIN D INHIBITION OF PLANT VIRUSES. M.S. Turner and W.O. Dawson, Department of Plant Pathology, University of California, Riverside, CA 92521.

Actinomycin D (AMD) has been shown to inhibit an early step in the replication of many plant viruses. To determine whether a host-coded function induced by one virus can function in the replication of other viruses, cowpea plants were inoculated with a virus and incubated until replication had progressed beyond the AMD-sensitive step. They were then inoculated with a second virus and AMD (50 $\mu\text{g}/\text{ml}$) was added at times afterward to leaf discs made from doubly infected plants. Amount of inhibition was determined by local lesion assay. Cowpea chlorotic mottle virus, tobacco mosaic virus (cowpea form), and cowpea mosaic virus were used in this study. In all combinations of viruses tested the amount of inhibition shown by the first virus was equal to that observed in the singly-inoculated controls.

83a

THE ROLE OF SEED-BORNE INFECTION AND RAINFALL IN BROWN BLOTCH (*COLLETOTRICHUM TRUNCATUM*) OCCURRENCE AND SEVERITY IN NIGERIA. DR. P.O. OYEKAN, UNIVERSITY OF IFE, I.A.R. & T., MOOR PLANTATION, P.M.B. 5029, IBADAN, NIGERIA.

Cowpea brown blotch caused by *Colletotrichum truncatum* was first observed on a small scale in the northern part of Nigeria in 1976. Within two years the disease had spread to the southern part of the country where it has since been causing severe epiphytotics on cowpea.

Cowpea planted early during the wetter part of the cropping season as currently recommended had severest attack of brown blotch while the crop that was grown during dryer period of the season had less disease. Seed infection varied with the severity of the disease on the crop in the field attaining over 90% in crops planted early in the season. Seed-borne infection is believed to be responsible for the rapid spread of cowpea brown blotch in Nigeria.

83b

SOLAR HEATING (SOLARIZATION) OF TOMATO SUPPORTS FOR CONTROL OF *DIDYMELLA LYCOPERSICI* KLEB. STEM CANKER. M. Besri, Institut Agonomique et Vétérinaire Hassan II B.P. 704, Rabat-Agdal, Morocco.

Previous studies have shown that *Didymella lycopersici* survives mainly in reed canes and Eucalyptus stakes, used to support tomatoes. To control this important disease, tomato supports were placed in April, on soil previously irrigated. They were abundantly moistened and then covered with transparent plastic film (180 μ). The plastic was removed in September. The maximum and minimum temperatures recorded in the atmosphere from May to September vary respectively from 26°C to 44°C and from 8°C to 16°C. The maximum temperatures recorded during the same period under plastic were always higher than 50°C and the minimum temperatures varied from 11°C to 20°C. In the plots trellised with solarized and unsolarized supports, the percentage of plants with *Didymella* cankers at the last month of tomato crop was respectively 2 and 20%. Therefore, we can conclude that tomato support solarization reduced the incidence of *D. lycopersici*.

83c

FIELD TESTS TO EVALUATE BAYLETON FOR CONTROL OF POWDERY MILDEW IN VINEYARDS. Frank P. Guerrero, Mobay Chemical Corporation, 1900 N. Gateway, Fresno, CA 93727.

Three applications of BAYLETON (triadimefon) at 3.0 oz a. i./a

on a 28-day interval provided 95% control of powdery mildew (*Uncinula necator*) on 'Chardonnays' and 100% control on 'Cabernet Sauvignon'. Even at 1.0 oz a.i./a control was 99% on the 'Cabernets'. The growers' sulfur treatment (up to 10 applications) gave poor control and resulted in a yield depression of 26% in the 'Chardonnays' and 60% in the 'Cabernets'. A test the following year on the highly susceptible 'Carignane' variety under a constant and severe source of inoculum showed that at least 5 applications of 2.0 oz a.i./a and 3 applications at 3 oz a.i./a were required on a 14 to 28-day interval. Overwintering infections of mildew were effectively controlled at the higher rate.

83d

THE ROLE OF THE FIBRILLAR RINGS ASSOCIATED WITH THE CELL NUCLEUS DURING GEMINIVIRUS REPLICATION. Ram6n Lastra and Francisco Gil, Laboratorio de Virus de Plantas, Instituto Venezolano de Investigaciones Cientificas, Apartado 1827, Caracas, Venezuela.

The presence of fibrillar rings in the nucleus of sieve elements and parenchymal cells infected with several geminiviruses has been established. However, their role in the virus replication is not understood. From observations of tissue infected with the following geminiviruses: Bean Golden Mosaic Virus, Mosaico Amarillo del Tomate and Lima Bean Golden Mosaic Virus, it seems that the fibrillar rings are the first abnormal cytopathological effect induced by these viruses in the cell. Fibrillar rings could be found in the cell nucleus even before the plant showed visual symptoms, usually between 4-7 days after mechanical inoculation. In the following days fibrillar rings were found associated with the virus particles until they are completely replaced by large masses of viruses which remained in the cell nucleus. The fibrillar rings seem to be associated with the early stages of viral replication and are the probably site of viral assembly.

83e

COMPARISON OF CROP LOSS IN PIMA AND UPLAND COTTON CAUSED BY PHYMATOTRICHUM OMNIVORUM. E. MULREAN, UNIV. OF AZ. COTTON RESEARCH CENTER, PHOENIX, AZ., J. MUELLER, DOW CHEMICAL, WALNUT CREEK, CA., R. HINE AND P. VON BRETZEL, UNIV. OF AZ, TUCSON, AZ.

Lint quality and seed cotton yield were significantly reduced by *Phymatotrichum omnivorum* infection in 37 upland cotton fields (2454.2 acres) and 10 Pima cotton fields (532.3 acres) in Arizona. Fields were ground surveyed and hand harvested lint samples were collected for fiber quality analysis. The amount of per acre yield reduction varied considerably between fields. This variation could not be related to cropping history or rotation practices. Infection reduced seed cotton yield in both varieties. Pima cotton retained its fiber quality inspite of infection and only exhibited significant reductions in seed index and fiber fineness. Infected upland cotton showed significantly lowered quality with respect to seed index, fineness, length, and uniformity.

84

PHYTOTOXICITY OF SOIL APPLICATIONS OF JET FUEL. A. L. Granett, H. E. Stone, and E. C. Smith, Statewide Air Pollution Research Center, University of California, Riverside, CA 92521.

JP4 jet fuel was pipetted onto soil near the bases of 15 species of 35-42-day-old potted plants. Applications at the rate of 0.07 ml/cm² per pot were made without fuel contacting the plant stem. Injury induced by the fuel was noted 24 hours after treatment and 7 days later when plants were harvested and weighed. Injury included discolored or chlorotic areas on stem and leaves of alfalfa, bean, corn and sunflower. Necrosis of stems and leaves were observed on cotton, lettuce and marigolds. Collapse or weakening of stems, usually near the soil line, occurred with barley, corn, sorghum, squash, wheat and zinnia plants. Global roots of radish withered, and a large proportion of bacterial nodules on alfalfa (100%) and bean (60%) roots were dead after treatment. Carrot plants exhibited no foliar injury due to the soil treatment, but had reduced and bleached roots. Fresh weights of treated plants were significantly less ($p < 0.05$) than controls for all species except wheat and zinnia.

85

MOVEMENT AND RETENTION IN SOIL OF AN UNIDENTIFIED TOXICANT IN JET FUEL. A. L. Granett, H. E. Stone, and E. C. Smith, SAPRC, University of California, Riverside, CA 92521.

Movement of a phytotoxic component of JP4 jet fuel through sandy greenhouse soil (UC Mix II) was measured using a sorghum seed (*Sorghum sudanense*) bioassay. Fuel was applied to the top of packed soil columns 50 cm long and 8 cm diameter. After 7 days the columns were cut into 10 equal-width sections and each section was potted and sown with seeds. As fuel applications in-

creased from 1 to 8 ml/cm², soil from 0-20 to 0-50 cm from tops of original columns did not support normal growth. Seeds in affected sections had germination rates ranging from 0 to 55% (mean = $2 \pm 18\%$) compared to a range of 65 to 100% (mean = $82 \pm 9\%$) germination for seeds in untreated soil. Lengths of seedlings in affected sections averaged 8.4 ± 6.5 mm whereas seedlings in the other soil were 118 ± 9.4 mm long. Shale-derived fuel was significantly more toxic than the petroleum form ($p < 0.05$). Inhibition decreased when seed sowing was delayed 14 days after soil treatment. This study verified movement and gradual loss of phytotoxicity in soil contaminated with jet fuel.

86

JET FUEL INHIBITS *PENICILLIUM RUBRUM* AND *TRICHODERMA VIRIDE* GROWTH. H. E. Stone and A. L. Granett, Statewide Air Pollution Research Center, University of California, Riverside, CA 92521.

In field situations, soil microorganisms are impacted by jet fuel spills. *In vitro* growth and sporulation of *Penicillium rubrum* and *Trichoderma viride* were observed in liquid medium containing JP4 jet fuel. The fungi were inoculated into separate flasks of potato dextrose broth plus 0 or 2% (v/v) of JP4 fuel. Stationary cultures were incubated at room temperature with 7 hours artificial light. Hyphal dry weights of both species were significantly reduced ($p < 0.05$) by the fuel treatment, and sporulation was delayed. *P. rubrum* did not change the yellow medium to deep red when fuel was present. *T. viride* cultures were harvested daily during a 10-day growing period; regression analysis of resulting growth curves revealed reduced hyphal dry weights for fuel-treated cultures compared to controls. Fungal growth reductions imply jet fuel may produce changes in the competition among soil fungi. The *in vitro* color reaction of *P. rubrum* may be useful for bioassay techniques.

87

IMPACT OF OZONE ON CORN YIELD. L. W. Kress and J. E. Miller, RER Division, Argonne National Laboratory, Argonne, IL 60439.

Field-grown plants of two hybrids of field corn (*Zea mays* L., Pioneer 3780 and PAG 397) were exposed to O₃ for 7-hr daily between June 20 and September 10, 1981, in open-top chambers. A randomized complete block design incorporated 3 replicates of 7 treatments with seasonal 7-hr average (0900-1600 CST) O₃ concentrations of 0.044 ppm (open plots), 0.015 ppm (charcoal-filtered air), and 0.044, 0.073, 0.100, 0.129, or 0.156 ppm (unfiltered air with O₃ added). Grain yield data indicated a threshold of 0.07 ppm for 3780 and 0.10 ppm for 397 for significant O₃ effects. Regression analysis of the data projected yield reductions of 21.6% (3780) and 9.9% (397) at an O₃ concentration of 0.10 ppm. At 0.06 ppm, predicted effects were 0.4% yield decrease (3780) and 0.2% yield increase (397) which indicates that field corn is significantly less sensitive than soybean to O₃ and no significant adverse effects should be expected at O₃ concentrations currently occurring (0.04-0.07 ppm) in the major corn producing regions.

88

GROWTH, NET PHOTOSYNTHESIS, AND YIELD OF SOYBEAN EXPOSED TO OZONE IN THE FIELD. R. J. Kohut and R. G. Amundson. Boyce Thompson Institute at Cornell Univ., Ithaca, NY 14853.

Hodgson soybeans were exposed in the field to charcoal-filtered air, ambient air, nonfiltered air, and nonfiltered air to which 0.03, 0.06, or 0.09 ppm O₃ was added. Seven-hour exposures were conducted in open-top chambers on 58 days between July 23 and September 30. Yield losses ranged from 8% in nonfiltered air to 41% in the nonfiltered + 0.09 ppm treatment. The latter treatment included 207 hours in which the average O₃ concentration exceeded 0.12 ppm while this level was never exceeded in the former treatment. Regression analysis indicated the relationship between yield in grams per square meter and the 7-hour average O₃ concentration was Yield = $273.7 - (1222.4 \times O_3)$. Foliar injury was observed only in the 0.06 and 0.09 ppm O₃ addition treatments. Above ground plant biomass decreased as the O₃ concentration increased. Reductions in net photosynthesis and increases in leaf resistance were monitored in plants receiving the 0.06 and 0.09 ppm O₃ additions.

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SULFUR DIOXIDE AND OZONE INJURY TO PLANTS IN SOUTH DAKOTA. Gardner, Wayne S. Department of Plant Science, South Dakota State University. Brookings, SD 57007

Since 1970, ozone (O₃) symptoms have been observed on sensitive indicator plants in 19 counties and at 23 locations in South Dakota. Plants showing symptoms of O₃ injury included: alfalfa, aspen, bean, grape, licorice, parsley, pea and Bel-W3, Bel-C, Turkish, Wisconsin-38 and *Nicotiana rustica* tobacco. Sulfur

dioxide (SO₂), resulting from coal or lignite combustion sources injured plants in Brookings, Grant and Walworth counties. Plants injured included: alfalfa, apple, apricot, begonia, black hills spruce, black walnut, bluegrass, box elder, catalpa, chokecherry, chrysanthemum, elder, elm, fern, geranium, gladiolus, hollyhock, lilac, maple, milkweed, morning glory, mulberry, oats, peony, petunia, plumegrass, prune, rhubarb, rose, tomato, spirea, sumac, woodbine, and zinnia. In Walworth County ozone injury was not detected with Bel-W3 tobacco in the years of operation of the SO₂ source. Ozone was detected in the four years after the SO₂ source ceased operation.

90

ULTRAVIOLET-B RADIATION INFLUENCES TRITICUM AESTIVUM GROWTH, PRODUCTIVITY, AND MICROFLORA. P. G. Webb. Fruit Crops Department, University of Florida, Gainesville, FL 32611.

A 1% decrease in stratospheric ozone concentration results in an amplification factor of 2 for ultraviolet-B radiation (280-320 nm) over North Florida. Studies have been initiated to assess responses of food crops to increases in UV-B radiation in the biosphere. Wheat (*Triticum aestivum* L., 'Florida 301') was grown under field conditions during December-April at four UV-B radiation enhancement levels, e.g., 0 (control), 16, 28, and 32%. Sample plants were harvested at 51, 64, 77, and 91 days after planting with a final harvest at 133 days. Plant dry weight, total leaf area, number of tillers, seed dry weight, and number of seed heads decreased as UV-B radiation levels increased. The amount of absorbance/g (chlorophyll) approached a cubic model through time and was not different between the treatments. The smallest UV-B radiation enhancement level occasionally stimulated certain aspects of wheat development. Occurrence of wheat rust was assessed during the final 15-day period.

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Growth of *Xanthomonas phaseoli* on the surface of leaves exposed to SO₂. J. A. LAURENCE AND K. L. REYNOLDS. Boyce Thompson Institute for Plant Research, Ithaca, NY 14853

Increases in epiphytic populations of rifampin-resistant *X. phaseoli* were measured on red kidney bean plants exposed to 0, 160, or 520 ug SO₂ m⁻³ for 6 hr day⁻¹ on each of 5 days before, after, or before and after application of the bacterium to the surface of the first trifoliate leaf. Plants were grown in a greenhouse at 20-25°C for two weeks, transferred to growth chambers for the 10 day exposure period and then returned to the greenhouse. At 0, 5, 10, 15, and 20 days after application of the pathogen, leaves were collected from each of 4 replicates of 9 treatments. Leaf area was determined, and each leaf was washed for 1 hour in water containing a surfactant. Washings were serially diluted and plated on rifampin agar medium for population determination. Visible foliar symptoms of SO₂ and common blight were not observed. SO₂ did not affect the relative growth rates or final population of the pathogen. Relative growth rates ranged from 0.32 to 0.63 day⁻¹.

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CATHODOLUMINESCENT DIAGNOSIS OF *TAXUS MEDIA* INJURED BY AMBIENT AIR POLLUTION.

Charles R. Krause, USDA, ARS, Nursery Crops Research Lab., 359 Main Rd., Delaware, OH 43015

Taxus media ramets were under field conditions from November, 1981, until March, 1982. Plants at Delaware, OH were exposed to air with low background levels of air pollutants while plants at three sites in Cleveland, OH were exposed to polluted air. In March, needles of *T. media* exposed to clean air were uninjured while those exposed to polluted air appeared chlorotic and mottled. Needles of plants grown at each site were compared with a scanning electron microscope equipped with a cathodoluminescence (CL) detector and cold stage. Epidermal cells from needles exposed to clean air were turgid, with uniform epicuticular wax and lacked CL properties. Energy dispersive X-ray analysis (EDXA) did not detect heavy metals. Surfaces of needles exposed to polluted air showed eroded wax and significant levels of particles with CL properties containing significant levels of S, Fe and other heavy metals as detected with EDXA.

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A WIND TUNNEL STUDY OF THE FLOW FIELD WITHIN AND AROUND OPEN-TOP CHAMBERS USED FOR PLANT DISEASE STUDIES. J.M. Davis, Departments of Marine, Earth and Atmospheric Sciences and Plant Pathology, and A.J. Riordan, Department of Marine, Earth and Atmospheric

Sciences, North Carolina State University, Raleigh, NC 27650

Open top field growth chambers are currently used for plant disease assessment. As wind speed increases, there is enhanced ingress of ambient air into these chambers, which hinders the maintenance of uniformity in pollutant concentration. A model of the field chamber was used in the EPA Meteorological Wind Tunnel to evaluate design modifications to reduce the ingress of ambient air. In addition, the effects of the chamber on the mean velocity and turbulence structure of the boundary layer were assessed. Finally, the relationship between chambers when they are placed in an array was examined. It was determined that a cone top baffle which preserves between 50 and 75% of the original chamber open area is a good compromise between maintaining openness and pollutant uniformity. Field observations indicate, and tunnel results confirm, that pollutants from upwind chambers will affect pollutant levels in downwind chambers.

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A PHASEOLOTOXIN-LIKE SUBSTANCE PRODUCED BY *PSEUDOMONAS SYRINGAE* PV. *GLYCINEA*. S. S. Gnanamanickam, A. N. Starratt and E. W. B. Ward. Research Centre, Agriculture Canada, University Sub P.O., London, Ont., Canada N6A 5B7.

Several distinct symptoms develop in soybeans infected with *Pseudomonas syringae* pv. *glycinea*. These include localized and systemic chlorotic mottling of leaves, stunting of plant growth and the formation of elongated leaves. Previously, it was shown that strains causing systemic symptoms produced coronatine. Symptoms of chlorotic mottling, however, could not be reproduced by application of coronatine. Small quantities of a second active principle have now been demonstrated in culture fluids of pv. *glycinea* strains and this material induces chlorotic mottling when applied to soybean leaves. Unlike coronatine, this toxin inhibits growth of *Escherichia coli* K-12, and the toxicity is reversed by arginine and tripeptides. It therefore has some of the characteristics of phaseolotoxin, produced by pv. *phaseolicola*.

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EVIDENCE THAT SYRINGOMYCIN, PRODUCED BY *PSEUDOMONAS SYRINGAE* PV. *SYRINGAE*, IS A FERRIC SIDEROPHORE. Dennis C. Gross, Dept. of Plant Pathology, Washington State Univ., Pullman, WA 99164-6430.

Syringomycin, produced by *Pseudomonas syringae* pv. *syringae*, is a non-host specific phytotoxin that is essential for pathogenicity. Evidence indicates that syringomycin is a ferric siderophore since its biosynthesis is regulated by iron, it has compositional similarities to siderophores of the hydroxamate type, and it appears to supply iron to the cell. Unlike most siderophores, available iron is essential for production of biologically active syringomycin. Production of syringomycin in deferrated potato dextrose broth is proportional to the Fe³⁺ concentration between 0.1 μM and 2 μM; higher levels of Fe³⁺ do not stimulate nor decrease production. Syringomycin appears to counteract iron deprivation of *P. syringae* cells, induced by the Fe³⁺-specific chelator ethylenediamine di-(o-hydroxyphenylacetic acid) (EDDA). Therefore, syringomycin may have a dual role in plant disease: promoting disruption of host cells and supplying iron in a form that can be assimilated by the pathogen to promote further growth.

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MEMBRANE FLUIDITY CHANGES INDUCED BY THE PHOTSENSITIZING TOXIN, CERCOSPORIN. Margaret E. Daub¹ and Steven P. Briggs², Departments of ¹Crop and Soil Sciences and ²Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824.

Cercosporin, a toxin from *Cercospora* species, rapidly kills plant cells when they are incubated in the light. Electron spin resonance spectroscopy with two fatty acid spin labels was used to study the mode of action of cercosporin on tobacco protoplasts. Fluidity of the protoplast membranes was decreased by cercosporin in the light. The rotational correlation time, a measure of membrane microviscosity, increased from 1.7 x 10⁻⁹ sec for untreated protoplasts to 4.4 x 10⁻⁹ sec for cercosporin-treated protoplasts. Cercosporin-treated protoplasts also showed an increase in the membrane phase-transition temperature, from 12.7 ± 1.1°C in the control to 20.8 ± 1.2°C in the treated protoplasts. Loss of unsaturated acyl chains decreases membrane fluidity; thus, these data support the hypothesis that cercosporin kills cells by causing the peroxidation of the membrane lipids.

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PURIFICATION AND PARTIAL CHARACTERIZATION OF *HELMINTHOSPORIUM CARBONUM* TOXIN. L.M. Ciuffetti, L.D. Dunkle, M.R. Pope, H.M.

Knoche, and J.M. Daly. Dept. of Botany and Plant Pathology, Purdue University, W. Lafayette, IN 47907 and Dept. of Agricultural Biochem., University of Nebraska, Lincoln 68583. Purified toxin from *H. carbonum* race 1 inhibited root growth of susceptible maize genotypes at 175 ng/ml (EC_{50}) but did not induce ion leakage from seedlings until 14-16 hr of treatment with 5-10 μ g/ml. An empirical formula of $C_{21}H_{32}N_4O_6$ and m/z of 436 were obtained by peak matching with chemical ionization-MS. Alanine and proline (2:1) account for 55% of the mass of the toxin; the remainder of the cyclic peptide is comprised of an unknown amino acid with the formula $C_4H_8NO_2$. Selective activity was reduced when toxin was stored in water at 4 C for long periods of time, exposed dry to air, or incubated at low pH. The decrease in toxicity was associated with the appearance of a second peak in HPLC which usually accounted for ca. 30% of the original quantity of toxin. However, other properties (reaction with bromocresol green, IR spectrum, pmr spectrum, and amino acid composition) of this product were very similar to those of the toxin.

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STRUCTURAL DETERMINATION OF HELMINTHOSPORIUM CARBONUM TOXIN. M.R. Pope, H.W. Knoche, L.M. Ciuffetti, L.D. Dunkle and J. M. Daly. Dept. of Agricultural Biochemistry, University of Nebraska, Lincoln, NE 68583 and Dept. of Botany and Plant Pathology, Purdue University, W. Lafayette, IN 47907. FAB-MS, CI-MS, and EI-MS confirmed a molecular weight of 436 and empirical formula of $C_{21}H_{32}N_4O_6$ for the toxin. ^{13}C -NMR yielded 21 signals and indicated the presence of 4 amide carbonyls, one ketone and groups consistent with an epoxide and a cyclic peptide containing one unusual ten-carbon amino acid, one proline and two alanine residues. Derivatives of the toxin were prepared by reduction of the keto-group with $NaBH_4$, opening the epoxide ring with HOAC, and acetylation of hydroxyl groups produced by these derivatization methods. MS-MS analysis of the toxin and three derivatives established the order of amino acids in the cycle and that the unusual amino acid contained a terminal epoxide with an adjacent ketone function. Proton-NMR with decoupling experiments allowed complete assignments of protons for the toxin. The stability of the toxin appears to be due to the structural features of the unusual amino acid.

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CHARACTERIZATION OF LOWER HOMOLOGUES OF HOST-SPECIFIC TOXINS FROM HELMINTHOSPORIUM SACCHARI. V. Macko, C. Grinnalds, J. Golay, Boyce Thompson Institute at Cornell University, Ithaca, NY 14853; and D. Arigoni, W. Acklin, F. Weibel, C. Hildebrand, Dept. Organic Chemistry, Swiss Federal Institut of Technology, CH-8092 Zürich.

The three isomeric host-specific toxins from *H. sacchari* consist of two 5-O- β -galactofuranosyl- β -galactofuranose units attached to a sesquiterpene aglycone. The three isomers differ only in the position of one double bond in the aglycone. We have now detected twenty one new glycosides in the culture filtrate of the fungus. NMR and mass spectra clearly show that these new compounds are lower homologues of the toxins consisting of 6 monoglycosides, 9 diglycosides and 6 triglycosides. We will discuss their mode of formation and will present bioassay data comparing the activity of these homologues to that of the toxins.

100

RATE OF VICTORIN-INDUCED DEATH OF ISOLATED ROOT CAP CELLS AND PROTOPLASTS FROM OATS. Martha Hawes, Plant Path. Dept., U. of KY, Lexington.

Cell walls of outer root cap cells were resistant to enzymatic degradation; but it was possible to isolate protoplasts from inner cells, which exhibited the same sensitivity to victorin as outer cells. It was thus possible to compare the rate of victorin-induced death of isolated cells, with or without walls, which had been exposed to identical conditions. Each root cap yielded 20-50 protoplasts, of which 80-90% were alive (as judged by fluorescein diacetate retention). Eighty-five to 90% of enzyme treated cells were viable compared with 98-99% of cells incubated in 0.6 M sorbitol alone. Plasmolysis inhibited victorin-induced death, and at 25°C protoplast death began only after 20 hours in 50 units/ml toxin. At 35°C, 50% of plasmolyzed cells and protoplasts were killed by 20 units/ml after 10 hours; 50 to 150 units/ml killed all protoplasts within 6 hours. There was no difference in rate of death of cells with or without walls, therefore the cell wall is probably not involved in toxin mode of action.

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RESPONSE OF OAT PROTOPLASTS TO HELMINTHOSPORIUM VICTORIAE TOXIN.

942 PHYTOPATHOLOGY

S. P. Briggs and R. P. Scheffer, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824

Protoplasts from oat mesophyll were exposed to HV-toxin and observed using light microscopy with and without Evans blue and neutral red stains. Protoplasts from toxin-sensitive plants did not appear to be affected by toxin. If the temperature was raised to 35 C, untreated protoplasts or those from toxin-resistant plants remained unaffected but toxin-treated protoplasts from sensitive plants collapsed within 3 h. However, use of the vital stain fluorescein diacetate revealed that at 23 C, 90% of the treated protoplasts from sensitive plants were killed within 5 h, even though they appeared to be normal. Preliminary results indicate that vacuoles isolated from protoplasts may also be sensitive to toxin, as indicated by fluorescein diacetate and microscopic examination. The results of further tests of vacuole sensitivity will be reported.

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PHYTOTOXIN PRODUCTION BY ALTERNARIA TAGETICA. P.J. Cotty, I.J. Misaghi, and R.B. Hine. Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

Alternaria tagetica causes a destructive stem, leaf, and flower blight of marigold in Northwestern Mexico where the crop is important. The fungus produces metabolites *in vitro* capable of inducing symptoms on marigold leaves similar to those caused by infection. Methanol extracts of dried cell-free culture filtrates were phytotoxic when assayed on excised pricked leaves. The dilution end-point of this fraction was between 1:25 and 1:50. Phytotoxicity increased with increasing temperature from 27 C to 37 C. This fraction was also phytotoxic to a number of non-hosts including sunflower, zinnia, cotton, okra, cucumber, wheat and tomato. Chloroform extracts of the filtrates caused wilting and darkening of the tissue around veins in excised marigold plants similar to that reported for zinnia, a chloroform soluble phytotoxin of *A. zinniae*, *A. dauci*, and *A. solani*. The identity of the toxin(s) and its possible involvement in pathogenesis are being investigated.

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SCREENING MAIZE SINGLE CROSSES FOR RESISTANCE TO PREHARVEST INFECTION OF KERNELS BY ASPERGILLUS FLAVUS. S. B. King and G. E. Scott, USDA-ARS, P. O. Drawer PG, Mississippi State, MS 39762.

Fifty-four single crosses representing 40 inbreds were screened for kernel infection by *A. flavus* in 1980-81. Ears were inoculated 20 days after the mid silk stage by pressing a pinbar (single row of sewing pins mounted in plastic) contaminated with *A. flavus* conidia through the husk and into a row of kernels. After harvest, kernels in the first and second rows of kernels adjacent to the inoculated row were shelled separately, surface-sterilized, plated on NaCl-amended Czapek solution agar and incubated at 28 C for 1 wk. Significant differences were found among hybrids, with mean *A. flavus* kernel infection frequencies ranging from 3% for Mp412 X Mp444 to 64% for Mp414 X Mp68:616. Reactions of many hybrids were consistent over the 2 yr of testing. For example, mean infection frequencies for the seven lowest and seven highest hybrids were 2 and 45%, respectively, in 1980 and 10 and 28%, respectively, in 1981. Mean kernel infection frequencies of inbreds estimated from data of hybrid response in five or more crosses, ranged from 8 to 27%.

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INCIDENCE OF RACE 2 OF HELMINTHOSPORIUM TURCICUM IN THE MIDWEST. E. G. Jordan, J. M. Perkins, R. A. Schall and W. L. Pedersen. Department of Plant Pathology, University of Illinois, Urbana, IL 61801 and USDA-APHIS, Urbana, IL and West Lafayette, IN 47901.

Samples of northern corn leaf blight tissue were collected during surveys of corn seed production and commercial grain fields in Illinois and Indiana in 1979, 1980 and 1981 and in Minnesota in 1981. Races of *H. turcicum* were determined using the corn inbreds B37, B37Ht1, B37Ht2 and Oh43Ht3. Race 2, virulent on plants carrying the resistance gene Ht1 and avirulent on plants carrying the Ht2 or Ht3 genes, was confirmed for 24 counties in Indiana and 53 counties in Illinois. These surveys show that race 2 is widespread in both states. A sample from Wright County, Minnesota, also was identified as race 2. This is the first report of race 2 from Minnesota. In addition, samples tested from New York, Ohio and Pennsylvania also were race 2.

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AN INOCULATION TECHNIQUE TO DETECT THE HtN GENE IN INBRED LINES OF CORN. S. Leath and W. L. Pedersen, Department of Plant

The effect of the resistance gene, *HtN*, against *Helminthosporium turcicum*, is characterized in the field by an increased latent period; however, susceptible lesions later develop. A technique was developed to identify the presence of *HtN* in seedlings of corn inbred lines and in segregating populations. A 10 μ l drop of inoculum containing approximately 400-500 conidia (40,000-50,000 conidia/ml) of *H. turcicum*, was applied to each adaxial leaf surface of 11-day-old corn seedlings. Ten days after inoculation, susceptible plants had typical water-soaked lesions while seedlings with the *HtN* gene remained symptomless. Four different isolates of race 1 and one isolate of race 2 were tested, and the technique was consistently effective in identifying the resistant seedlings. This technique also proved effective in characterizing known segregating populations.

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REACTION OF CORN SINGLE CROSSES TO GRAY LEAF SPOT. J. E. Ayers, R. R. Hill, Jr., and M. W. Johnson, Jr., Department of Plant Pathology and Agronomy, The Pennsylvania State University and USDA-ARS, University Park, PA 16802

Two replications each of four corn (*Zea mays*) single cross tests were planted on May 8, 1981, no-till, in a field that had been in continuous corn for several years and in which gray leaf spot (*Cercospora zeae-maydis*) severity had been high the two previous seasons. Inbreds used in the crosses consisted of released public and experimental lines. Disease severities were determined on August 19 (soft dough) and September 7 (late dough-early dent). Analysis of variance procedures demonstrated significant differences for disease reaction among single crosses in each test. Further partitioning of the sum of squares for single crosses allowed a test for the significance of males adjusted for females not present in all crosses and for females adjusted for males. The inbreds Pa887P, B68, Va59, and several Pennsylvania experimental lines consistently demonstrated resistance to gray leaf spot.

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EVALUATION OF MAIZE HYBRIDS AND INBREDS FOR RESISTANCE TO SPHACELOTHECA REILIANA. E. L. Stromberg, Department of Plant Pathology & Physiology, VPI & SU, Blacksburg, VA 24061, T. Kommedahl, W. Stienstra, C. E. Windels, and C. Matyac, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Maize hybrids (161) and inbreds (14) were screened in the field for resistance to head smut caused by *Sphacelotheca reiliana*. All entries were planted at three dates within a 30 d period. Although seeded into naturally infested soil, entries were jab-planted and 120 ml of an artificially infested soil (200:1, v/v, soil to *S. reiliana* teliospores) was placed over each seed. At maturity, incidence of smutted ears, tassels, ears and tassels, partially smutted ears, ear proliferation, and dwarfness was recorded. Resistance was expressed as incidence of head smut as a grand mean percentage for the three planting dates (0%, resistant; >0-5%, moderately resistant; >5-10% moderately susceptible; >10%, susceptible). Eight entries were resistant, 111 moderately resistant, 34 moderately susceptible, and 22 susceptible.

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INHERITANCE OF RESISTANCE TO SORGHUM DOWNY MILDEW IN CORN J. Craig, USDA-ARS, Plant Sciences Dept., Texas A&M University College Station, TX 77843

The inheritance of resistance to sorghum downy mildew was studied in the F_1 , F_2 , F_3 and backcross progenies from a cross of the resistant corn inbred Tx601 and the susceptible N28. *Peronosclerospora sorghi*, causal agent of the disease, was unable to colonize inoculated leaves of Tx601. In Tx601 the chlorosis of leaf tissue caused by pathogen colonization was restricted to small spots surrounding stomata invaded by conidia. In the susceptible N28, the pathogen induced wide spread chlorosis of the inoculated leaves. Reactions of the inoculated leaves of the test populations indicated that susceptibility was partially dominant and that resistance to *P. sorghi* was conditioned by two linked genes.

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A NEW SOURCE OF RESISTANCE TO SOYBEAN MOSAIC VIRUS IN A SOYBEAN LINE AND ITS INHERITANCE. S. M. Lim, USDA-ARS and Department of Plant Pathology, University of Illinois, Urbana, 61801.

A new source of resistance to all known seven strains of soy-

bean mosaic virus (SMV) was found in Suweon 97 soybean obtained from the Crop Experiment Station, Suweon, Korea. Reactions of this line to each of the seven SMV strains were symptomless in greenhouse tests under artificial inoculations. Soybean cultivars, Williams and Franklin, susceptible to the seven SMV strains, were used as the female in crosses with Suweon 97 to determine the inheritance of resistance to SMV. Parental, F_1 , F_2 , and F_3 populations of these crosses were inoculated in the greenhouse with two strains of SMV: G-2 (common) and G-7 (severe). Segregation ratios indicated that resistance to each of the two strains was conditioned by a single dominant gene.

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RACE NON-SPECIFIC RESISTANCE IN BEAN VARIETIES TO RUST AND ITS ASSOCIATION WITH SOME LEAF EPIDERMAL CHARACTERS. Meher Shaik, Botany Department, University of the West Indies, Mona, Jamaica.

Race non-specific resistance of five varieties of beans (*Phaseolus vulgaris* L.) to three rust races (*Uromyces appendiculatus* (Pers.) Unger var. *appendiculatus*, races J4, J10, and J15) was investigated. Disease intensity (pustules/cm²), considered as a measure of race non-specific resistance, was significantly different between the varieties. Furthermore, successive leaves on a plant -inoculated at comparable developmental stages- showed increasing resistance. Varieties most likely to possess race non-specific resistance were identified. In a separate experiment epidermal characters of fully expanded leaves of each variety were examined. Resistant varieties had lower stomatal density and higher hair density than the susceptible varieties. Similarly, increased resistance of upper leaves was associated with lower stomatal density and higher hair density than on the more susceptible lower leaves.

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DECREASING INCIDENCE AND SEVERITY OF BROWN STEM ROT AND INCREASING YIELDS BY CONTINUOUS CROPPING OF RESISTANT SOYBEAN. H. Tachibana, A. H. Epstein, and J. D. Hatfield. USDA-ARS, Department of Plant Pathology, Seed and Weed Science, Iowa State Univ. Ames, IA 50011

Incidence and severity of brown stem rot (BSR) of soybean was progressively reduced by continuous cropping with resistant soybean. BSR incidence remained high and severity fluctuated with continuous cropping with susceptible soybean. Yield increase averaged 17% for the four susceptible cultivars (Coles, Weber, Hardin and Pride B216) on land that had been cropped continuously with BSR resistant germplasm line A3 for four years. Higher yields were obtained also with four resistant soybeans, including A3. The study was conducted utilizing maturity group I and II cultivars at Kanawha in north central Iowa. The test land was heavily infested with *Phialophora gregata* at the start of the experiment. Our conclusion is - current BSR resistant soybeans have immediate as well as long term benefits for disease control and higher yields.

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CHANGES IN THE ISOFLAVONOID CONSTITUENTS OF SOYBEAN CELL CULTURES ON EXPOSURE TO FUNGAL ELICITOR. R. M. Zacharius, E. B. Kalan and P. Ripa, Eastern Regional Research Center, USDA, ARS, 600 E. Mermaid Lane, Philadelphia, PA 19118

Changes in isoflavonoid content of soybean cv. Mandarin cell suspension cultures were studied during interaction with a fungal elicitor of *Phytophthora infestans* using TLC and HPLC methods. Daidzein (I), genestein (II) and coumestrol (III) were the major isoflavonoids identified in the control cell cultures. The isoflavonoid level varied considerably for the same cell line depending on culture age, light and probably cell aggregate size. Darkening of the cell cultures occurred on exposure to elicitor but there was no evidence of cell death. Within 12 hours, glyceollin accumulated with a concomitant decline (40% or more) of I, II and III. Cell cultures that initially contained very low levels of I, II and III failed to produce glyceollin when exposed to elicitor.

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AN APPROACH FOR LABORATORY TESTING OF CERTIFIED DRY BEANS. H.F. Schwartz and D.E. Legard. Dept. of Botany and Plant Pathology, Colorado State University, Fort Collins, CO 80523.

A dry bean seed testing procedure is being developed by CSU and University of Nebraska scientists to provide laboratory verification of the pathology status of seed visually inspected by Colorado Seed Growers Association personnel in western Colorado. The test will eventually use serological methods to detect pathogens responsible for halo blight, common bacterial blight, and bean common mosaic virus. The procedure currently relies upon a modified North Dakota Dome seed-soak bioassay to test certi-

fied seed for internal contamination by bacterial pathogens. Surface sterilized seed lots are incubated in a dilute nutrient solution for 18-24 hours. Fresh seeds are vacuum-infiltrated with the solution and planted in moist vermiculite sealed within a clear plastic bag. Seedlings are then scored 12-13 days later for the presence or absence of bacterial symptoms on stems or leaves. Work is in progress to correlate lab results with disease development under favorable environmental conditions in the field.

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IMMUNOLOGICAL PROCEDURES FOR THE DETECTION OF *XANTHOMONAS PHASEOLI*. E. M. Malin, D. A. Roth and E. L. Belden, Department of Plant Science and Department of Microbiology and Veterinary Medicine, University of Wyoming, Laramie, WY 82071.

Xanthomonas phaseoli was detected in mixed culture and in artificially and naturally infected bean seed by immunofluorescent staining. Immunofluorescence was highly specific for over 40 *X. phaseoli* isolates from Wyoming, Nebraska and Michigan. Numerous other *Xanthomonas*, *Pseudomonas* and common bean seed bacterial contaminants did not cross-react with *X. phaseoli*-antisera. *Xanthomonas phaseoli* cells were detected in mixed populations containing 500 *X. phaseoli* cells/ml and 10^8 cells/ml of common bean seed contaminants. *Xanthomonas phaseoli* cells also were detected in naturally and artificially infected bean seed samples with *X. phaseoli* infection levels of 0.1%. Immunofluorescent staining when used with dilution plating of samples on a semi-selective medium provides a rapid and reliable method for the detection of *X. phaseoli* in dry bean seed.

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ASCOCHYTA LENTIS: INCIDENCE AND TRANSMISSION IN IMPORTED LENTIL SEED. W. J. Kaiser, and R. M. Hannan. USDA, ARS, Regional Plant Introduction Station, Washington State University, Pullman, WA 99164.

Ascochyta lentis was isolated from seeds of different exotic lentil accessions in the USDA plant germplasm collection maintained by the Western Regional Plant Introduction Station at Pullman, WA. Seeds of 186 introductions from 23 countries were surface-sterilized in 0.26% NaOCl for 5 min and plated on water agar. The fungus was isolated from seeds of 39 accessions that originated in Australia, Ethiopia, Greece, Hungary, Italy, Morocco, Soviet Union, Spain, Syria, and Turkey. Seed transmission ranged from 1.7 to 58.3% in infected accessions. The pathogen survived in infected seeds of several accessions for over 30 years at 4-8°C. Isolates of *A. lentis* varied greatly in growth rate, sporulation, and colony appearance. Although plants infected by *A. lentis* have not been observed in commercial lentil plantings in the Palouse region of eastern Washington and northern Idaho, commercial lentil varieties are susceptible to infection by different isolates of the pathogen.

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LOCATION EFFECTS ON SCREENING CORN KERNELS FOR INTERNAL FUNGI IN MISSOURI IN 1981. O.H. Calvert, A.S. Foudin, and H.C. Minor. Dept. of Plant Pathology; USDA, APHIS, PPQ; and Dept. of Agronomy, Univ. of Missouri-Columbia, Columbia, MO 65211, respectively.

Corn kernels from hybrids grown at eight different locations (State yield trials) were examined for internal fungi. Duplicate 100-kernel samples of sound, intact seed were surface sterilized in a Clorox solution (1:5, v/v) for 10 min, placed on potato-sucrose agar and incubated in the dark for 5 d at 28°C. Kernels surveyed from six sites north of the Missouri River produced approximately half of the numbers of fungal colonies as kernels from two southeast locations. The predominant species cultured during 1980 and 1981 was *Fusarium moniliforme*. The 1981 corn crop had little disease stress as a result of excellent growing conditions, yielding about double that produced in the previous 1980 season of severe drought. We observed very limited numbers of *Aspergillus flavus* (<0.1%) colonies and fewer colonies of all fungi than in kernels from the 1980 crop season.

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PREVALENCE OF THE CAUSAL ORGANISMS OF STEM CANKER AND POD AND STEM BLIGHT ON SOYBEAN PODS AND SEEDS. D. C. McGee, Dept. of Plant Pathology, Seed and Weed Sciences, Iowa State University, Ames, IA 50011.

Infection of soybean pods by *Diaporthe phaseolorum* var. *caulivora* (Dpc), the cause of stem canker, and *Phomopsis* sp. and *Diaporthe phaseolorum* var. *sojae* (Dps), the causal organisms of pod and stem blight, was measured in 15 soybean fields in Iowa in 1982. Pod infection averaged 8.8, 17.8, and 1.3% for Dpc, *Phomopsis* sp., and Dps, respectively, at the R8 stage of growth. The

relative amounts of *Phomopsis* sp. and Dpc infection varied greatly between fields, but were consistent for measurements made at different times within the same field. Seed infection, determined at harvest maturity, was negligible when the seed was plated directly on PDA. However, when detached pods were held at 98% relative humidity for 7 days before seeds were tested, seed infection averaged 25.8, 39.0, and 1.6% for Dpc, *Phomopsis* sp., and Dps, respectively, for all fields. The ratios of seed infection by the three fungi were well correlated with those for pod infection in the same field.

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THE INFLUENCE OF DIFFERENT INCIDENCES OF PHOMOPSIS-INFECTED SEED ON THE YIELD OF SOYBEANS. Steven B. Johnson and R. D. Berger, Department of Plant Pathology, University of Florida, Gainesville, FL 32611.

Soybean seeds, healthy and infected with *Phomopsis sojae*, were mixed manually to obtain treatments of 0, 0.1, 1, 10, and 100% diseased seeds. The seed were planted in four-row plots arranged in a randomized complete block design with four replications. Forage sorghum was used as a barrier between plots to minimize interplot interference. Visual ratings of the intensity of infection by *P. sojae* did not differ among the treatments. Based on regression analysis, seed yield decreased significantly with increased proportions of diseased seed in the planting mixture. Soybean seed yield in commercial fields would be expected to increase from planting disease-free seed.

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INTERACTION OF *CERCOSPORA SOJINA* AND *PHOMOPSIS SOJAE* ON SOYBEAN SEED QUALITY AND YIELD COMPONENTS. V. S. Bisht and J. B. Sinclair, Dept. of Plant Pathology, University of Illinois at Urbana-Champaign, IL 61801.

Soybean cultivars Corsoy 79 and Wells were noninoculated (N) or inoculated with race 10 A of *C. sojae* (C) or *P. sojae* (P) alone or in combination (CP). Leaf area at growth stage R₆ was significantly ($P=0.05$) reduced by C and CP below N and correlated with 1000 seed weight ($r=0.60$). Yield was higher, but not significantly, with CP compared to C. Both C and P reduced seed germination, but only C reduced seedling vigor. Both C and P reduced % clean seeds and CP significantly reduced % clean seeds below either alone. Total number of fungi recovered from seeds was correlated to C ($r=0.77$) and to P ($r=0.51$). Fungi significantly reduced seed germination ($r=-0.77$). C and P act independently and have an additive effect on deterioration of seed quality.

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BACTERIZATION OF PEANUT SEED WITH *BACILLUS SUBTILIS*. P. A. Backman, R. P. Clay, and M. A. Crawford. Dept. Botany, Plant Pathology and Microbiology, Auburn University, AL 36849.

Field trials in 1980 and 1981 supported by germinator studies have indicated that peanuts (*Arachis hypogaea*) exhibit significantly improved rates of emergence and sustained improvements in plant vigor when treated with *Bacillus subtilis* (Abbott, ABG-4000). Evaluation of seeds and seedlings indicate that the probable mechanisms involved in plant stimulation are: 1) hormonal stimulation of germination that afforded probable disease escape; and 2) antibiotic production that reduced seed and seedling disease severity. Color and antibiotic markers indicate that the strain can survive until harvest on the rhizoplane at $> 10^5$ CFU's/g fresh roots. *Bacillus subtilis* was compatible with all common peanut seed treatment fungicides and Rhizobium nodulation was significantly improved. Although not significant, bacterization with *B. subtilis* resulted in an 18% yield increase.

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IMPROVED ASSAY MEDIA FOR ISOLATING *XANTHOMONAS CAMPESTRIS* PV. *CAMPESTRIS* FROM CRUCIFER SEEDS. P. S. Randhawa and N. W. Schaad, Dept. of Plant Pathology, University of Georgia, Georgia Experiment Station, Experiment, GA 30212.

Crucifer seed lots contain numerous saprophytic and antagonistic bacteria which often interfere with the isolation of *X. campestris* on such non-selective differential media as nutrient starch cycloheximide agar (NSCA). Adding nitrofurantoin and vancomycin at 20 and 1.0 µg/ml, respectively, to NSCA (= NSCAA) reduced significantly the numbers of saprophytic and antagonistic bacteria in 10 commercial seed lots tested without

reducing the recovery of *X. campestris*. Adding the same antibiotics at 10 and 1.0 µg/ml to a modified basal starch cycloheximide agar [(BSCA, Phytopathology 64:876-880) (= BSCAA)] resulted in a further significant reduction of the saprophytic microflora. However, BSCAA reduced significantly the growth and recovery of one of 10 strains of *X. campestris* tested. The use of NSCA together with the two selective media should increase the recovery of *X. campestris* in commercial seed testing.

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AIRBORNE DISPERSAL OF XANTHOMONAS CAMPESTRIS P.V. CAMPESTRIS
T.-L. Kuan, G.V. Minsavage, N.W. Schaad, P.O. Box 1, Asgrow Seed Company, San Juan Bautista, CA 95045, and University of Georgia, Georgia Experiment Station, Experiment, GA 30212.

Xanthomonas campestris p.v. *campestris* causes black rot of crucifers. The seed-borne phase of this pathogen is considered one of the primary inoculum sources. Other inoculum sources have received limited study. The role of dispersal of the pathogen has not been determined. Crucifer weeds are known to be a reservoir of *X. campestris* p.v. *campestris* in the Salinas Valley, CA (Phytopathology 71: 1215-1220). To determine the importance of weeds as a reservoir of inoculum, and the role of air-borne dispersal, we obtained air samples with Andersen air sampler and selective medium. We isolated the pathogen from the air at 0.002 cell/l near the crucifer weeds (*Brassica campestris* and *B. nigra*) in Salinas, California. Further studies are being conducted to investigate the nature of air-borne dispersal of the pathogen.

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RELATIONSHIP OF MOLECULAR WEIGHT AND VISCOSITY TO WILT-INDUCING PROPERTIES OF ERWINIA AMYLOVORA-EPS. K. Sijam, A.L. Karr, and R.N. Goodman. Department of Plant Pathology, University of Missouri, Columbia, MO 65211.

The extracellular polysaccharide (EPS-amylovorin) produced by *E. amylovora* is a large poly-anion. While it is antigenically identical to lipopolysaccharide, it is known not to contain LPS since the molecule bearing the antigenic determinants is hydrolyzed by an EPS specific depolymerase phage. EPS is capable of causing wilt in apple shoots. When the viscosity of EPS solutions and the molecular weight of EPS is reduced by treatment with the depolymerase phage, the capacity to cause wilt is not lost. The viscosity of EPS solutions, but not the molecular weight of EPS is reduced with increasing the ionic strength of the solution. This effect is independent of the ionic species (cationic or anionic) employed. When the viscosity of EPS solutions is decreased in this fashion, the ability to cause wilt is lost. The results suggest that the interaction between these large poly-anionic molecules is critical to their ability to cause wilt in cut apple shoots.

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CAPSULAR DEPOLYMERASE FROM BACTERIOPHAGE INFECTED ERWINIA AMYLOVORA. J.S. Hartung, P.B. Rosenthal, D.W. Fulbright and E.J. Klos, Department of Botany & Plant Pathology, Michigan State University, East Lansing, MI 48824

E. amylovora infected with bacteriophage PEal(h) was shown to produce an enzyme which degrades the extracellular polysaccharides (EPS) of non-infected cells, without killing them. The enzyme was partially purified, to study the possible role of EPS as a factor in pathogenicity and as a barrier to uptake of antibiotics. EPS was isolated by ethanol precipitation from *E. amylovora*-infected green pear fruits and used as substrate for enzyme assays. In partially purified cell lysates the enzyme has a pH optimum of 5.0; activity is enhanced by a high molarity buffer. The enzyme reversibly binds to DEAE cellulose at pH 7.3. PEal(h) produces clear plaques when plated with *E. amylovora* in soft agar overlays; however, PEal(h) fails to completely lyse cultures of *E. amylovora* grown in either rich or minimal broth. PEal(h) has a double stranded DNA genome which is cleaved into 3 fragments by restriction endonuclease Eco RI.

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CHARACTERIZATION OF THE EXTRACELLULAR POLYSACCHARIDE FROM CORYNEBACTERIUM INSIDIOSUM. N. K. Van Aifen, B. D. McMillan, and P. Dryden, Department of Biology, UMC 45, Utah State University, Logan, Utah 84322.

Corynebacterium insidiosum (McCulloch) Jensen, when grown on any of several media in shake culture produces at least three size classes of soluble high molecular weight polysaccharides. The two larger size classes have the same neutral sugar composition (fucose:glucose:galactose; 2:1:1), and are both retained on DEAE cellulose equilibrated with 10mM sodium acetate buffer,

pH 5. The larger, near the void fraction on Sepharose Cl-2B, will disassociate into the smaller (ca. 5 million MW) upon boiling or treatment with SDS, but is unaffected by 8M urea. It is associated with a small amount of protein, while the smaller is not. The smallest of the size classes (ca. 50,000 MW) will not bind to DEAE cellulose under the above conditions, and differs in neutral sugar composition (galactose:mannose:fucose; 3:1:1). Molecular sizes may be important in determining at which sites vascular blockage may occur in the host.

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LOCAL CONCENTRATIONS OF TWO PHYTOALEXINS AT SITES OF BACTERIAL COLONIES IN COTTON LEAVES. M. Essenberg, E.C. Cover, M.L. Pierce, P.E. Richardson, Depts. of Biochemistry and Botany, Oklahoma State Univ., Stillwater, OK 74078; V.E. Scholes, Dept. of Microbiology/Immunology, Oral Roberts Univ., Tulsa, OK 74171; B.K. Hamilton, Dept. of Plant and Soil Science, Texas Tech Univ., Lubbock, TX 79410.

In near-isogenic susceptible and immune lines of cotton inoculated with *Xanthomonas malvacearum* pv. *malvacearum* (Xcm), yellow-green fluorescent material with an emission spectrum similar to the spectra of the phytoalexins lacinilene C (LC) and lacinilene C 7-methyl ether (LCME) was detected at clusters of brown palisade cells. LC and LCME constituted 60% of the yellow-green fluorescent substances in acetonitrile extracts of inoculated immune leaves. We concluded that LC and LCME were localized in the fluorescent cells. Concentrations of LC and LCME at the fluorescent cells were calculated using total extractable quantities from leaves and the percentages of leaf cells containing yellow-green fluorescence. When bacterial multiplication stopped in immune plants, the calculated local concentrations of LC were high enough in immune plants, but not in susceptible plants, to be at least partially inhibitory to Xcm.

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GEORGIA PECAN TREES WITH CROWN GALL YIELDED MAINLY AGROCIIN SENSITIVE BIOVAR 1 STRAINS OF AGROBACTERIUM TUMEFACIENS. H. Bouzar, and L. W. Moore, Oregon State University, Corvallis, 97331, and N. W. Schaad, University of Georgia, Experiment, 30212.

Georgia accounts for about 50% of the total pecan production in the U.S., and crown gall is a major concern. Disease incidence in 40-60 year old pecan orchards is 25 to 60%. *Agrobacterium* strains were isolated from 18 galled trees located in orchards in several counties. According to standardized diagnostic tests, biovar 1 strains were isolated on Schroth et al. medium, whereas most of the biovar 2 strains were isolated on the New and Kerr medium. Virulent strains of both biovars were isolated from the same gall in 4 of the 18 samples, but biovar 1 strains accounted for 70% of the pathogens. Most of them were sensitive to agrocin 84, a bacteriocin produced *in vitro* by *A. radiobacter* strain K84. A representative biovar 1 strain was also inhibited from infecting tomato seedlings by *A. radiobacter* strain K84. Therefore, we would expect that biological control of crown gall of pecan in Georgia has a good chance of success.

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TAGETITOXIN PRODUCTION BY PSEUDOMONAS SYRINGAE PV. TAGETIS. D. J. Styer and R. D. Durbin, Department of Plant Pathology, ARS, USDA, University of Wisconsin-Madison, WI 53706.

Tagetitoxin, a toxin produced by *Pseudomonas syringae* pv. *tagetis* in culture, causes apical chlorosis of marigold and zinnia. Similar apical chlorosis and ultrastructural appearance are observed in plants inoculated with this bacterium suggesting that the same toxin is produced in both cases. Although all 10 strains of the pathogen isolated in the Netherlands, Zimbabwe, Australia or the U.S. cause chlorosis of plants, only the Australian strain produces tagetitoxin *in vitro*. In Woolley's medium containing 110 mM glucose, toxin production occurs at 50 mM but not at 10 mM nitrogen. Production occurs at both 20 and 28 C and requires high aeration; it does not occur in nutrient broth. Commonly, single cell variants of the Australian strain can be isolated that lack the ability to produce tagetitoxin *in vitro* but still produce tagetitoxin *in planta*. Plasmids isolated from both cell types have identical migration patterns on agarose gels.

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CHARACTERIZATION OF A BACTERIOCIN FROM ERWINIA HERBICOLA STRAIN C9-1. C. Ishimaru, R. R. Brubaker, and E. J. Klos, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824

Erwinia herbicola strain C9-1, isolated in Michigan from blighted apple tissue, produced a bacteriocin (herbicocin C9-1) active against all tested isolates of *E. amylovora*,

Escherichia coli, and *Yersinia* spp. Bacterial isolates from 18 other genera were insensitive to herbicolin C9-1. All sensitive isolates yielded spontaneous insensitive mutants. Insensitive mutants of *E. coli* 9, a standard colicin indicator, were tolerant to group B colicins. Herbicolin C9-1 was dialyzable and heat stable (95°C for 10 min); activity against *E. coli* in cross-streak assays was reduced by trypsin. The bacteriocin was not significantly induced by ultra-violet light or radiomimetic agents. Bacteriocinogenic strains of *E. herbicola* have been proposed as biological control agents of fire blight. Such strains are common in Michigan.

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A NEW BIOASSAY FOR CORONATINE PRODUCTION BY *P. SYRINGAE* PV. *GLYCINEA*. J. W. Willis and J. V. Leary. Department of Plant Pathology, University of California, Riverside, CA 92521.

There are nine known physiological races of the soybean pathogen *Pseudomonas syringae* pv. *glycinea* (*P. glycinea*), which were identified on the basis of differential host/pathogen interactions using indicator soybean cultivars. Strains of some races produce a chlorosis-inducing phytotoxin, coronatine. Investigation of the use of coronatine production as a basis for identification of *P. glycinea* and of the genetics of toxin production have been hampered by the lack of a convenient, rapid bioassay. We screened 21 different species of gram-positive and gram-negative bacteria, including phytopathogens, human pathogens, and saprophytes for sensitivity to coronatine. Only *Micrococcus* (*Sarcina*) *luteus* demonstrated growth inhibition in the presence of the toxin and the inhibition was quantifiable. Thus, a convenient bioassay for production of coronatine is now available.

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DETECTION OF BACTERIOCIN-LIKE SUBSTANCES PRODUCED BY *XANTHOMONAS CAMPESTRIS*-PV *ORYZAE*. T. W. Mew, J. S. Huang and E. Echandi. The International Rice Research Institute, Los Banos, Philippines and North Carolina State Univ., Raleigh, NC 27650.

Twenty-two strains of *X. campestris* pv. *oryzae* produced bacteriocin-like substances on solid media. Most of the 22 strains produced inhibition zones less than 2 mm from colony margins, against pv. *oryzae* indicator strains, and 4 to 6 mm against pv. *vasculorum* strains. Bacteriocin-like substances active against pv. *oryzae* strains were also produced by *X. campestris* pv. *campestris* and pv. *vasculorum* strains. The pv. *oryzae* bacteriocin-like substances were detected on six media tested but production was the highest on nutrient agar amended with 15 g/l sucrose. Inhibition zones were larger at 20°C than at 28°C, the optimum temperature for bacterial growth. The bacteriocin-like substances were heat-stable, sensitive to trypsin and protease, and they were not induced by UV light and mitomycin C. One µg/ml mitomycin C completely suppressed bacterial growth. The bacteriocin-like substances were precipitated by $(\text{NH}_4)_2\text{SO}_4$.

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LOCALIZATION AND QUANTIFICATION OF ICE NUCLEI AND ICE NUCLEATION ACTIVE BACTERIA ASSOCIATED WITH DORMANT AND GROWING PEAR TISSUE. D. Haelele and S. E. Lindow, Dept. Plant Pathology, University of California, Berkeley, CA 94720.

Spatial distribution of bacteria on plants was determined by their sensitivity to topical application of 50 mM CuSO_4 . The surface and internal populations of total, and ice nucleation active bacteria, and ice nuclei active at -5°C or -9°C were determined for pear buds, twigs, flowers and or leaves during dormancy, flowering and leaf expansion. Dormant twigs contributed ice nuclei and had a warmer mean supercooling temperature (MST) than buds before flowering. Viable ice nucleation active bacteria were not always found on dormant tissue containing ice nuclei active at -5°C. Populations of total bacteria increased on the surface and decreased internally at bud break. Concomitantly, ice nucleation active bacteria reached 1.5×10^2 cfu/g on the surface and 5.6×10^3 cfu/g internally. The MST of pear twigs treated with methyl benzethonium hydroxide and CuSO_4 was -4.2°C and -4.9°C respectively compared with -2.4°C for untreated twigs. Extensive washing of twigs reduced the MST to -3.8°C. It appears that ice nuclei on dormant pear are superficial and are probably bacterial in origin.

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INTEGRATED CONTROL OF FROST INJURY AND FIRE BLIGHT OF PEAR WITH ANTAGONISTIC EPIPHYTIC BACTERIA. S. E. Lindow, Dept. of Plant Pathology, University of California, Berkeley, CA 94720.

Epiphytic populations of ice nucleation active strains of *Pseudomonas syringae* pv. *syringae* and *Erwinia herbicola* as well as *Erwinia amylovora* on leaves and flowers of Bartlett pear were reduced significantly on trees treated at 10% bloom with 14 of 16 non-ice nucleation active antag-

onistic bacteria. Antagonistic bacterial populations on treated trees ranged from ca. 10^4 cells/g fresh weight to over 10^6 cells/g fresh weight for eight weeks following inoculation, comprising up to 70% of the total bacteria on pear surfaces. Frost injury to small pear fruit in a mild radiative frost (minimum air temperature -3°C) was reduced up to 89% on trees treated with certain antagonistic bacteria. The incidence of fire blight on fruiting spurs of pear one month following freezing temperatures was also reduced up to 64% on trees treated with antagonistic bacteria. The incidence of either frost injury or fire blight on trees treated with most antagonistic bacteria did not differ significantly from trees treated at 10 day intervals with a mixture of streptomycin and oxytetracycline.

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THE INVOLVEMENT OF ICE NUCLEATION-ACTIVE BACTERIA IN FROST INJURY TO FRUIT TREES: DISTRIBUTION, POPULATION DYNAMICS, AND CHARACTERISTICS OF THE NUCLEATOR. D. C. Gross, Y. S. Cody, E. L. Proebsting, Jr., G. K. Rademaker, and R. A. Spotts*. Wash. St. Univ., Pullman, WA 99164; *Irr. Agr. Res. Ext. Ctr., Prosser, WA 99350; and **Mid-Columbia Expt. Sta., Hood River, OR 97301.

Frost sensitivity of fruit tree floral tissues is largely dictated by the number and nucleation frequencies of ice nucleation active (INA) bacteria and the inherent ability of tissues to supercool and survive ice formation. In the Pacific Northwest populations of INA bacteria in pear, apple, and cherry orchards, during periods of frost susceptibility, range from undetectable to over 10^6 /g fr. wt. of tissue. These INA bacteria are efficient ice nuclei and aggressive colonizers of young vegetative and floral tissues. Populations peak around full bloom. All of the isolates of INA bacteria are *Pseudomonas syringae*. Most of the INA bacteria are pathogenic to pear, suggesting a relationship between frost injury and disease. INA bacteria appear to limit supercooling of flowering fruit trees; however, nucleation sites within the woody tissue also limit supercooling to similar temperatures ($\approx -3^\circ\text{C}$).

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COLONIZATION OF THE CIBARIA OF SHARPSHOOTER VECTORS, *ONCOMETOPIA NIGRICANS* AND *HOMALODISCA COAGULATA* BY XYLEM-INHABITING BACTERIA. R. H. Brlansky and L. W. Timmer, Univ. Fla., IFAS, AREC, 700 Experiment Station Road, Lake Alfred, FL 33850.

The sharpshooter leafhoppers, *Oncometopia nigricans* (Walker) and *Homalodisca coagulata* (Say), are vectors of the xylem-inhabiting bacteria which cause Pierce's disease (PD), phony peach disease (PPD), plum leaf scald (PLS) and periwinkle wilt (PW). These bacteria were observed in the cibaria, apodemal groove of the diaphragms and precibaria of these sharpshooters by scanning electron microscopy. Cibaria, precibaria and diaphragms of *O. nigricans* were colonized after insects had fed on PD-affected *Vitis vinifera* L. or PW-affected *Catharanthus roseus* L. (G. Don). These bacteria were also found in these locations in *H. coagulata* collected from peach orchards with PPD or fed on PLS-affected *Prunus cerasifera* L. In both sharpshooters, bacteria were found attached by one end to wall lining and they appeared to be undergoing cell division. Bacteria were located in the precibarium above and below the precibarial valve.

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IMPROVED PROCEDURES FOR GAS-CHROMATOGRAPHIC DETERMINATION OF NITROGEN-FIXATION RATES FOLLOWING RHIZOBIUM INVASION OF LEGUME ROOTS. A. W. Helton and Richard Dilbeck, Department of Plant and Soil Sciences, University of Idaho, Moscow, ID 83843.

The acetylene-ethylene assay served as the basic test method for determination of nitrogen-fixation rates at various times following inoculation of field-pea roots at planting time in a greenhouse with a commercial preparation ("Nitragin") of *Rhizobium leguminosarum*. Blood serum tubes (13-cc Vacutainers) were evacuated to 600 mm Hg, sealed with liquified Parawax to preserve the vacuum, and used to store 15-cc acetylene-ethylene-air samples from containers in which excised roots of Alaska pea plants had been incubated in a 10% acetylene-in-air atmosphere for 1 hr. The acetylene-ethylene samples required 7 days to achieve homogeneous distribution of gasses in the tubes, as many as 25 0.1-cc subsamples could be withdrawn from each tube without decline in gas-chromatograph-indicated concentration of ethylene, and the tubes could be stored in darkness at $21 \pm 1^\circ\text{C}$ for 95 days without decline in GC-indicated concentration of ethylene.

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AN INOCULATION TECHNIQUE FOR *PSEUDOMONAS SYRINGAE* ON SEEDLING BARLEY. H. E. Bockelman, D. C. Sands, and A. L. Scharen, Dept. of Plant Pathology and USDA-ARS, Montana State Univ., Bozeman 59717.

A pressure injection technique was used to inoculate barley

seedlings with a Montana isolate of *Pseudomonas syringae* pv. *syringae*. The primary leaf of seedlings in the two-leaf stage was inoculated using a "tongue-seizing" forceps fitted with a syringe. Inoculated plants were placed in a lighted dew chamber at 5°C for four days. Hypersensitive reaction (HR)-like symptoms (similar to the HR observed on the flag leaf in the field) were observed two days after plants were removed from the dew chamber. No chlorosis or HR was observed beyond the point of inoculation. The extent of HR-like symptoms were rated on 50 barley cultivars. 'Steptoe' gave HR at 10^6 cfu/ml and above, while 'Shabet' gave HR at 10^8 cfu/ml and only slight chlorosis at 10^6 cfu/ml. The clearest differential reactions were obtained with 10^6 cfu/ml. Two other inoculation methods (application of inoculum to clipped plants and spraying intact plants) were ineffective, even at 10^8 cfu/ml.

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THE USE OF ULTRASOUND TO FACILITATE THE HARVESTING AND QUANTIFICATION OF EPIPHYTIC AND PHYTOPATHOGENIC MICROORGANISMS. D. Haeefe and R. Webb. Dept. Plant Pathology, University of California, Berkeley, CA 94720.

Ultrasound energy has many applications which exploit its ability to safely remove unwanted particles from delicate surfaces which may be otherwise inaccessible. Ultrasound energy has, for example, been used to remove contaminating microorganisms from macroalgae in culture. Here we report the use of ultrasound energy for rapid, reliable collection of epiphytic bacteria from the above ground portion of terrestrial plants and conidia from agar culture of pycnidial fungi without the use of extensive washing procedures or surfactants. Epiphytic bacteria are collected by immersing the host plant parts in a sterile buffered solution contained in a small flask. This flask is partially submerged for five minutes in a 240 watt ultrasonic bath. Agar culture of pycnidial fungi are harvested utilizing a similar technique. Quantification then follows standard microbiological techniques. Replicated trials show accuracy equal to that obtained using traditional techniques, with considerable time saving. There is also less damage to host tissue.

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A NOVEL METHOD OF MICROBIAL IDENTIFICATION, Valerie N. Hall, D. C. Sands, H. K. Kim, D. P. Bancroft¹, and E. H. Abbott¹. Dept. of Plant Pathology and (1) Dept. of Chemistry, Montana State University, Bozeman, MT 59717.

Carbon 13 Nuclear Magnetic Resonance spectroscopy (nmr) allows non-destructive identification of small organic molecules within bacterial cells. High resolution nmr permits natural abundance spectroscopy, whereas a less sensitive system requires the use of carbon 13 enriched substrates. Our research with natural abundance spectroscopy showed that strains of *Xanthomonas campestris* pv. *translucens* gave similar spectra, but other xanthomonads and pseudomonads exhibited widely dissimilar spectra. Cells grown on a rich peptone medium vs. a glucose-mineral salts medium gave identical results, as did cells from different phases of growth. Nmr also has potential for determining pathway intermediates and the mode of action of pesticides.

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THE SYSTEMATIC ANALYSIS OF PLANT-ASSOCIATED FLUORESCENT PSEUDOMONADS. B. C. Hemming, C. Orser, D. L. Jacobs, G. A. Strobel and D. C. Sands. Dept. of Plant Pathology, Montana State University, Bozeman, MT 59717. The experiments reported represent the first systematic study of iron-induced changes on microbial antagonism of plant-associated fluorescent pseudomonads. A comprehensive numerical analysis which included data on 113 characters of nearly 200 isolates from a variety of plants served as the foundation on which the strain specific iron-induced changes in inhibition were examined. Isolates tended to cluster on the basis of their host plant origin when characterized by carbon utilization tests. The numerical analysis facilitated the evaluation of the relative merit of sole carbon source tests, bacteriocin tests and antibiotic production tests in the characterization of strains. In addition, hypersensitivity response production in *Nicotiana* sp., phytopathogenicity in *Tagetes* sp., and ice nucleation ability of several isolates were determined. The ability to grow in the presence of 10.0 mM EDTA was restricted to a limited number of isolates.

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RAPID FIELD IDENTIFICATION OF A BACTERIAL PATHOGEN BY AN ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA). A. Alvarez and K. Lou. Dept. of Plant Pathology, University of Hawaii, Honolulu, HI 96822.

An ELISA double antibody technique was developed to detect the presence of *Xanthomonas campestris* pv. *campestris* in leaf disks sampled from cabbage in the field. Assay time was reduced from

4-5 days (using semi-selective SX medium) to 5 hr using ELISA. Sheep and rabbit anti-*Xanthomonas* were used as primary and secondary antibodies. Goat anti-rabbit peroxidase and purified 5-aminosalicylic acid were used as the enzyme indicator system, measured at 450 nm. Based on 438 samples with disease prevalence of 54%, ELISA detected 96.2% of known positives as confirmed by isolation and pathogenicity tests. The level of detection was 2×10^6 cells/ml released from a 5 mm diam leaf disk after 2-24 hr, which is below the number of cells ($5 \times 10^8 - 1 \times 10^9$) commonly present in equivalent tissue exhibiting symptoms. ELISA permits processing of large sample numbers and facilitates determinations of infection rates for epidemiological studies.

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A NEW BACTERIAL BLIGHT OF CHICKPEA. F.L. Caruso and D.J. Dampen, Department of Botany & Plant Pathology, University of Maine, Orono, Maine 04469.

Five cultivars of chickpea (*Cicer arietinum* L.) exhibited stem lesions, leafspots and a generally blighted appearance in a field in July, 1981. Isolations from affected tissue consistently yielded a white pigmented, gram-negative, rod-shaped bacterium. Forty isolates reproduced stem lesions and leafspots in greenhouse inoculations. Reisolations yielded the same organism used as inocula. Biochemical and other tests have tentatively identified the causal bacterium as a non-fluorescent *Pseudomonas* species. All chickpea cultivars tested were susceptible, and the bacterium also induced stem lesions on garden pea and pinto bean. We believe this to be the first report of a blight of chickpea caused by a species of *Pseudomonas*.

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QUANTITATIVE ESTIMATION OF *Rhizoctonia solani* ANASTOMOSIS GROUPS IN SOIL. C. Castro, J. R. Davis and M. V. Wiese, Univ. of Idaho Dept. of Plant and Soil Sciences, Moscow, ID 83843.

A sensitive procedure involving the use of Prochloraz (a selective fungicide) permitted quantification of propagules of *Rhizoctonia solani* anastomosis groups (AG) in soil. Soil was treated with 5 ppm Prochloraz, formed into pellets and incubated on Ko and Hora's medium also modified with 5 ppm of Prochloraz. After 24 to 72 hours of incubation, *Rhizoctonia* isolates were selectively recovered from the pellets and examined for nuclei number. Multinucleate isolates were then identified by mating against known AG-types. With this method, the efficiency of recovery of *R. solani* AG-3 and AG-4 from soil artificially infested with mycelial fragments and sclerotia was 75 and 88%, respectively. In contrast, recovery without the use of Prochloraz was 17 and 33% for AG-3 and AG-4. Populations of *R. solani* in freshly collected soil from potato field plots ranged from 0 to 20 propagules/100 g and the predominantly occurring isolate was *R. solani* AG-3.

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ANTIBIOSIS AS THE MECHANISM OF ANTAGONISM OF STREPTOMYCES HYGROSCOPICUS VAR. GELDANUS TO RHIZOCTONIA SOLANI. C. S. Rothrock* and D. Gottlieb. Dept. of Plant Pathology, Univ. of Illinois, Urbana, IL 61801,*Dept. of Plant Pathology, Georgia Station, Univ. of Georgia, Experiment, GA 30212.

S. hygroscopicus var. *geldanus* controlled root rot of pea caused by *R. solani*, as well as reducing saprophytic growth and the population of the pathogen in sterilized soil when the antagonist was incubated for 7 days prior to infesting soil with the pathogen and planting. Evidence for antibiosis as the mechanism of antagonism is: (1) production of a zone of inhibition to *R. solani* on media, (2) inhibition of growth of *R. solani* by geldanamycin in vitro by as little as 0.5 ug/ml, (3) production of geldanamycin in soil (88 ug/g) in which the antagonist was incubated, (4) that the period of incubation necessary for disease control and production of the antibiotic are similar, and (5) geldanamycin when added to soil in concentrations similar to that produced in soil reduced disease and saprophytic growth. No evidence for antagonism due to other mechanisms was found.

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VIRULENCE OF RHIZOCTONIA SOLANI AG-2 TYPES 1 AND 2 AND AG-4 FROM PEANUT SEED ON CORN, SORGHUM, LUPINE, SNAPBEAN, PEANUT AND SOYBEAN. D. K. Bell and D. R. Sumner, Coastal Plain Exp. Stn. Tifton, GA 31793.

Three isolates of *Rhizoctonia solani* AG-2 type (T)-1, seven of AG-2 T-2 and 14 of AG-4 from surface disinfested, visibly sound peanut seed were tested for virulence on corn, sorghum, lupine, snapbean, peanut and soybean in heat treated (60 C, 30 min)

Tifton loamy sand in a greenhouse. Compared to the control, there was no difference ($P=0.05$) in emergence for crops with AG-2 T-1 isolates and for corn, sorghum, snapbean, peanut and soybean with AG-2 T-2; emergence of lupine was reduced by AG-2 T-2. Emergence of all crops except corn was reduced by AG-4. There was no increase in root or hypocotyl disease index (RHD1, min 1-max 5) among crops with AG-1 T-1 and for peanut with AG-2 T-2; the RHD1 for all other crops was increased by AG-2 T-2. The RHD1 was increased for all crops except corn by AG-4. There was no relationship between rate of growth of isolates on tannic acid-benomyl selective medium and virulence.

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CONTROL OF RHIZOCTONIA SOLANI AND PYTHIUM SPP. WITH METHAM SODIUM APPLIED THROUGH SPRINKLER IRRIGATION. Donald R. Sumner and S. C. Phatak, Coastal Plain Experiment Station, Tifton, GA. 31794.

Metham sodium (32.7% a.i.) was applied onto Bonifay sand through irrigation water in an overhead sprinkler system at 93, 468, 935, or 1870 liters/ha in 6.4, 12.7, or 25.4 mm of water; or by injection 20-25 cm deep into soil with chisels 25 cm apart followed by irrigation with 12.7 mm of water. Metham sodium at 468 l/ha or more in 12.7 or 25.4 mm of water reduced populations of *Rhizoctonia solani* and *Pythium* spp in soil 0-8 and 8-16 cm deep consistently and increased yields of vegetables, whereas application with chisels was inconsistent. Metham sodium at 93 l/ha, or at higher rates in only 6.4 mm of water, was ineffective. In a greenhouse test, metham sodium was applied to infested soil at 93, 234, or 468 l/ha in 12.7 mm water. Populations of *R. solani* AG-4 and AG-2, and *Rhizoctonia* spp. (binucleate) CAG-2, CAG-3, and CAG-5 were controlled 0-8 and 8-16 cm deep with 234 and 468 l/ha, but not with 93 l/ha. Hypocotyl and root rot in snapbean and corn were reduced with 234 l/ha and controlled with 468 l/ha; 93 l/ha was ineffective.

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EFFECTS OF TILLAGE PRACTICES ON POPULATION DENSITIES OF RHIZOCTONIA SPP. IN A RYE-SOYBEAN, MULTICROPPING SYSTEM. R. C. Ploetz, D. J. Mitchell, and R. N. Gallaher, Univ. of Florida, Gainesville, FL 32611.

Soilborne populations of *Rhizoctonia* spp. (RS) were monitored ca. every 5 wks in 1981 and 1982. Rye planted in Nov. was harvested in May; soybeans planted in June were harvested in Oct. Tillage treatments (tilled to 15 cm or not tilled), replicated four times, were imposed annually in Nov. beginning in 1976. Total RS populations fluctuated with seasonal environmental changes, changes of crop, and in particular the imposition of tillage treatments. In general, RS populations were higher in no-till plots than in tilled plots during the rye crop; no significant differences occurred in the soybean crop. *Rhizoctonia solani* AG-4 and several uncharacterized binucleate isolates of RS were isolated routinely; *R. zeae* and CAG-4 isolates were isolated rarely. Although tillage regimes appeared to influence individual species, mean population differences among tillage treatments were seldom significant due to large sample variances. Nonsignificance may have resulted, in part, from clumping of RS inoculum which occurred in preliminary work.

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GROWTH AND POPULATION-DISEASE RELATIONSHIPS OF RHIZOCTONIA SOLANI IN SOIL UNDER CONTROLLED TEMPERATURE AND MOISTURE. R. S. Kinsbursky and A. R. Weinhold, Dept. Plant Pathology, University of California, Berkeley, CA 94720.

To determine growth of *R. solani*, inoculum was placed on the intersection of two polyester threads held in place by a circular wire frame. The threads were placed in the center of a 3 cm deep soil column contained in a 5.5 cm diam. brass ring. After 38 hrs. the length of mycelial growth along the threads was measured. Population-disease relationships were determined by placing radish seeds in soils containing various populations of natural propagules of *R. solani*. Soil was placed in the rings and after 4 days seedling survival was determined. A pressure bomb was used to regulate soil moisture and after a 12-18 hr. equilibration period to bring the water potential to -0.1 bars, the rings were covered with a clear plastic film to retain moisture and transferred to an incubator or growth chamber held at 27°C. Growth in 72 soils was determined and ranged from 0.20 to 0.55 mm/hr. For six soils the effective inoculum density (EID) to give 50% seedling survival ranged from 18 to 48 prop./100 g soil. There was no relationship between rate of mycelial growth and EID₅₀.

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THE PERSISTENCE OF ENDOCONIDIAL AND MYCELIAL CHLAMYDOSPORES OF FUSARIUM CULMORUM W.G. SM. IN WHEAT-FIELD SOILS OF EASTERN WASHINGTON. D.A. Inglis and R.J. Cook, Dept. of Plant Pathology, Washington State University, Pullman, WA 99164-6430.

Fusarium culmorum, cause of foot rot of wheat, survives in soil

as chlamydospores formed in macroconidia (endoconidial) or hyphae (mycelial). The difference in survival between the two chlamydospore types was not significant in a 3-yr survival study under field conditions in Palouse and Ritzville Silt Loam soils at Pullman and Lind, WA (50 and 25 cm annual precipitation), respectively. Both types of chlamydospores were shorter-lived at Lind; yet fields infested with the pathogen and occurrence of severe foot rot are much more common in the Lind than in the Pullman area. Endoconidial chlamydospores are more prevalent in culture than mycelial chlamydospores and occur in wheat-field soils of both areas at a ratio of 9:1, respectively. The higher number of infested fields near Lind relates to more favorable conditions for disease, and hence to production of more inoculum which offsets the shorter life of chlamydospores. The fungus survives better in soil in Pullman, but unfavorable conditions for disease preclude the formation of chlamydospore inoculum.

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MECHANISMS OF SUPPRESSION OF CHLAMYDOSPORE GERMINATION FOR FUSARIUM OXYSPORUM F. SP. PISI IN SOILS. S.F. Hwang, R.J. Cook, and W.A. Haglund, Dept. Pl. Path., WSU, Pullman, WA 99164-6430.

Glucose at 0.8mg/g soil supported 80-90% chlamydospore germination of *F. oxysporum* f. sp. *pisi* race 1 in wilt conducive soil but only 10-15% in suppressive soil. In suppressive soil, doubling the glucose gave 30% germination. A four-fold or eight-fold increase of glucose gave 50-60% germination, indicating other limiting factors in the suppressive soil. Supplementing glucose (3.2mg/g soil) with asparagine (0.8mg/g soil) or FeCl₃ (up to 200uM) gave no more germination than occurred with the glucose alone. However, EDDHA (200uM) but not EDDHA-Fe reduced germination to 31% in conducive soil. One mechanism of suppressiveness apparently involves inadequate carbon and energy available for the pathogen, but other mechanisms must operate which cannot be nullified by adding glucose, asparagine, or iron. Suppressiveness was eliminated by moist heat-treatment (60C/30min). Adding 1% suppressive soil to conducive soil reduced germination slightly, but adding 20% suppressive soil to the mixture was equal to 100% suppressive soil.

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THE EFFECT OF FREQUENCY OF IRRIGATION ON SEVERITY OF FUSARIUM YELLOWS OF CELERY. J. C. Correll and R. W. Schneider, Dept. Plant Pathology, University of California, Berkeley, CA 94720.

Field observations consistently indicated that *Fusarium* yellows, caused by *Fusarium oxysporum* f. sp. *apii*, is more severe where irrigation water accumulates and oxygen stress conditions develop. A field experiment was conducted in uniformly infested soil in which celery was grown under heavy (twice per 8 days) or light (once per 8 days) irrigation schedules for the first 2 months of the season after transplants were established. Incidence of vascular discoloration under the heavy and light irrigations, respectively, were 90.1 and 81.7% in roots, 50.8 and 38.4% in the lower crowns, and 25.0 and 11.7% in the upper crowns (marketable portion). Measurements of oxygen diffusion rates indicated that plants receiving the heavy and light irrigations were exposed to cycles of oxygen deficits of 3-5 and 1-2 days, respectively. These data suggest that progression of the disease from roots to lower crown and particularly from lower crown to the marketable crown tissue is accelerated by periods of oxygen stress. Greenhouse experiments also indicated that root colonization by the pathogen is increased following brief periods of oxygen stress.

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INOCULUM-DENSITY-LEVEL STUDY OF HELMINTHOSPORIUM SATIVUM AND FUSARIUM GRAMINORUM AND THE INCIDENCE OF WHEAT ROOT ROT DISEASE IN OKLAHOMA. C. I. Umechuruba, Sch. of Biol. Sciences, Univ. of Port Harcourt, Nigeria and L. L. Singleton, Dept. of Plant Pathology, Oklahoma State Univ. Stillwater, Oklahoma 74078.

Inoculum-density-level of *H. sativum* and *F. graminorum* and the incidence of wheat root rot disease on hard red winter wheat cultivar, 'Danne' was evaluated in the greenhouse. Percentage recovery of *H. sativum* from subcrown internodes ranged from 48 - 97% as the inoculum-density-levels increased from 1-1000 conidia per gm. of soil and plateaued between 250-1000 conidia per gm. of soil. Percentage recovery of *F. graminorum* from subcrown internodes ranged from 41-57% as inoculum-density-levels increased from 1-100 macroconidia per gm. of soil, plateaued between 100 and 500 macroconidia per gm. of soil, and finally declined to 45% at 100 macroconidia per gm of soil. Disease severity rating of the subcrown internodes based on percentage lesions covering the surface areas of the internodes followed similar trend for each of the pathogens.

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ENHANCED BIOLOGICAL CONTROL OF WHEAT TAKE-ALL WHEN INHIBITORY PSEUDOMONAS STRAINS ARE INTRODUCED ON INOCULUM OR SEED AS OPPOSED TO DIRECTLY INTO SOIL. H.T. Wilkinson, D.M. Weller and

Four strains of *Pseudomonas* spp., two that inhibit *Gaeumannomyces graminis* var. *tritici* (Ggt) in-vitro and two that do not, were tested for ability to prevent infections when: (i) infiltrated into particles of Ggt-colonized oat grains; (ii) coated onto wheat seeds; and (iii) drenched into soil. The inhibitory strains were isolated from wheat roots grown in take-all suppressive soil and the non-inhibitory strains were from wheat roots grown in take-all conducive soil. Inhibitory strains gave best control of infections when infiltrated into inoculum particles. Coating the seed gave good control but less than when bacteria were infiltrated into inoculum. Drenching the soil gave no control. Non-antagonistic strains gave no control by any method of application. It appears that *Pseudomonas* strains, may not only inhibit development of lesions but may also have an important continuing inhibitory effect on the pathogen after the root dies and becomes a source of inoculum.

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ESTIMATED DISTANCES FOR INFECTION OF WHEAT ROOTS BY *G. GRAMINIS* VAR. *TRITICI* IN TAKE-ALL SUPPRESSIVE AND CONDUCTIVE SOILS. H.T. Wilkinson, J.R. Alldredge, and R.J. Cook, WSU, Pullman, WA 99164

A model was developed to estimate the distance for infection (EDI) between a wheat root and an inoculum particle based on the number of infections produced by particles infested with Ggt. Inoculum particles of axenically colonized oat grains were added at five concentrations (0.1-10.0 mg/g soil) into a soil naturally suppressive to take-all (SSL) and a naturally conducive soil (RSL). The mean EDI in SSL soil (5.9 mm) and RSL soil (5.3 mm) were not significantly different. In both soils, inoculum particles 1.0-2.0 mm had a greater mean EDI value (11.2 mm) than particles 0.5-1.0 mm (EDI=4.8 mm) and particles 0.25-0.5 mm had an EDI value of 2.4 mm that was significantly less than that of the larger particles. The EDI values in fumigated or pasteurized soils were similar and significantly greater than in natural soils (6.5, 6.1, 4.2 mm, respectively). Infiltration of the inoculum particles with inhibitory strains reduced the EDI by 1/2 compared to untreated inoculum particles or particles infiltrated with non-inhibitory strains.

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COLONIZATION OF WHEAT ROOTS BY A TAKE-ALL SUPPRESSIVE *PSEUDOMONAD*. David M. Weller, USDA, WSU, Pullman, WA 99164.

Seed treatment with fluorescent *Pseudomonas* strain 2-79 suppresses take-all of wheat caused by *Gaeumannomyces graminis* var. *tritici* (Ggt). An antibiotic-resistant strain of 2-79 was used to study colonization of wheat roots infected by Ggt. Three weeks after planting treated winter wheat (108 CFU/seed) in the field, more than 10^6 CFU of 2-79/0.1 g root were detected. They composed about 50% of the total bacteria and about 90% of all *pseudomonads* on the roots. The population of 2-79 declined to 10^3 CFU/0.1 g root during the winter but in the spring increased at least 10 fold. The bacteria were initially detected on the seminal and later on the coronal roots. Higher levels of 2-79 were detected on roots with take-all than on healthy roots. To study the movement of 2-79 with the growing root, consecutive 1.5 cm sections of individual seminal roots from their origins to their tips were sampled for 6 wk. The distal 1.5 cm section was always colonized by 2-79 and the population of 2-79 increased 10 to 100 fold in sections preceeding the tip.

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EXUDATION AND THE FUNGISTASIS OF *COCHLIOBOLUS VICTORIAE* CONIDIA IN SOIL. L. Epstein and J.L. Lockwood, Dept. of Botany and Plant Pathology, Michigan State University, E. Lansing, MI 48824

Fungistasis in soil could be due to the presence of germination inhibitors or to the loss of germination promoters from propagules. ^{14}C -labeled *Cochliobolus victoriae* conidia released a glucose-rich exudate during the first 30 min after wetting. To determine if this early exudate was involved in fungistasis, the conidia were incubated in a germination-suppressive environment (soil or two model sterile systems that simulated fungistasis in soil) or in a germination-conducive environment (sand). Conidia first incubated in the suppressive environment for 2-8 hr, then transferred to the conducive one, germinated more rapidly than conidia incubated only in the conducive environment. Temperature, pH, and osmotic potential had greater effects on the stasis imposed by the model systems than on stasis in soil. Exudation in the first 30 min occurred regardless of environmental conditions or subsequent germination. This and other evidence indicated that the primary exudate was probably not involved in soil fungistasis.

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EFFECT OF SOIL MICROBIOTA ON GERMINATION OF *COCHLIOBOLUS VICTORIAE* CONIDIA. L. Epstein and J. L. Lockwood, Dept. of Botany and Plant Pathology, Michigan State Univ., East Lansing, MI 48824.

A fungistatic environment was generated by the saturation of a sand substratum with a suspension of 0.3-14% Capac clay loam. Fungistasis was reduced by 1) the addition of 50 ppm of vancomycin, chloramphenicol or rifampin, or 2) by reduction of the number of soil particles, and hence microbes, in a soil suspension by dilution, centrifugation or ultrafiltration. Thirty-eight isolates obtained on several culture media from either fungistatic (soil extract) or non-fungistatic (non-soil) environments were inoculated individually into sterile soil extract which was used to saturate sand. Most of the bacterial isolates inhibited germination of *Cochliobolus victoriae* conidia, regardless of the medium used for the initial isolation. Prior colonization of the sand substratum was not required for the isolates to suppress conidial germination. There was no evidence that antibiotics or other inhibitory volatile or non-volatile compounds were involved in fungistasis. The results indicated fungistasis was caused nonspecifically by microbes.

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THE ROLE OF SOIL BORNE FUNGI IN THE DRY BEAN ROOT ROT COMPLEX. M.F. Mulligan, G.R. Safir and A. Smucker, Departments of Botany & Plant Pathology and Crop & Soil Sciences, Michigan State University, East Lansing, MI 48824

The severity of root rot of *Phaseolus vulgaris*, caused by *Fusarium solani* f.sp. *phaseoli*, was monitored in naturally infested Michigan fields throughout the 1981 growing season. Data were collected for several bean varieties at several levels of soil compaction in both sandy and clay soils. Varieties differed significantly in their root rot ratings and higher average root rot ratings were associated with lower yields. Disease incidence was not significantly different between varieties. Soil compaction significantly increased the severity of root rot and decreased mycorrhizal infection especially in sandy soils. Disease severity ratings taken at the time of flowering correlated best with reduced plant bio-yield than were ratings taken at other times. The severity of root rot was expressed as the percentage of the root area infected.

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INFECTION OF LATERAL ROOTS OF ALFALFA BY PLANT PATHOGENIC FUNGI. J. G. Hancock, Dept. Plant Pathology, University of California, Berkeley, 94720.

The number of infections and degrees of colonization of lateral roots of alfalfa by *Pythium* spp. were more extensive between October and April (20-25 infection sites/100 cm roots) than between June and August (9-15 infection sites/100 cm roots) in the Sacramento Valley of California. *Pythium ultimum* was the species most frequently isolated from roots during the summer whereas *P. violae* was the species most frequently isolated during the winter. The degree of infection by *Fusarium* spp. (mainly *F. acuminatum*, *F. culmorum*, and *F. oxysporum*) was relatively constant over all seasons but was slightly greater during the warm months: a similar pattern was observed with *Rhizoctonia solani* (anastomosis group 4). The numbers of infections were much greater in the upper 15 cm of soil (tillage layer) than between 30 and 80 cm depths. Root infection by *Pythium* spp. and *R. solani* was negligible at soil depths below the tillage layer. The living : dead root ratios were lowest in the tillage layer of soil and highest below this depth.

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ASSESSMENT OF COMMERCIAL FIELDS FOR BEAN ROOT ROT AND PEA ROOT ROT POTENTIALS IN WISCONSIN'S CENTRAL SANDS. K. M. Kobriger and D. J. Hagedorn, Dept. Plant Path., Univ. of Wis., Madison 53706.

Vegetable growers in the central sands area of Wisconsin are practicing increased double-cropping by planting peas, *Pisum sativum*, early in the season and snap beans, *Phaseolus vulgaris*, after pea harvest. This practice may lead to increased problems with root rots considering the recent discovery of a bean strain of *Aphanomyces euteiches* and the implication of *Pythium* spp. as causal agents of the root rot complex. We sought to determine the relationship between the bean and pea root rots in the central sands. Soil samples were collected from production fields and bioassayed in the greenhouse to determine the severity indices of bean and pea root rots. Optimal environmental conditions for disease development were provided. A total of 91 fields were tested from 1979-1981. A bean root rot index greater than 65 was recorded in 49 field samples while only 21 of these samples had comparable pea root rot indices. Thus bean and pea root rot potentials may be assessed with the same soil sample.

STUDIES ON FUNGAL COLONIZATION OF THE COTTON RHIZOSPHERE. J. S. Gerik and O. C. Huisman, Dept. Plant Pathology, University of California, CA 94720.

Colonization of cotton root tips by epiphytic root fungi was examined for field grown plants. The fungi studied included members of the genera *Verticillium*, *Fusarium*, *Trichoderma*, *Penicillium*, *Gliocladium*, *Cladosporium*, *Doratomyces*, *Aspergillus*, and *Stemphylium*. Samples were collected throughout the growing season and frequencies of the various fungal colonies were determined for 1 mm intervals from the root tips. The frequency of colonization of most fungi increased with distance from the root tip. Maximum frequency, usually occurring within the first 5 mm from the tip, varied with fungal species and time. During the early part of the growing season, the maximum frequency was closer to the root tip than later in the year, indicating a possible effect of root growth on the point of initial colonization. Positive and negative correlations between the frequency of root colonies of one fungus relative to others indicate the possibility of interaction among genera.

EFFECT OF pH ON NUTRITION AND RESPIRATION OF *PISOLITHUS TINCTORIUS*. W. A. Taber and R. A. Taber. Departments of Biology and Plant Sciences, Texas A&M University, College Station, TX 77843.

In vitro growth of the mycorrhizal fungus, *Pisolithus tinctorius*, cultured on synthetic media was favored by low pH (4.5-6.2). Little or no growth occurred at pH 6.9 on either glucose or sucrose. Both exogenous and endogenous respirations were also pH sensitive. Studies on respiration of both ^{14}C -glucose-labeled sucrose and ^{14}C -fructose-labeled sucrose revealed poorer respiration of the fructose moiety, suggesting some fructose is passed to an organic acceptor. Free fructose was readily respired and respiration was not reduced by the presence of unlabeled glucose. Ionized succinic acid (pH 7) was not respired whereas mainly unionized succinic acid (pH 4.5) was as readily respired as glucose (equivalent carbon basis). The bulk of respiration appears to be at the expense of some endogenous substrate.

EFFECT OF POST-COLONIZATION TREATMENTS ON SPORULATION OF VESICULAR-ARBUSCULAR MYCORRHIZAL FUNGI. D.M. Sylvia and N.C. Schenck, Plant Pathology Dept. Univ. of Florida, Gainesville, 32611.

Availability of spores is a limiting factor in mycorrhizal research. Our objective was to find treatments to stimulate sporulation in pot culture. *Glomus mosseae*, *G. clarum* and *Gigaspora margarita* were grown on *Paspalum notatum* in the greenhouse. During the logarithmic phase of root colonization five pots/species were exposed to one of six treatments: shoots removed (pruned), water withheld 9 days (drought), exposed to 5 C for 4 days in the dark (cold/dark), sprayed with paraquat at 69g a.i./ha, soil drenched with P_2O_5 at 500 ppm/pot, or soil drenched with ethazole at 12.5 ppm/pot. On an average, spores/g soil at 18 wk for *G. mosseae* and *G. margarita* (soil pH 6.5) increased 99, 40, or 24 percent in response to P_2O_5 , drought, or paraquat; and decreased 4, 9, or 48 percent with pruning, cold/dark, or ethazole treatments, respectively, when compared to the no treatment control. Spore numbers for *G. clarum* (soil pH 4.5) were not affected by P_2O_5 or drought but were suppressed by pruning, drought, cold/dark, and paraquat treatments.

ULTRAVIOLET-INDUCED AUTOFLUORESCENCE: A NEW METHOD FOR DETECTING AND QUANTIFYING VESICULAR-ARBUSCULAR MYCORRHIZAL ROOT INFECTIONS. R. N. Ames, E. R. Ingham, and C. P. P. Reid. Natural Resource Ecology Laboratory, Colorado State University, Fort Collins, Colorado 80523.

Arbuscules but not hyphae or spores of vesicular-arbuscular mycorrhizal (VAM) fungi autofluoresce when excited with ultraviolet light. Washed roots were viewed with an epifluorescent microscope at 455-490 nm excitation and 520-560 nm emission wavelengths. There was no significant difference in determining VAM infection by the UV or staining methods. Pathogenic fungal root infections did not autofluoresce. The UV method may not be useful for haustoria-forming foliar pathogens. Further examination of the autofluorescent compound(s) may provide information on the mechanisms of carbon and/or phosphorus exchange between the host and fungus. Significance and applications of the UV method will be discussed.

EFFECT OF SPORE VERSUS SOIL INOCULUM OF THE MYCORRHIZAL FUNGUS *GLOMUS MOSSEAE* ON GROWTH OF SOYBEAN. N. C. Schenck, G.S. Smith, and L. Zambolim. Plant Pathology Dept., Univ. of Florida, Gainesville 32611 and Univ. of Viçosa, Viçosa, M.G., Brazil. Our purpose was to compare the rate of root colonization and plant growth to initial differences in inoculum of *G. mosseae*. Inoculum consisted of spores alone or spores plus associated soil and roots from a bahiagrass pot culture (soil inoculum). Spore concentrations ranged from 20 to 2000 per 15-cm pot of steamed soil (2kg). Inoculum was placed 5-10 cm below the surface or was distributed uniformly throughout the soil just prior to planting. The experiment had 5 replicates of each treatment and was repeated 3 times. In some experiments, soil inoculum resulted in less growth stimulation than spore inoculum because *Fusarium solani* was introduced with the soil inoculum. In all experiments, regardless of inoculum source, soybean nodulation was consistently increased by *G. mosseae* and weight of mycorrhizal soybean was greater than nonmycorrhizal soybeans at 10-11 wks but were less than nonmycorrhizal soybeans at 2-4 wks. Both the rate of and final root colonization by *G. mosseae* increased as inoculum levels increased.

MYCOTROPHIC GROWTH OF HOST PLANT AND ENDOPHYTE DEVELOPMENT IN MYCORRHIZAL SOYBEANS. G.J. Bethlenfalvay and G. Fuller, Western Regional Research Center, USDA/ARS, Berkeley, CA 94710

The relationships between growth parameters of host plant and fungal endophyte were studied. Soybean [*Glycine max* (L.) Merr.] plants colonized by the vesicular-arbuscular mycorrhizal (VAM) fungus *Glomus fasciculatus* Gerdemann and Trappe were grown in pot cultures utilizing a composite greenhouse rooting medium. Development of fungal mycelia inside and outside the host root and total fungal biomass were determined from assays of fungal chitin. Growth and phosphorus uptake by VAM plants and uncolonized controls were compared. Mycotrophic growth in VAM plants occurred during the final six weeks of the 19-week growth period, when the concentration of available soil P fell below 10 $\mu\text{g P/g soil}$. Growth enhancement was most pronounced in the reproductive organs. The data suggest a relationship between the initiation of the reproductive phase in the host and the cessation of growth in the endophyte. Source-sink relationships and P availability appear to be factors influencing interactions between the symbionts.

CONDITIONS AFFECTING PARASITIC OR MUTUALISTIC GROWTH IN MYCORRHIZAL PLANTS. G. J. Bethlenfalvay, Western Regional Research Center, USDA/ARS, Berkeley, CA 94710

Soybean [*Glycine max* (L.) Merr.] plant colonized by the vesicular-arbuscular mycorrhizal fungus *Glomus fasciculatus* Gerd. and Trappe were grown in pot cultures using hydroxyapatite [$\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$] as a slightly soluble source of P. Growth response of host plants to VAM fungal infection varied from growth depression (parasitic growth of the endophyte) to growth enhancement (mutualism). Host-plant and endophyte development were correlated with P availability. At very low levels of NaHCO_3 -extractable P (1 $\mu\text{g P/g soil}$) host plant and endophyte development were inhibited. At an intermediate level of P availability (4 to 10 $\mu\text{g P/g soil}$) host-plant growth was enhanced, while at higher levels (10-20 $\mu\text{g P/g soil}$) it was inhibited. Total fungal biomass varied with P availability and plant age, and was maximally 20% of root dry weight. It is concluded that host-plant growth response to VAM fungal infection depended on the availability of and competition for P and photosynthate.

SELECTION OF VA MYCORRHIZAL FUNGI FOR USE ON ALFALFA GROWN IN ARIZONA. M.D. Steinberg and H.E. Bloss. Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

Alfalfa varieties were inoculated with five species of VA mycorrhizal fungi and screened for optimum biomass enhancement in the greenhouse. Based on results from the study conducted in the greenhouse three species of *Glomus* (*G. deserticola*, *G. mosseae*, and an undescribed *Glomus*) were selected for use with one variety of alfalfa (Hayden Px-1) to obtain biomass increases in Arizona field conditions. Alfalfa plants were either preinoculated with the mycorrhizal fungi, grown in the greenhouse and transplanted to the field, or were direct seeded into rows in which a layer of mycorrhizal inoculum had been incorporated. Phosphorus was added to the soil (100 lbs treble super-phosphate/acre) whereas phosphorus was not added to controls. A 65% increase biomass of plants preinoculated with the undescribed *Glomus* in the absence of phosphorus occurred over controls. A 90% increase occurred in plants preinoculated

with the undescribed *Glomus* with added phosphorus. Significant biomass increases did not occur with the other treatments.

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INCIDENCE AND SPECIES OF VESICULAR-ARBUSCULAR MYCORRHIZAL (VAM) FUNGI ASSOCIATED WITH WINTER WHEAT AS COMPARED WITH UNDISTURBED TALL GRASS PRAIRIE GRASSES. B. A. Daniels and J. Bloom, Department of Plant Pathology, Kansas State University, Manhattan, Kansas 66506.

The Konza prairie and several geographically distinct wheat fields were sampled monthly throughout a growing season. Two previously undescribed sporocarpic VAM species were observed in prairie samples. Moderate infection occurred in regularly burned or unburned prairie plots throughout the year. Infection levels fluctuated monthly, but no seasonal variation was observed. More VAM species sporulated in spring and fall than during other seasons. In contrast, little or no root infection was evident in wheat before flowering in May and June. More VAM species sporulated in spring but spore numbers were consistently lower in these fields than in the native prairie. VAM fungi probably function in nutrient cycling in undisturbed perennial tall grass prairie but the low spore numbers and late growing season infection in wheat make the benefit of VAM infection in wheat less likely.

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EFFECT OF CITRUS ROOT EXUDATES ON GERMINATION OF CHLAMYDOSPORES OF THE VESICULAR-ARBUSCULAR MYCORRHIZAL FUNGUS, *GLOMUS EPIGAEUM*. J. H. Graham, University of Florida, IFAS, Agril-cultural Research and Education Center, Lake Alfred, FL 33850.

The effect of root exudates from Troyer citrange (*Poncirus trifoliata* X *Citrus sinensis*) on germination of surface-sterilized chlamydospores of the vesicular-arbuscular mycorrhizal (VAM) fungus, *Glomus epigaeum*, was examined on water agar. Optimum temperature for spore germination was 25°C. Exudates, collected from roots in water and filter-sterilized, contained 1.5 mg/ml and 12.2 mg/ml total amino acids and sugars, respectively. Exposure of spores to root exudates increased germination 2 to 4 times compared to water alone. Growth of germ tubes from exudate-treated spores was more than 400% greater than growth from untreated spores. Root exudates stimulated branching of germ tubes in all directions, whereas germ tubes of untreated spores branched infrequently. These results suggest that root exudates could affect the activity of *G. epigaeum* in the rhizosphere and thereby alter VAM formation.

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MYCORRHIZAL FUNGI ASSOCIATED WITH NATIVE AND IMPROVED VARIETIES OF PECANS IN TEXAS. Ruth A. Taber, Dept. Plant Sciences, Texas A&M Univ., College Station, TX 77843; J.W. Worthington, Texas A&M Research Center, Stephenville, TX 76401; J.M. Trappe, USDA Forest Sciences Laboratory, Corvallis, OR 97331; and W.A. Taber, Dept. Biology, Texas A&M Univ., College Station, TX 77843.

Six genera of ectomycorrhizal fungi were associated with *Carya illinoensis* in managed orchards, urban plantings, and native habitats in 8 counties in Texas. First documentation of the natural occurrence of *Pisolithus tinctorius* on pecan was recorded from managed orchards in 3 counties. Sporocarps were produced in orchards treated with pesticides - Karmex, Princep, Surflan, Benlate, Du-Ter, Zolone, Sevin, and Roundup. They were abundant under canopy drip-lines, in soil depressions, and on roots severed by cultivators. *Astraeus hygrometricus* was found in orchards in 2 counties. *Scleroderma*, including *S. cepa*, fruited sometimes on roots which also had *Pisolithus* associations. *Tuber* was found in urban plantings. *Gyrodont*, *Tylopilus*, and *Russula* fruited only on native trees. VAM fungi included species of *Glomus*, *Gigaspora*, and *Sclerocystis*.

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INTERACTION OF FERTILIZER AND *Glomus fasciculatus* ON GREENHOUSE GROWN POINSETTIA (*Euphorbia pulcherrima*). J.W. Kaye, F.L. Pfeleger and E.L. Stewart, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Rooted poinsettia plants were either inoculated with *Glomus fasciculatus* (GF) or not inoculated (controls) and planted in a potting medium of 1 part peat:1 vermiculite:1 soil containing 5 or 12 ppm phosphorus (P). Both GF-inoculated and control plants were fertilized with 125 or 250 ppm (once/wk) or with 300 ppm (twice/wk) NH_4NO_3 and KNO_3 . Inoculation with GF had the following effects on the plants: (1) greater dry weights and total numbers of cuttings taken from plants grown at 5 ppm P fertilized with 250 or 300 ppm N and K, (2) foliar analysis

revealed a greater uptake of P and Cu at 12 ppm P fertilized with 125 ppm N and K, and at 5 ppm P fertilized with 125, 250 or 300 ppm N and K, (3) increased uptake of K at 5 ppm P fertilized with 125 or 250 ppm N and K. In general, % root infection by GF increased with decreasing levels of fertilizer. More chlamydospores were recovered from soil of plants grown at 5 ppm P fertilized with 250 ppm N and K.

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VA MYCORRHIZAL INFECTION IN BRASSICA, A NON-HOST
Marian G. Glenn, Department of Plant Pathology, University of Wisconsin-Madison, 53706.

This work examines the hypothesis that a general lack of functional VA mycorrhizae in the cruciferae and related families is due to presence of glucosinolates (gs) and their hydrolysis products, isothiocyanates, in and around roots. Hyphal growth of *Glomus mosseae* near roots of *Brassica napus* and *B. campestris* cultivars with low levels of gs was observed with light and electron microscopy before and after penetration, compared to growth elicited by cultivars with high levels of gs, and also to growth elicited by compatible hosts *Nicotiana tabacum* and *Allium cepa*. All brassica roots elicited fewer and shorter pre-penetration branches than did hosts, resulting in fewer attempts to penetrate. Hyphae penetrating brassica fail to produce coils or arbuscles, but grow within the cortex parallel to the root axis, and often form vesicles. The vesicles may serve a protective function, since they resemble vesicles produced externally under adverse conditions.

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SPORE POPULATIONS AND SPECIES COMPOSITION OF VESICULAR-ARBUSCULAR MYCORRHIZAL FUNGI IN RELATION TO SOIL FUMIGATION AND CROP SPECIES. A.-C. McGraw and James W. Hendrix, Department of Plant Pathology, University of Kentucky, Lexington, KY 40546.

Crop rotations involving four treatments of pepper (cv Yolo Wonder) or tomato (cv Jet Star) grown in fumigated (methyl bromide + chloropicrin [67/33%]) and nonfumigated soil, sudex, or fallow were established in June 1981. Pepper and tomato transplants were planted in rows of fumigated and nonfumigated soil. Sudex was planted in plots of nonfumigated soil. Pepper, but not tomato, yields were improved slightly in fumigated compared with nonfumigated soil. More than ten species of mycorrhizal fungi representing four genera (*Acaulospora*, *Gigaspora*, *Glomus*, *Sclerocystis*) could be identified from each treatment in June and October. *Glomus macrocarpus* comprised approximately 50% of the total population of mycorrhizal fungal spores. Crop treatment, particularly sudex, was more important than soil fumigation in manipulating the population level but not the species composition of the mycorrhizal fungi. Certain minority species were detected only with select soil and crop treatments.

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SUITABILITY OF WEEDS AS HOSTS FOR PRATYLENCHUS SPP. Ralph H. von Quallen and Glenn B. Bergeson, Department of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907.

Jimsonweed, johnsongrass, giant foxtail, giant ragweed, and lambsquarter were compared to corn as hosts for lesion nematodes (primarily *Pratylenchus hexincisus*). Each weed species was grown for six weeks with corn in soil infested with lesion nematodes from corn roots. The weeds were evaluated as hosts using two ratios: the ratio of weed to corn for the number of nematodes/gram of dry root, and the ratio of weed to corn for the number of nematodes/root system. The ratios were respectively: lambsquarter 0.47, 0.12; jimsonweed 0.28, 0.07; giant ragweed 0.36, 1.53; johnsongrass 1.19, 0.83; and giant foxtail 0.23, 0.04. Johnsongrass was a significantly ($P=0.01$) better host than the other weed species based on nematodes/gram of root; however, both johnsongrass and giant ragweed were significantly ($P=0.01$) better hosts based on nematodes/root system. None of the weeds were significantly better hosts than corn.

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COWPEA RESISTANCE TO MELOIDOGYNE INCOGNITA AND M. JAVANICA. T. A. Swanson and S. D. Van Gundy, Dept. of Nematology, U.C. Riverside, Riverside, CA 92521.

Cowpea cv.'s were compared for resistance by inoculating plants with an egg suspension ($P_i=5000$) and counting eggs extracted by the blender/bleach technique 6 wks later. Plants were grown at 27°C in a growth chamber. Highest reproduction occurred on Groit. Magnolia, Mississippi silver and Mississippi purple were resistant to both nematode spp. ($P_f=10\%$ of Groit while CBE 5 was

resistant to *M. i.* only. Other cv.'s were slightly resistant ($P_f=50\%$ of Groit). *M. i.* Race 2 reproduction was significantly greater than Races 3 and 4 on CBE 5. Race 1 was intermediate. Magnolia, *M. silver* and *M. purple* were resistant to all races while CBE 3 was resistant to Races 2 and 4. Queen Ann was resistant to Race 2. The optimum temperature for *M. i.* reproduction on CBE 3 was 26°C. Raising the temperature from 25 to 35°C did not break resistance and reduced reproduction on resistant and susceptible cv.'s. *M. i.* reproduction 3 weeks after inoculation was similar on CBE 3, CBE 5, and Magnolia but by 6 wks was significantly greater on CBE 3.

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MORTALITY IN SWARMING APHELENCHUS AVENAE. J. P. Hollis and E. P. White, Dept. Plant Path. & Crop Physiol., La. State Univ. Agric. Expt. Sta., Baton Rouge, La. 70803

Swarming populations from soybean roots of *Aphelenchus avenae* Bastian 1865 when dispersed in water showed 25% survival at 5 weeks. Swarmer mass cultured principally on *Fusarium oxysporum* Schlecht. in Petri plates containing 3% acid water agar showed 2% survival after extraction at 4 weeks, and all were non-swarmer; at 7 weeks swarms again appeared comprising 25% of populations, with 100% survival of swarmer and non-swarmer. Some specimens in undisturbed swarms survived (exhibited life movements) in water up to 6.5 months after extraction from soybean root surfaces. Dead specimens in all populations were darkened, greyed or browned with disorganized internal contents and characteristic open - C posture. *A. avenae* larvae hatched from surface sterilized swarmer eggs were nonswarming, but their progeny maintained in fungus culture transformed in 2 weeks again into swarmer, as previously observed in *Tylenchorhynchus martini* Fielding 1956 (Phytopath 71:226, 1981).

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NEMATODE DAMAGE IN CROPS LIMITED BY SYMBIONT SUPPLIED NUTRIENTS. J. P. Hollis. Dept. of Plant Path. & Crop Physiol., La State Univ. Agric. Expt. Sta., Baton Rouge, LA 70803

Comparisons of crop responses to factors reversed by soil nematocides show nematode damage: (a) corn in nitrogen adequate, nitrogen and minerals deficient soils, and (b) soybeans only in *Rhizobium* nitrogen and minerals deficient soils. Graphically: soil nitrogen \rightarrow nematodes \rightarrow plant; nematodes \rightarrow *Rhizobium* nitrogen \rightarrow plant. Soil nitrogen uptake in nonlegumes is reduced by damage to roots by intervening nematodes. Where legumes are grown in *Rhizobium* nitrogen adequate soils the nitrogen fixation process is interposed between nematodes and plants and supplies nitrogen to them independent of nematode damage to roots. Thus crop ecosystems involving donor symbionts supply nutrients directly to plants and bypass nematode effects on their uptake. Cooperation between nematologists and symbiont systems scientists involving transfers of symbionts and symbiont system genes to nonlegumes will reduce the agricultural significance of plant parasitic nematodes.

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EFFECT OF SOIL SOLARIZATION AND TELONE II ON PLANT-PARASITIC NEMATODES IN CALIFORNIA SOILS. J.J. Stapleton and J.E. DeVay. Dept. of Plant Pathology, Univ. of California, Davis, CA 95616.

Ten field sites in Yolo, Merced, Napa, and Sonoma Co. of CA, were pre-plant or post-plant treated for 4-6 wk with soil solarization and/or 122 l/ha Telone II (=50% label dose of 92% 1,3-dichloropropene). Subsequent soil assays included 42-100% reductions of *Meloidogyne*, *Heterodera*, *Pratylenchus*, *Paratrichodorus*, *Xiphinema*, *Crictonemella*, and *Paratylenchus* spp., and "total" nematodes ($P=0.05$ or 0.01). Population decreases were greater when solarization and fumigant were combined in 1/2 of the cases, as compared to either treatment alone. No significant reductions of population densities were detected below 46 cm soil depth. Greenhouse tests showed increased plant growth response (IGR) of 29-133% ($P=0.05$) in some solarized soils. No IGR was found in soils treated with fumigant alone; in treated soil from Napa or Sonoma Co.; or in soil from post-plant stone-fruit orchard treatments. No visible damage to orchard trees resulted from any of the treatments, and treated peach tree roots remained mycorrhizal after solarization.

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NEMATODE, BACTERIAL, AND NEMATOCIDE EFFECTS ON SOYBEAN NODULES. W. Birchfield and M. M. Joshi, USDA, ARS, Dept. Plant Path. & Crop Physiol., La. State Univ. Agric. Expt. Sta., Baton Rouge, LA 70803, and E. I. duPont de Nemours & Co., Wilmington, DE 19898.

Heterodera glycines, *Meloidogyne incognita*, *Rotylenchulus reni-*

formis, *Rhizobium japonicum*, Dasanit, Mocap, and Nematicur were used alone and in various combinations to determine their effects on soybean nodulation. Bacteria used were strain 6 and 10, antibiotic resistant, *Rhizobium japonicum*. Nematicide rates were comparable to 3.0 a.i./A of 15% granules mixed with the soil. Lee 74 soybeans were grown on a greenhouse bench under inflorescent and window lighting at 85°F. Nodule and nematode counts were made after a 78 d average. The highest nodule counts on soybeans were obtained with the *Rhizobium* inoculum alone and in combination with nematicides. Reniform nematode increased nodulation. Root knot had no effect and cyst decreased soybean nodulation.

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OCCURRENCE OF SOYBEAN CYST NEMATODE, HETERODERA GLYCINES AND OTHER NEMATODES PARASITIC ON SOYBEANS IN LOUISIANA. Bachireddy, V.R. and R. Payne, Jr. USDA-CRSR, Department of Plant and Soil Science, Southern Univ., Baton Rouge, LA. 70813.

During 1977-1981 a comprehensive survey of plant parasitic nematodes was carried out in all soybean growing parishes of Louisiana. Soil samples were collected from farmers fields in 54 parishes that have had soybeans growing on them for two or more years and analyzed for soybean parasitic nematodes. Results from the surveys showed that twelve parasitic nematode species associated closely with soybean roots. Soybean cyst nematodes (SCN) infestations are found concentrated mainly in the parishes bordering the Mississippi River and a gradual spread towards the South Central and Southwest parishes of the state. Distribution of SCN in 1981 encompassed 26 of 64 parishes (40.63%) and the survey showed that the area of infestation is expanding rapidly. Other genera recovered were *Meloidogyne*, *Rotylenchulus*, *Tylenchorhynchus*, *Helicotylenchus*, *Pratylenchus*, *Xiphinema*, *Trichodorus*, *Aphelenchoides*, *Crictonemoides*, *Tylenchulus*, and *Hoplolaimus* spp.

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FIGWORT MOSAIC VIRUS: A NEW CAULIMOVIUS. M. K. Handley, J. E. Duffus, and R. J. Shepherd, Department of Plant Pathology, University of California, Davis, 95616

Figwort mosaic virus (FMV), isolated from naturally infected *Scrophularia californica* Cham. (figwort), is transmitted by aphids and has inclusion bodies characteristic of the caulimovirus group. We have established that the virus has a circular double stranded DNA genome of about 8000 base pairs. Clones (in *E. coli* plasmid pBR322) of two strains of the virus and of a naturally occurring, non-infectious deletion have been compared by restriction endonuclease mapping. A physical map of the native virus has been prepared by determining the locations of the single stranded discontinuities with respect to the locations of several restriction sites. Four discontinuities occur in the viral DNA; three of these occur in one strand, and one occurs in the other strand. These clones of FMV show little or no sequence homology with cauliflower mosaic virus in Southern blot hybridizations.

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PROPERTIES OF THE LENTIL STRAIN OF PEA SEEDBORNE MOSAIC VIRUS. J. Goodell and R. O. Hampton, Dept of Bot & Plant Pathology, Oregon State University, Corvallis, OR 97331

The lentil strain, PSBMV-L, was distinguishable from the standard strain of PSBMV by an inoculum reservoir in lentil seed vs. pea seed, by separate resistance genes in *Pisum* and *Lens*, and by the instability of PSBMV-L in standard purification procedures. PSBMV-L was more prone to particle breakage, aggregation, and binding to host components, and unlike PSBMV was sensitive to low concentrations of organic solvents and tended to aggregate irreversibly at low Mg++ concentrations and when precipitated by PEG. PSBMV-L was purified by extraction in neutral buffer, clarification with 7.5% chloroform, centrifugation through PEG(6%)-sucrose(35%) cushions and equilibrium centrifugation in CsCl gradients to yield 0.01mg virus per g tissue. A260/280 was 1.15. The two strains were indistinguishable by SDS immunodiffusion or SDS immunoelectrophoresis but were clearly separated by quantitative ELISA serology. The protein subunit size of both strains was estimated at 33,000 d. Preliminary results of cDNA hybridization studies are presented.

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NUCLEIC ACIDS OF TOMATO BUSHY STUNT VIRUS. B. Hillman and T. J. Morris, Plant Pathology Department, University of California, Berkeley, CA 94720.

Tomato bushy stunt virus (TBSV) is a 30nm spherical virus with a single stranded RNA genome of about 1.6×10^6 . We have analyzed the virus

with respect to virion encapsidated ssRNA as well as replicative form (RF) dsRNA isolated from infected tissue. Results of virion ssRNA analysis indicate that in addition to genomic 1.6×10^6 RNA there is a subgenomic species of about 1.0×10^6 . Also, a small RNA species of about 0.15×10^6 is present in at least one isolate. This small RNA may be satellite-like in that modulation of symptoms on *Nicotiana clelandii* is associated with its presence or absence. Results of RF-RNA analysis indicate there are 4 major species in each of 4 strains tested. These have molecular weights of approximately 3.2, 2.0, 0.6 and 0.3×10^6 . The functional significance of the ssRNAs and their dsRNA counterparts will be discussed with respect to similar 30nm spherical viruses.

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A COVALENTLY LINKED PROTEIN AT THE 5'-ENDS OF THE GENOMIC RNAs OF PEA ENATION MOSAIC VIRUS. G. A. de Zoeten and D. Reisman, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.

A covalently linked protein that is attached to the 5'-ends of genomic RNAs of pea enation mosaic virus (PEMV) was found. Nuclease digestion of PEMV-RNA releases protein with an apparent molecular weight of approximately 17,500. The absence of a 5'-cap and the inability to introduce 32 P-phosphate into the 5'-termini of these RNAs suggests that the protein is covalently bound to PEMV-RNA.

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TRANSLATION OF THE RNA OF BEAN POD MOTTLE VIRUS USING RABBIT RETICULOCYTE LYSATES. C. J. Gabriel and K. S. Derrick, Dept. of Plant Pathology and Crop Physiology, and D. S. Shih, Dept. of Biochemistry, Louisiana State University Agricultural Experiment Station, Baton Rouge, LA 70803.

The translation of the two RNA components (B-RNA and M-RNA) of bean pod mottle virus (BPMV) in rabbit reticulocyte lysates resulted in the formation of eight virus proteins. Maximum incorporation occurred with magnesium acetate, potassium acetate, and viral RNA concentrations of approximately 0.3 mM, 30 mM, and 20-30 μ g/ml respectively. B-RNA coded for a 181K protein which was cleaved into 158K and 30K products. M-RNA coded for two polypeptides (107K and 97K) which were processed only in the presence of B-RNA translation products into 58K, 49K and 40K products. The BPMV protease was specific for BPMV polypeptides in that it would not process cowpea mosaic virus (CPMV) polypeptides. Conversely, CPMV protease would not cleave BPMV polypeptides.

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CITRUS TRISTEZA VIRUS RNA TRANSLATED WITH A RABBIT RETICULOCYTE LYSATE: CAPSID PROTEIN IDENTIFIED AS ONE OF THE PRODUCTS. J. Nagel, E. Hiebert, R.F. Lee, Dept. of Plant Pathology, Univ. of Florida, Gainesville, FL 32611, and Univ. of Florida, IFAS, AREC, Lake Alfred, FL 33850.

Citrus tristeza virus (CTV) is a closterovirus and contains a single-stranded RNA of $6-7 \times 10^9$ m.wt. Citrus tristeza virions, prepared by a procedure to enhance recovery of intact particles, were dissociated with an equal volume of 200mM ammonium carbonate (pH 9.0), 2% SDS, 2mM EDTA, and proteinase K. RNA was fractionated from sucrose linear-log gradients and ethanol precipitated. Translations were with rabbit reticulocyte lysates under conditions used with potyviruses. Capsid protein, ~26,000 m.wt. (26K), was identified as a translation product by immunoprecipitation with antiserum specific for CTV capsid protein. A major product of 50K and other products of 65K and 33K were also formed, all of which were non-reactive with antiserum to capsid protein. It is concluded that CTV RNA has messenger activity and is therefore plus-stranded.

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PURIFICATION OF INCLUSION BODIES OF CITRUS TRISTEZA VIRUS. R. F. Lee, S. M. Garnsey, R. H. Brlansky, and L. A. Calvert. AREC, University of Florida, Lake Alfred, FL 33850, and *USDA Horticultural Research Laboratory, Orlando, FL 32803.

Citrus tristeza virus (CTV)-infected tissue contains inclusions of aggregated virus. These inclusions were extracted by dicing young bark tissue in 0.1 M sodium citrate buffer, pH 6.0, containing 1% Driselase[®] enzyme. After incubation, Tris buffer, pH 8.0, with 2% Triton X-100[®] was added and the mixture was homogenized, filtered through cheesecloth, and centrifuged at low speed. The resuspended pellet was centrifuged for 3.5 hr at 36,000 rpm (SW41 rotor) on a stepwise gradient of 0.5, 1.0, and 2.0 molal Cs_2SO_4 made in 30% (w/v) sucrose. Zones containing inclusion bodies were identified by immunofluorescent

and SDS-immunodiffusion assays. Several apparently viral specific proteins were detected by SDS-polyacrylamide gel electrophoresis of solubilized inclusion bodies from step gradients. The free virus particles extracted by this procedure were also recovered from the low speed supernatant and purified.

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PROPERTIES OF VIRUS SPECIFIC MONOCLONAL ANTIBODIES TO PRUNUS NECROTIC RINGSPOT (NRSV), APPLE MOSAIC (ApMV), TOBACCO STREAK (TSV) AND ALFALFA MOSAIC (AMV) VIRUSES. E. L. Halk, H. T. Hsu and J. Aebig. ATCC, Rockville, MD 20852.

Eighteen somatic hybrid cell lines (hybridomas) that secrete antibody specific for TSV, AMV, NRSV and ApMV or to both NRSV and ApMV have been produced. Hybridoma clones produced antibodies of the IgG1, IgG2a, IgG2b and IgM subclasses. ELISA titers of hybridoma ascites against homologous antigen ranged from 1/12,500 to 1/39,000,000 and agar double diffusion (ADD) titers ranged from 1/270 to >1/2130. However, some monoclonal antibodies with an ELISA titer of >1/312,000 did not precipitate virus in ADD tests. Differential reactions occurred among a panel of NRSV and ApMV strains in indirect ELISA using NRSV and ApMV specific monoclonal antibodies. Antibodies from clone 70C9 reacted with all NRSV and ApMV strains tested. Antibodies from clone 46D5, but not clone 63F10, reacted with NRSV-G and the Danish plum line pattern strain of NRSV. The differential reactions suggest antigenic strain differences have been detected by these monoclonal antibodies.

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DETECTION OF MAIZE STRIPE VIRUS USING NONCAPSID VIRAL PROTEIN ANTISERUM AND INDIRECT ELISA. B. W. Falk and J. H. Tsai. University of Florida, Agricultural Research and Education Center, Belle Glade, 33430 and Agricultural Research and Education Center, Fort Lauderdale, 33314.

Maize stripe virus (MStpV) induced noncapsid viral protein (NCVP) is produced in much higher concentrations in infected plants (1-2 mg/g) than MStpV nucleoprotein (80-120 μ g/g). Therefore, antiserum was prepared to NCVP for MStpV indexing experiments. NCVP was isolated from infected plants using the method of Gingery et al. (Virology 112:99-108), and further purified by SDS-polyacrylamide gel electrophoresis or by incubation at 4 C for one week. Antiserum was prepared in rabbits and tested by immunodiffusion and enzyme-linked immunosorbent assay (ELISA). Antiserum reacted with sap from MStpV infected but not healthy plants when tested by immunodiffusion, but not by double-antibody sandwich ELISA. Good reactions were obtained, however, by indirect ELISA, but healthy tissue extracts gave relatively high background reactions (0.2 A_{405} for healthy vs 1.0 A_{405} for infected tissue extracts).

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MAXIMIZING THE SENSITIVITY OF A BEET WESTERN YELLOWS VIRUS ELISA SYSTEM. Adrianna D. Hewings and Cleora J. D'Arcy, Dept. of Plant Pathology, Univ. of Illinois, 1102 S. Goodwin, Urbana, IL 61801

A California isolate of BWVY was purified from *Capsella bursa-pastoris* using an improved extraction procedure. Average yields of 2 mg/kg were obtained by grinding tissue frozen in liquid N_2 in a Waring Blender first without and then with .05M PO_4 buffer, pH 6. The mixture was stirred for 24h at room temperature (RT) in .5% Na₂S₂O₅ and 1% Rohment-P, a macerating enzyme. The virus was finally purified by methods reported earlier. A high-titre (4096) antiserum was made and an ELISA system set up. No differences in sensitivity were observed when coating IgG's were incubated at 4C or RT overnight or at 37C for 2h. For sources with low levels of virus incubation of antigen at 4C was adequate but RT gave best results. Conjugate was added for 6h and substrate for 1h. Replicated comparisons of sensitivity and variability with and among plates of the same batch showed no differences at the 99% level. Inside and outside wells were also found to be homogeneous. Purified BWVY was detected at 12.5 ng/ml and in single late-instar *Myzus persicae*.

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PRETREATMENT OF POLYSTYRENE CUVETTES WITH STRONG BASES INCREASES SENSITIVITY OF ELISA FOR POTATO VIRUSES S, X AND Y. Paul Goodwin and E.E. Bantari, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Pretreatment of polystyrene cuvettes with strong bases improved sensitivity of the double antibody sandwich enzyme-linked immunosorbent assay (ELISA) for potato viruses S, X and Y. New cuvettes used in the Gilford[®] system soaked for 48 hr, in 5M NaOH mixed 1:1 with 95% ethanol or in 15M NaOH in water alone increased virus specific reactions (absorbance A_{405} of hydrolyzed substrate) an average of 0.590 for PVS, 0.152 for

PVY, and 0.320 for PVX over that of new nontreated cuvettes. Comparable pretreatment of cuvettes with 17M KOH increased virus-specific absorbancies A_{405} over those in new non-treated cuvettes by 0.249 for PVS, 0.266 for PVY and 0.640 for PVX. The hypothesis is that strong bases improved binding and reduced leakage of coating γ -globulins of these antisera on cuvette plastics.

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CHARACTERISTICS OF AN ELISA TEST TO DETECT LETTUCE MOSAIC VIRUS IN LETTUCE SEEDS. B.W. Falk and D.E. Purcifull. University of Florida, Agricultural Research and Education Center, Belle Glade, 33430, and Department of Plant Pathology, University of Florida, Gainesville, 32611.

The enzyme-linked immunosorbent assay (ELISA) was evaluated as an alternative to the *Chenopodium quinoa* test to index lettuce seed for lettuce mosaic virus (LMV). ELISA results indicated substantial variability in the titer of LMV in individual infected seeds. When healthy seed samples containing more than 100 seeds in 5 to 10 ml PBS-Tween buffer were tested, high non-specific absorbance values were common ($\bar{x}=0.17$). Non-specific reactions were found to decrease substantially by grinding the samples and incubating them overnight at room temperature before placing them in microtiter plates. Absorbance values of infected seeds were not affected by this treatment. ELISA tests of individual seeds gave the same seed transmission percentage for three seed lots as did the seedling grow-out and *Chenopodium quinoa* assay. The test was sufficiently sensitive to consistently detect one infected seed in a 500 seed sample.

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COMPARISON OF DIRECT AND INDIRECT ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA) IN THE DETECTION OF BEAN COMMON MOSAIC VIRUS. Wei-Young Wang, G.I. Mink, and M.J. Silbernagel. Washington State University, I.A.R.E.C., Prosser, WA 99350.

Direct and indirect ELISA were compared using nineteen strains of Bean Common Mosaic Virus (BCMV) and three antisera. A narrow range of strain specificity was found using direct ELISA. Seven of 19 isolates could be detected by anti-Type conjugate; two by anti-CR conjugate and only NY-15 detected by anti-NY15 conjugate. The indirect ELISA with a commercial preparation of goat-anti-rabbit IgG conjugate gave a much broader range of strain specificity. Any one of the three antisera detected all strains except NL-3 and NL-5 in crude extracts of infected leaf tissue. Because of the narrow specificity, direct ELISA can be used to differentiate certain strains or at least, serotypes. In contrast, indirect ELISA is useful for general surveys for BCMV infected plants. BCMV was detected in seed extracts by indirect ELISA test. A good correlation was found between the results of the seed assay and subsequent tests on the seedlings.

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IMPROVED DETECTION OF HETEROLOGOUS DOUBLE-STRANDED RNA (dsRNA) ANTIGENS BY THE INDIRECT METHOD OF THE SOLID-PHASE ENZYME IMMUNOASSAY. D. H. Zanzinger and S. M. Tavantzis, Dept. of Botany and Plant Pathology, University of Maine, Orono, ME 04469

The double antibody sandwich method of the enzyme-linked immunosorbent assay (ELISA) used for detection of dsRNA (Phytopathology 72:268) was modified with the objective to improve detectability of heterologous dsRNA antigens. Poly-L-lysine was used to directly bind the dsRNA molecules to the polystyrene plates. Antibodies (rabbit immunoglobulins, 10 μ g/ml) to polyinosinic: polycytidylic acid [p(I:C)] were then bound to the dsRNA, followed by the addition of enzyme-conjugated anti-rabbit immunoglobulins. Our data suggests that this system is very reliable for detecting a variety of plant virus replicative forms and dsRNA of fungal origin, whereas the previously described double antibody sandwich ELISA is designed primarily for detection of homologous dsRNA antigens. This test can be completed within a single day and utilizes a commercially available preparation of enzyme-labelled anti-rabbit immunoglobulins.

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DIFFERENTIATION AND SEROLOGICAL DETECTION OF CUCUMBER MOSAIC AND PEANUT STUNT VIRUSES. I. B. Ahmad and H. A. SCOTT, Dept. of Plant Pathology, Univ. of Arkansas, Fayetteville, AR 72701

Isolates of cucumber mosaic (CMV) and peanut stunt (PSV) viruses produced different symptoms in bean, cowpea, cucumber and tobacco. Limited proteolysis of these viruses (purified by a chloroform-butanol method) followed by analysis in DISC-PAGE resulted in peptide maps which showed major reproducible differences between the CMV and PSV groups and minor differences between isolates of CMV. In most agar gel media the serological

detection of CMV in crude sap resulted in straight precipitin bands characteristic of protein subunits. However, tests utilizing agarose made up in borate-EDTA plus Triton X-100 and sodium thioglycolate, and CMV extracts from leaves ground in the same buffer plus bentonite, resulted in curved bands characteristic of intact capsids. Agarose made up in phosphate plus Triton X-100 and sodium azide was effective in serologically detecting intact capsids of PSV in crude sap.

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SEROLOGICAL DIFFERENCE IN HELPER COMPONENT FROM POTATO VIRUS Y AND TOBACCO VEIN MOTTLING VIRUS-INFECTED PLANTS. D. W. Thornbury & T. P. Pirone, Dept. of Plant Pathology, University of Kentucky, Lexington, KY 40546

Antisera were prepared to partially purified helper component (HC) from tobacco plants infected with either tobacco vein mottling virus (TMV) or potato virus Y (PVY). The activity of TMV-HC or PVY-HC was completely abolished when HC preparations were treated with the homologous antiserum for 2 hr at 4°. In contrast, treatment of either TMV-HC or PVY-HC with the heterologous antiserum or antiserum to a comparable preparation from healthy tobacco had little or no effect on HC activity. It is likely that HC activity is due to either host proteins produced as a response to potyvirus infection or to virus-coded proteins. These results indicate that serologically distinct HC proteins are produced in response to specific potyvirus infection and favor the hypothesis that HC is virus-coded.

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VARIATION IN SEROLOGICAL AND INFECTIVITY VALUES OF VIRUS INFECTED LEAVES STORED AT VARIOUS TEMPERATURES. R.P. Singh, Agriculture Canada, Research Station, Fredericton, N.B., Canada E3B 4Z7

For virus assay of large numbers of field samples there is a need to develop the methodology for storage of leaf samples that will prevent the loss of their virus infectivity and serological properties. In the present studies, storage temperatures of 25, 4, -20 and -70 C were used and potato viruses A, Y, X, S and leafroll were tested. Serological tests were performed by enzyme-linked immunosorbent assay (ELISA) and the infectivity was tested using various local lesion hosts. It was observed that serological readings obtained with potato virus A, Y and leafroll were very similar at 25, 4 and -70 C, but very variable at -20 C. Similarly, infectivity of PVA and PVY were significantly reduced at -20 C. Potato viruses S and X were less affected by -20 C storage. Storage at -70 C was very reliable and maintained the infectivity and serological ability of all viruses at levels similar to that of fresh material.

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THE EFFECT OF SODIUM DEXTRAN SULFATE ON SOME SPHERICAL PLANT VIRUSES. J.H. Tremaine, W.P. Ronald, and E.M. McGauley, Research Station, Agriculture Canada, Vancouver, B.C. V6T 1X2.

Sodium dextran sulfate (NDS) was added to nine spherical plant viruses at pH 5.0 and at pH 7.5, with and without ethylenediamine tetraacetic acid (EDTA). The effect of NDS on these viruses was assessed by sucrose density gradient centrifugation, and by electron microscopy and protein analysis of fractions of these gradients. None of the viruses were affected by NDS at pH 5.0. Turnip yellow mosaic and cowpea mosaic viruses were unaffected by any of the conditions. At pH 7.5, bromo mosaic, carnation ringspot and turnip crinkle viruses were each dissociated into a component sedimenting at the same rate as RNA, as was tomato bushy stunt virus in the presence of EDTA. In EDTA at pH 7.5 three sobomoviruses (southern bean mosaic, sowbane mosaic and turnip rosette viruses) were dissociated into RNA by high levels of NDS (20% by weight) but in lower concentrations of NDS components sedimenting between the RNA and swollen virus were formed and they contained spherical T=1 particles and distorted T=3 particles.

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OCCURRENCE IN BEE-STORED POLLEN OF AN ENZYME-LIKE FACTOR WHICH DEGRADES INTACT VIRUSES. G. I. Mink, Anabel Cole, Washington State University, Irrigated Agriculture Research and Extension Center, Prosser, WA 99350.

When bee-stored pollen samples were added to purified suspensions of *Prunus* necrotic ringspot virus, prune dwarf virus or potato virus X and incubated at 42C, no virus could be detected by ELISA after 4-6 hrs and intact virus could not be detected in sucrose density-gradients. These viruses were not affected when incubated with or without bee-stored pollen

at temperatures below 20C or by incubation with hand-collected pollen at any temperature. Extraction and kinetic experiments suggested that bee-stored pollen contains an enzyme-like factor that is capable of degrading intact virus particles at temperatures above 30C. This factor was not found on hand-collected pollen.

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GENETIC COMPLEMENTATION BETWEEN CARNATION RINGSPOT VIRUS AND RED CLOVER NECROTIC MOSAIC VIRUS. S. A. Lommel and T. J. Morris, Dept. Plant Pathology, University of California, Berkeley, CA 94720.

A stable, recombinant hybrid virus was produced by co-inoculation of red clover necrotic mosaic virus (RCNMV) RNA-1 and carnation ring-spot virus (CRSV) RNA-2 into *Nicotiana glauca*. The hybrid exhibited a coat protein serologically identical to RCNMV. This data suggest that for CRSV and RCNMV the coat protein gene resides on the long RNA (RNA-1). Recovery of hybrid virions from infected plants was significantly lower than either of the parents, although analysis of replicative forms (dsRNA) and virus protein indicated no deficiency in replication and coat protein production. Electrophoretic analysis of dsRNA's from infected tissue show that CRSV, RCNMV and the hybrid produce three replicative forms. Two of the dsRNA's are twice the size of the genomic RNA's, whereas the third dsRNA is presumed to be subgenomic. Comparison of relative mobilities of replicative forms of the parents and the hybrid illustrate that RCNMV RNA-1 and CRSV RNA-2 are replicating in complement to produce a hybrid recombinant.

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MEASUREMENT OF GROWTH REDUCTION OF WHITE CLOVER BY CLOVER YELLOW VEIN VIRUS AND *CODINAEA FERTILIS*. C. Lee Campbell and J. W. Moyer, Department of Plant Pathology, N.C. State University, Raleigh, NC 27650.

Clover yellow vein virus (CYVV)-infected or CYVV-free plants from two clones (T7 and T17) of 'Tillman' white clover were grown in pasteurized loam:sand mix (3:1 v/v) infested or non-infested with *Codinaea fertilis*. Plant response was evaluated after 20 weeks in the greenhouse by measuring or counting flowers/plant, stolons/plant, stolon length, nodes/stolon, rooting nodes/stolon, and root and shoot weights. CYVV infection reduced stolon length and number and all weight measurements taken in one or more cases. Plants of clone T7 were more tolerant of CYVV infection than those of clone T17. *C. fertilis* infection also reduced some components of growth. Root rot induced by *C. fertilis* was generally greater in CYVV-infected than in CYVV-free plants. CYVV and *C. fertilis* appear to act additively in reducing growth of white clover.

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USE OF DIFFERENTIAL CULTIVARS TO ASSESS RELATIVE SEVERITY OF FOLIAGE DISEASES OF ALFALFA. F. A. Gray, Pl. Sci. Div., Univ. of Wyoming, Laramie, 82071.

Resistant and susceptible alfalfa cultivars, recommended by the USDA to characterize disease reaction, were used to measure relative severity of foliage diseases of alfalfa in six geographically diverse areas of Wyoming. Plots were rated for disease on a scale of 1-5 (1=none, 5=very severe). Spring black stem (SPBS), common leaf spot (CLS), downy mildew (DM) and yellow leaf blotch (YLB) were detected in test plots during 1981. The highest disease ratings (means) for SPBS, DM, CLS, and YLB at Laramie, Riverton, Afton, Powell, Torrington, and Sheridan were: 3.8, 3.8, 0, 0; 0, 2.8, 2.8, 0; 3.0, 4.2, 0, 0; 2.1, 1.9, 2.9, 0; 0, 0, 4.1, 0; and 0, 0, 0, 2.1; respectively. SPBS, DM and CLS were major diseases in plots grown under irrigation. Only one of the locations, however, had all three of these diseases. YLB was the major disease at the dry-land location and was not detected in irrigated plots. Diseases found in plots were the same as those observed in field surveys of test site areas.

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VERTICILLIUM ALBO-ATRUM ESTABLISHED ON ALFALFA IN WYOMING. D. A. Roth and F. A. Gray, Pl. Sci. Div., Univ. of Wyoming, Laramie, 82071.

Verticillium Wilt of alfalfa, caused by *Verticillium albo-atrum*, was found in Wyoming for the first time in the fall of 1981. The disease currently appears to be restricted to irrigated alfalfa in north central Wyoming. Pathogenicity tests conducted in the greenhouse showed alfalfa and sainfoin to be highly susceptible to the Wyoming isolate of *V. albo-atrum*. Four wk after inoculation the percent healthy, diseased, and dead plants of 9-wk-old alfalfa (cultivar 'Vernal') and sainfoin (cultivar 'Remont') were: 10, 30, 60, and 0, 50, 50, respectively.

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CYLINDROCLADIUM ROOT AND CROWN ROT OF ALFALFA IN HAWAII. J. J. Ooka and J. Y. Uchida, Department of Plant Pathology, University of Hawaii, Honolulu, Hawaii 96822.

Cylindrocladium spp. were consistently isolated from stunted and chlorotic or dead *Medicago sativa* plants from three sites in Hawaii. Black and sunken lesions were observed on the roots and in the crown area of diseased plants. A brown root rot was sometimes also present. Isolations from necrotic tissues produced four *Cylindrocladium* spp. and other fungi. *Calonectria crotalariae* made up 45% of the *Cylindrocladium* spp. isolated from plants collected from Waimanalo, 85% from Kohala and 100% from Wailua. *Cylindrocladium clavatum*, which accounted for 40% of the Waimanalo isolates, was not found at Kohala or Wailua. *Cylindrocladium scoparium* accounted for 2 and 4% of the Waimanalo and Kohala isolates respectively. Inoculation of cultivar 'Ranger' with *C. crotalariae* or *C. clavatum* produced slightly depressed hypocotyl lesions and dark brown to black root lesions. An average of 60% of the seedlings were killed by *C. crotalariae*, while *Cylindrocladium clavatum* killed 15% of inoculated seedlings.

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DISEASES OF THE BUFFALO GOURD, *CUCURBITA FOETIDISSIMA*, IN ARIZONA. M. E. Rosemeyer, B. H. Wells, and A. Zaid, Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

Buffalo gourd, a drought-resistant cucurbit native to the semi-arid western United States, has many potential economic uses as human food and as an energy source. In domestication field plots at the University of Arizona (1973-present), the causal agents of five previously unreported diseases of buffalo gourd have been isolated and identified: leaf mosaic caused by squash mosaic virus and cucumber mosaic virus; root rot caused by an *Erwinia carotovora*-*Fusarium solani* complex; root rot caused by *Phymatotrichum omnivorum*; and galling of roots caused by the root-knot nematode, *Meloidogyne javanica*. These results indicate that buffalo gourd is susceptible to a number of recognized plant pathogens.

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AREAL PATTERN OF RHIZOCTONIA CROWN AND ROOT ROT IN SUGARBEET FIELDS. L. J. Herr, Department of Plant Pathology, Ohio Agricultural Research and Development Center, Wooster 44691 and The Ohio State University.

In early summer, two sugarbeet fields with one to two *Rhizoctonia solani*-infected plants per primary infection focus (PIF) were selected. Disease increase in 20 PIF on four counting dates (late June to Sept.) was recorded in one field. Regression analysis of diseased plants per PIF on days gave the equation $Y=1.49+0.195X$, $R^2=0.56$ and $S.E.=0.019$. Average increase in diseased plants per PIF was 10.8 fold. Both fields were also divided into 10 sets of four double row plots 91.4 m in length. Numbers of dead and diseased plant doublets were counted on three dates (July to Sept.). The actual number of doublets exceeded the expected number by more than two times the S.E. in both fields on all counting dates. Thus, disease pattern was clustered (non-random) indicative of secondary plant-to-plant spread.

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DIFFERENTIATION OF *USTILAGO SCITAMINEA* ISOLATES IN GREENHOUSE TESTS. A. G. Gillaspie, Jr., R. G. Mock, USDA, ARS, Beltsville, MD 20705, and J. L. Dean, USDA, ARS, Canal Point, FL 33438.

Young shoots from germinated setts of seven sugarcane clones were inoculated by an injection technique with *U. scitaminea* teliospores obtained from Argentina, Florida, Hawaii, Taiwan, and Zimbabwe. Tests were done in containment greenhouse facilities (30 ± 1 C, c. 50% RH) at Frederick, Maryland. The spore suspension (5 x 10⁴ viable spores/ml) was injected twice into the meristematic region of each 8-12 cm shoot (Ferreira and Comstock. *Phytopathology* 71:873). Inoculated plants were incubated for 1-2 days at 30 C, potted in soil, and observed for 7 months. Six isolates could be differentiated on their virulence to clones CP 65-357, F 134, H 50-7209, H 68-1158, and NCo 310. The sugarcane clones varied in the time required for appearance of smut whips and the isolates varied in their virulence. This method will differentiate *U. scitaminea* isolates.

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ETIOLOGY OF ATYPICAL SYMPTOMS OF CHARCOAL ROT OF SUNFLOWER. S. M. Yang. USDA-ARS, P.O. Drawer 10, Bushland, TX 79012.

The typical symptom of charcoal rot of sunflower (*Macrophomina*

phaseolina [Tassi] Goid.) is gray discoloration of stem. Sunflower plants simultaneously infected with *M. phaseolina* and infested with *Cylindroclonus adspersus* (LeConte) larvae showed atypical symptom in the form of brown and black discoloration on stems. *Alternaria alternata* (Fries) Keissler, *Fusarium solani* (Martius) Saccardo and *Rhizopus arrhizus* Fischer were frequently isolated from the *C. adspersus* larvae and from the sunflower plants showing these atypical symptoms. Greenhouse grown sunflower plants inoculated with the three fungi alone or in combination with *M. phaseolina* exhibited atypical symptoms, but sunflower plants inoculated with *M. phaseolina* alone showed typical symptoms. Stem injuries by tunneling larvae prior to inoculation with the three fungi hastened the development of the atypical symptom. The results indicate that the three saprophytic fungi contribute to the development of atypical symptom on sunflower stalks infected with *M. phaseolina*.

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IN VITRO EFFECTS OF PREPLANT INCORPORATED HERBICIDES ON FUNGAL PATHOGENS OF COTTON SEEDLINGS. D. M. Ingram and K. W. Roy, Plant Pathology and Weed Science, P. O. Drawer PG, Mississippi State, MS 39762.

The effects of trifluralin, fluchloralin, profluralin and pendimethalin on the growth of three isolates each of *Rhizoctonia solani* (RS), *Fusarium oxysporum* (FO), *Pythium ultimum* (PU), and *Thielaviopsis basicola* (TB) were determined using herbicide-amended agar at 19C. At concentrations of 1-1,000 ug/ml a.i., all herbicides significantly reduced the growth of RS and PU. Concentrations of 50-1,000 ug/ml reduced the growth of TB. The greatest reductions in growth occurred at herbicide concentrations of 1,000 ug/ml. At that concentration, all herbicides reduced the growth of RS, PU and TB isolates by more than 75, 65 and 50%, respectively. Growth of one FO isolate was reduced by more than 35%. All four herbicides increased the growth of one FO isolate, with pendimethalin causing the greatest increase (37%). Trifluralin and pendimethalin increased the growth of another FO isolate, with trifluralin causing the greatest increase (14%).

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MORPHOLOGICAL AND PATHOLOGICAL STUDIES OF *COLLETOTRICHUM CAPSICI* AND *C. INDICUM* FROM COTTON BOLLS. Rodney Golden Roberts and J. P. Snow, Dept. Plant Path. & Crop Physiol., La. State Univ. Agric. Expt. Sta., Baton Rouge, LA 70803.

Colletotrichum capsici (Syd.) Butler & Bisby and *C. indicum* Dastur are falcate-spored species reported from cotton bolls. Morphology and pathogenicity of 4 isolates of each taxon were studied to determine if synonymy exists between the 2 species. Pathogenicity on unwounded cotton bolls was similar for all isolates. Plastic embedded thick sections from culture and host tissue showed marked differences in the degree of stromatic development among isolates. Mass isolates sectioned into distinct forms, indicating that these species are heterogeneous taxa which cannot be adequately separated. The name *C. capsici* should be used until further studies elucidate broader relationships among falcate-spored *Colletotrichums*.

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IN VITRO INHIBITION OF PLANT PATHOGENS BY *VERTICILLIUM DAHLIAE* AND *V. ALBO-ATRUM*. R. R. Romanko, Joy Jaeger, and A. Chernik. SWIREC, Parma, ID 83660, and Univ. of Olstyn, Olstyn, Poland

Plant pathogenic *Verticillium* spp. have shown inhibitory activity against a range of bacteria and fungi including other plant pathogens. This phenomenon was first detected in *Verticillium dahliae* isolated from vascular tissue of potato tubers. Microsclerotial colonies with scant aerial mycelium strongly inhibit fungi such as *Rhizoctonia*, *Alternaria*, *Colletotrichum*, *Sclerotinia*, and *Fusarium*, at a distance. This inhibition is readily demonstrable when *V. dahliae* is seeded on thin layer, low nutrient agar plates a few days prior to a challenge seeding. Detectable activity of inhibitor in solution was obtained from cultures in liquid PDB and YDP broth. These solutions inhibited bacterial plant pathogens in the genera: *Agrobacterium*, *Corynebacterium*, and *Xanthomonas*. *V. dahliae* isolates from mint, hops, eggplant, alfalfa, sugarbeets and cotton also produced this inhibitor, as did *Verticillium albo-atrum* from alfalfa. No distinct inhibition could be shown from cultures of *V. nigrescens*, *V. nubilum*, *V. tricorpus*, or *V. lateritium*.

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VERTICILLIUM DAHLIAE GROWTH IN THE PRESENCE OF AMINO ACIDS. D.R. Duncan and E. B. Himelick, Department of Plant Pathology, University of Illinois and Illinois Natural History Survey, Urbana, IL 61801.

Verticillium dahliae when grown on a Czapeks-Dox medium, in

which amino acids were substituted for NaNO_3 , yielded variable conidial and dry weight production depending upon what amino acid or combination of amino acids was present in the medium. Asparagine, glutamine and arginine stimulated the greatest conidial production. Proline, valine and a 50:50 mixture of asparagine and arginine stimulated the greatest dry weight production. Similar responses to individual amino acids were noted when the amino acids were added to sugar maple sap and the amended sap was used as a growth medium. The nitrogen in tree sap is predominantly in the form of amino acids and the types and quantities of these amino acids vary with environmental stresses. The response of *V. dahliae* to these amino acids could influence the development of *Verticillium* wilt of trees and may be an explanation for variability in disease progression that has been previously reported.

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CHLAMYDOSPORE FORMATION AND GERMINATION IN SELECTED ISOLATES OF *PHYTOPHTHORA PARASITICA*, *P. CINNAMOMI*, *P. PALMIVORA* MF1 AND MF4. Azizollah Alizadeh and Peter H. Tsao. Department of Plant Pathology, University of California, Riverside, CA 92521.

Chlamydospore formation and germination were compared in 12 isolates representing 4 species or morphological forms (MF) of *Phytophthora*. Abundant chlamydospores formed in 3 weeks at 18 C with most isolates following the transfer of fungal mats grown in a liquid medium to deep water. This method revealed the hitherto unreported formation of chlamydospores in the macadamia isolate of '*P. palmivora*' MF4 = (*P. capsici* of Kunitomo et al.). The mean chlamydospore diam of 12 isolates ranged 27-40 μm , with smaller ones formed by 6 isolates of '*P. palmivora*' MF4. Viability of chlamydospores ranged 27-97%; the higher values were noted for '*P. palmivora*' MF4 and lower for *P. cinnamomi*. Germination of viable spores of all isolates after 10 hr was higher in carrot broth than in buffered glucose-asparagine solution and ranged 15-72%. If '*P. palmivora*' MF4 is indeed *P. capsici* as suggested by others, our data constitute the first observation of chlamydospore formation in this species.

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INDUCTION OF PHOSPHOLIPASE ACTIVITY IN *PHYTOPHTHORA INFESTANS*. Robert A. Moreau. Eastern Regional Research Center, USDA, ARS 600 E. Mermaid Lane, Philadelphia, PA 19118

Phytophthora infestans (race 4) grew well on lipid rye steep medium (Can. J. Plant Sci. 44:583) that was modified by omitting glucose. The fungus produced an extracellular phospholipase B when low levels of phospholipids (5 mg/100ml) were added to the modified medium, but no phospholipase activity was observed when phospholipids were omitted. Very little phospholipase activity was detected when the fungus was grown on the complete (+ glucose) medium, even after the addition of phospholipids. Phospholipase activity also was induced by triacylglycerols, wax esters, steryl esters, and Tween 20. Maximal levels of phospholipase activity were detected in the media 7-14 days after incubation. A sensitive radioisotopic assay employing phosphatidylcholine (dipalmitoyl- ^{14}C) was used to measure phospholipase activity. The extracellular phospholipase exhibited an optimum activity at pH 9.0 and was not inhibited by EDTA.

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EFFECTS OF WATER POTENTIAL ON GROWTH AND RESPIRATION IN *PHYTOPHTHORA CRYPTOGEA* AND *FUSARIUM MONILIFORME*. D.M. Woods and J. M. Duniway, Department of Plant Pathology, University of California, Davis, CA 95616

Mycelial growth and respiration rates of *Phytophthora cryptogea* and *Fusarium moniliforme* were evaluated in media adjusted to various solute potentials (ψ_s). Growth by *P. cryptogea* in standing liquid cultures was reduced by 50% at -9 to -14 bars ψ_s while a 50% reduction in the growth of *F. moniliforme* occurred at -100 to -120 bars, the exact values depending on the media and solutes used. At equivalent ψ_s values, osmotic decreased growth in the following order: sucrose < mannitol < NaCl = KCl = sea salt < MgSO_4 < Na_2SO_4 = PEG 300. Growth measured as fresh weight was generally decreased more by slight decreases in ψ_s than was growth in dry weight. Both fungi grew at lower ψ_s values in complex liquid media than in defined media, and at still lower ψ_s values when growth was measured as colony diameter on solid media. Respiration rates were significantly higher for mycelial mats grown at low ψ_s values.

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EFFECT OF LIGHT ON SPORANGIUM FORMATION, MORPHOLOGY, ONTOGENY, AND CADUCITY OF *PHYTOPHTHORA CAPSICI* AND THE BLACK PEPPER ISOLATES OF '*P. PALMIVORA*' MF4. Azizollah Alizadeh and Peter H. Tsao. Department of Plant Pathology, University of California, Riverside, CA 92521.

The influence of continuous light on sporangium formation, morphology, ontogeny, and the detachment of sporangia was studied. Sporulation on carrot agar was enhanced under continuous light, whereas no sporangia formed in the dark. Sporangia were formed in the dark when the mycelial disc-in-water technique was employed. The sporangial arrangement was fan-shaped in the light with all black pepper (BP) and those *P. capsici* (PC) isolates that have elongated sporangia with a high L/B ratio, tapered bases and long pedicels. In the dark, irregular sporangium ontogeny resulted with round-based sporangia (lower L/B ratio) which were not easily dislodged from the vegetative mycelium. The use of sporangia L/B ratio as a taxonomic criterion in identifying PC and BP appears to have little validity under these conditions. Furthermore, the sporangial morphology, ontogeny, and the degree of caducity are different in the light and dark.

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THE EFFECT OF NITROGEN SOURCE AND PH ON OXALIC ACID PRODUCTION AND PATHOGENICITY OF *SCLEROTINIA MINOR*. C. L. Patterson, C. M. Waters, and R. G. Grogan, Department of Plant Pathology, University of California, Davis, CA 95616

Oxalic acid (OA) production by *Sclerotinia minor*, determined by potassium permanganate titration, was positively correlated with pathogenicity in an *in vitro* bean leaf-disc assay. Acid production was greatest when the fungus was grown on an inorganic salts medium containing peptone, L-asparagine, L-glutamine, or L-threonine as a nitrogen source, and 1% glucose as a carbon source. Little or no OA was produced when the nitrogen sources were ammonium nitrate, potassium nitrate, and urea. OA production was also influenced by the pH of the medium; at pH 6.0, acid production was greater than at pH 5.0 or 4.0. Pathogenicity of *S. minor* *in vitro* was positively correlated with OA production suggesting that OA plays a role in the initiation of disease.

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BIOCHEMICAL DETERMINATION OF FUNGAL SPORE VIABILITY
S. Q. Yu and E. J. Trione, Dept. Botany and Plant Path., Oregon State University, Corvallis, Oregon 97331

Germination is commonly used to monitor spore viability, but many fungal spores do not germinate readily. Several methods based on enzyme activity were developed to monitor viability. Methylumbelliferyl (MU) substrates (-glucoside or -phosphate) are non-fluorescent, but when hydrolyzed by glucosidase or phosphatase enzymes, the highly fluorescent MU moiety is produced and can be readily seen and measured. Preparations from 25 µg of living spores contain sufficient glucosidase to hydrolyze MU-glucoside at 37°C in 2 hours. Dead spores will not bring about the enzymatic fluorescent reaction. The ATP content (measured by the Luciferase reaction) is also much higher in living than in dead spores. Hydration often causes the ATP content of living spores to increase greatly. Approximately 40 µg of spores are required to measure the ATP level. We have used these methods to test the viability of the spores of these fungi: *Tilletia controversa*, *Tilletia caries*, *Ustilago scitaminea*, *Puccinia striiformis*, *Pisolithus tinctorius*, *Elaphomyces* sp., and *Rhizopogon colossus*.

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AUTOFLUORESCENCE OF FUNGAL PROPAGULES: ITS RECIPROCAL RELATIONSHIP TO VIABILITY AND APPLICATION TO PATHOLOGICAL RESEARCH. Huey-Iwa Wu, and H. L. Warren. USDA, ARS, and Dept. Botany & Plant Pathology, Purdue University, West Lafayette, IN 47907.

This research determined if fungi emitted autofluorescence, and how it could be used as an experimental tool. Conidia, ascospores, or mycelia of fourteen fungal species were examined with fluorescent microscopy for autofluorescence. Cultural conditions were used to study natural fluorescence and physiological, biological, chemical, and physical stresses were used to study induced fluorescence. Qualitative observations gave a perfect negative correlation between fluorescence and germination. Quantitative assays also gave a very high negative correlation ($r = -0.982$). This reciprocal relationship between fluorescence and viability permits the fluorescent microscopic technique to be used as a tool with many advantages. This technique which reflects true viability can complement or replace germination tests. The technique was also extensively evaluated for mycological and pathological studies of *Colletotrichum graminicola* conidia.

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POTASSIUM ION INDUCES BEAN RUST GERMLINGS TO DEVELOP INFECTION STRUCTURES. R. C. Staples, H. J. Grambow, and H. C. Hoch, Boyce Thompson Institute for Plant Research, Ithaca, NY 14853; Institute für Biologie III, RWTH Aachen, West Germany; and

New York State Agricultural Experiment Station, Cornell University, Geneva, NY 14456.

Uredospores of the bean rust fungus (*Uromyces phaseoli*) germinating on agar buffered with 50-mM Tris-HCl were induced to form complete infection structures by a variety of potassium salts. The optimum pH was 7.0, and the ED_{50} was 10 mM. Potassium appears to stimulate bean rust germlings to reduce nucleotides for replication of nuclear DNA since deoxyadenosine and deoxycytidine separately added also stimulate differentiation at low concentrations of K^+ . Other interpretations of the potassium ion effect are possible and will be discussed. Similar results with wheat stem rust (*Puccinia graminis tritici*) will also be presented.

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BIOLOGICAL CONTROL OF CHESTNUT BLIGHT WITH NATIVE MICHIGAN HYPOVIRULENT STRAINS OF *ENDOTHIA PARASITICA*. D. W. Fulbright and W. H. Weidlich, Department of Botany & Plant Pathology, Michigan State University, East Lansing, MI 48824

Naturally occurring and laboratory converted hypovirulent strains of *E. parasitica* were tested in 1981 for their ability to control blight in a stand of chestnut trees (*Castanea dentata*). Cankers were initiated with virulent cultures from the same stand, by placing *E. parasitica* in corkborer holes cut into the bark. Five weeks later, hypovirulent isolates were inoculated around the expanding canker. Measurements indicated that most cankers stopped expanding after reaching the hypovirulent inoculum. There was a similar test with natural cankers; approximately 90% of the treated cankers ceased expansion. Callus tissue formed around several cankers. One hypovirulent strain (GH2) sporulated profusely at the end of the season and studies are underway to determine the fate of the spores. An isolate resistant to a fungicide (Olin TerraCoat L205) was found; this isolate may be useful in studies on the spread of the fungus in Michigan.

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EFFECTS OF SELECTED NORTH AMERICAN AND ITALIAN VIRUS-LIKE CYTOPLASMIC HYPOVIRULENCE AGENTS ON NORTH AMERICAN AND ITALIAN STRAINS OF *ENDOTHIA PARASITICA*. J. E. Elliston, The Connecticut Agricultural Experiment Station, Box 1106, New Haven, CT 06504.

Each of four North American virus-like cytoplasmic hypovirulence (CH) agents, H_{M1} , H_{M2} , H_{T2} , and H_{V1} , and two Italian agents, H_{I1} and H_{I2} , were transmitted by hyphal anastomosis on agar in to each of 22 isolates of *Endothia parasitica*. Half of the isolates were from North America and half from Italy, and eleven vegetative compatibility groups were represented. Three of the North American and five of the Italian isolates had been naturally infected with CH agents and freed of these by single conidial isolation. The others had no known history of natural infection with CH agents. Each agent caused a characteristic disease in each isolate, regardless of geographical origin or previous history. These results suggest that the wide variation found among CH strains may be due more to variation among CH agents than to differences in strains of *E. parasitica*.

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UNUSUAL PACKAGING OF A NAKED VIRAL GENOME ASSOCIATED WITH HYPOVIRULENCE OF *ENDOTHIA PARASITICA*. D. R. Hansen, K. Gillies, and N. K. Van Alfen, Department of Biology, UMC 45, Utah State University, Logan, Utah 84322.

Two membrane-bound particulate fractions can be isolated from extracts of *Endothia parasitica* strain 113. Composition of both fractions are similar with the exception of the presence of dsRNA in the denser one. Assays indicate a carbohydrate content of over 40%, while protein is less than 30% of the total dry weight of the particle. Radiolabelling studies indicate that no one polypeptide is present in an amount sufficient to be a capsid. The hexoses that are in these particulate fractions were found to be the same ones present in the fungal cell wall. Our data suggest that these particles are not typical mycoviruses but rather are fungal vesicles that package the dsRNA. These vesicles are similar to those involved in fungal cell wall synthesis. The dsRNA associated with transmissible hypovirulence of *E. parasitica* thus appears to be the genome of a multi-component virus that has lost the need for a capsid.

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GROWTH EFFECTS OF JUGLONE AND HYDROJUGLONE GLUCOSIDE ON PATHOGENS AND NON-PATHOGENS OF BLACK WALNUT. Steven Cline and Dan Neely, Department of Plant Pathology, University of Illinois and Illinois Natural History Survey, Urbana, IL 61801.

The growth of *Gnomonia leptostyla*, *Cylindrosporium juglandis*, *Cristulariella moricola* (black walnut pathogens), and *Gnomonia*

platani, G. quercina, Sclerotinia sclerotiorum (non-pathogens) was assayed against commercially prepared juglone (5-hydroxy-1, 4-naphthoquinone) in shake culture. ED₅₀ values were evaluated by dry weight measurements. Juglone inhibited mycelial growth at concentrations between 0.2 and 0.5 ppm in 4 of 6 fungi tested. Gnomonia leptostyla and C. juglandis were more tolerant to juglone with ED₅₀ values ranging from 1.5-4.5 ppm. Hydrojuglone glucoside (HJG) (4,8-dihydroxy-1-naphthalenyl-B-D-glucopyranoside), an endogenous precursor to juglone, was isolated from nut hulls of black walnut. It was assayed in potato dextrose broth shake culture with G. leptostyla. HJG progressively stimulated fungal growth up to 750 ppm. Recent evidence implicates juglone as a factor in disease resistance to pecan scab and it may be similarly effective against G. leptostyla in black walnut.

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RANGE-WIDE STUDY OF DECAY RESISTANCE OF BLACK WALNUT. John H. Hart, C. S. Thomas and T. L. Kamps. Depts. of Botany & Plant Pathology and Forestry, Mich. State Univ., E. Lansing, MI 48824

To determine the variability in decay resistance throughout the range of black walnut, Juglans nigra L., and from different locations within a tree, 3 mature black walnut trees were collected from 8 states: IL, IN, KS, KY, MI, MS, PA, and WI. Blocks 1.9 cm³ were cut from 3 vertical positions (1, 5 and 9 m above ground line) from the sapwood (SW), outer heartwood (HW), middle HW and inner HW. Using the agar-block method, blocks were exposed to Coriolus vesicolor, Poria placenta or Schizophyllum commune for 10 weeks at 25 C. SW was more decay susceptible than the HW. There were no significant gradients in decay resistance of the HW associated with radial or longitudinal position. HW of individual trees from the same state varied significantly in decay resistance, attributable mainly to genetic variability. Similarities in HW decay resistance of trees collected in different states suggest that site is not an important factor in determining decay resistance. Growth rate of the trees was not found to be indicative of HW decay resistance.

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OCCURRENCE AND FREQUENCY OF CERATOCYSTIS FAGACEARUM ON FREE-FLYING NITIDULIDS IN MINNESOTA. J. Juzwik and D.W. French, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Washings from 1043 free-flying nitidulids (Coleoptera: Nitidulidae) attracted to two types of odor bait traps from March 30 - June 26, 1981, in an oak wilt area in Minnesota were plated on agar media to detect the presence of Ceratocystis fagacearum (Bretz) Hunt. The fungus was isolated from seven beetles trapped during 5 of 13 weeks. The percentage of positive isolation based on total number of beetles tested per trap type ranged from 1.5 to 22.2. Isolation of the fungus from a beetle collected on April 1 suggests that overland transmission of oak wilt could occur in early spring. The maximum number of nitidulids trapped coincided with the time of maximum mycelial mat production in the infection center; however, isolation of C. fagacearum occurred 2 to 3 weeks after large numbers of mycelial mats were detected. SEM observation of beetles collected from mycelial mats in May suggest that ascospores were the predominant spore type on the free-flying beetles.

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CONTROL PROGRAM FOR THE PINWOOD NEMATODE AT THE MORTON ARBORETUM. Thomas L. Green, The Morton Arboretum, Lisle, Illinois 60532.

The pinewood nematode (PWN), Bursaphelenchus xylophilus, was discovered at the Morton Arboretum, Lisle, IL, in 1979. In 1981 the nematode was found in Pinus armandii, P. cembra, P. contorta, P. flexilis, P. mugo, P. nigra, P. ponderosa, P. resinosa, P. sylvestris, and P. tabulaeformis. This is the first known record from P. armandii. The PWN was found in 38 of 119 trees sampled, 28 (74%) of the 38 were P. sylvestris. The nematode has not been found in P. strobus, a known host. The control program at the Arboretum involves a survey of the Pinaceae to verify occurrence of the disease, developing reliable sampling methods to locate diseased trees, identifying nematode vectors and their periods of activity, removal of dead trees to reduce the beetle vector population, and destruction of diseased trees before 1 June by burning to prevent spread of the disease. Monochamus carolinensis, the Carolina pine sawyer, seems to be the only PWN vector at the Arboretum. There is evidence that this program is reducing loss of pines.

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SELECTIVE DELIGNIFICATION OF OAK BY INONOTUS (POLYPORUS) DRYOPHILUS. L. Otjen and R.A. Blanchette, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Inonotus (Polyporus) dryophilus selectively delignified oak

xylem in localized areas. White pockets consisted of delignified tissues that lacked middle lamellae and degradation of the cell walls was characterized by the presence of cellulosic microfibrils. Chemical analyses showed delignified tissue to be composed of 93.5% total sugars and 2.5% lignin; whereas sound wood contained 64.5% and 24.9%, respectively. Histological and scanning electron microscopy techniques demonstrated that selective delignification occurred in axial parenchyma cells surrounding vessels of earlywood and latewood. Flame-shaped tracts of vessels with accompanying axial parenchyma, present throughout the latewood, provided avenues for radial movement of I. dryophilus. Occluded latewood fibers and medullary rays were often left intact forming borders between white pockets.

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BUTT ROT IN OAK IN THE CENTRAL HARDWOOD REGION. Frederick H. Berry, NEFES, Forestry Sciences Laboratory, Delaware, Ohio 43015.

Butt rot was found to be responsible for considerable cull in oak stands in the central hardwood region. Rot was present in the butt log of 29 percent of 2,278 oaks examined. Volume loss due to cull was greatest in scarlet oak (Quercus coccinea Muench.), followed in order by black oak (Q. velutina Lamarck), chestnut oak (Q. montana Willd.), northern red oak (Q. rubra L.), and white oak (Q. alba L.). Although 26 species of decay fungi were isolated and identified, two species Laetiporus (Polyporus) sulphureus (Bull. ex Fr.) Bond. & Sing., and Poria cocos (Schw.) Wolf, caused 26 percent of all infections and almost 43 percent of the cull volume. Fire wounds were the major infection court for decay fungi, serving as the entry point for 60 percent of the infections. Rot fungi also entered through parent stumps (22 percent), roots (13 percent), mechanical injury (2 percent), and unknown, 3 percent. Over 68 percent of the cull volume was associated with fire wounds.

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DROUGHT AS A CAUSE OF OAK DECLINE AND MORTALITY ON THE SOUTH CAROLINA COAST. F. H. Tainter, T. M. Williams, and J. B. Cody, Associate Professors, Department of Forestry, Clemson University, Clemson, SC 29631.

Extensive oak decline and mortality evident in 1981 along the South Carolina coast from Georgetown to Myrtle Beach was apparently caused by two severe summer droughts, the first in 1978, from which resulted only scattered incidences, and another more severe drought in 1980 which caused rapid decline and death of urban and forest trees in spring of 1981. A lowered water table has been blamed as the cause of injury to the shallow-rooted red oak species most affected. These included willow oak (Quercus phellos), laurel oak (Q. laurifolia), water oak (Q. nigra), and southern red oak (Q. falcata). Hypoxylon atropunctatum was an evident early colonizer of both the declined and dead trees.

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VARIATION AMONG ISOLATES OF BOTRYODIPLODIA HYPODERMIA FROM CANCKERS ON SIBERIAN ELM. J. M. Krupinsky, USDA-ARS, Northern Great Plains Research Center, P.O. Box 459, Mandan, ND 58554.

Isolates of Botryodiplodia hypodermia were obtained from Siberian elm cankers which were collected from the northern Great Plains. Variation in spore type and virulence of 218 isolates were examined. Seven percent of these isolates were atypical. Spores from atypical isolates were slightly narrower and longer than spores from typical isolates; however, the two types could not be differentiated by spore size. Approximately 50% of the spores from mature cirrhi of atypical isolates were septate; spores of typical isolates were aseptate. Atypical isolates were less virulent than typical isolates. Branches above the point of inoculation were killed on 20% of 132 branches inoculated with atypical isolates and on 73% of 266 branches inoculated with typical isolates. Atypical isolates should not be used in evaluating germplasm. Several typical isolates should be used to evaluate Siberian elm germplasm for resistance.

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INHIBITION OF DECAY IN BACTERIAL-STAINED WOOD OF AMERICAN ELM (ULMUS AMERICANA L.). J.S. Coleman, R.J. Campana, and C.W. Murdoch. Department of Botany and Plant Pathology, University of Maine, and New England Plant, Soil and Water Laboratory, USDA-ARS, Orono, ME 04469.

Differential rates of decay in sapwood and bacterial-stained heartwood of elm were evaluated in an agar block test. Average moisture content of each wood type was determined by oven dry weight of 10 blocks. Test blocks were obtained from a single,

freshly-cut elm, cut to 2.54 cm² x 1.27 cm, surface-sterilized or autoclaved and inoculated with *Corioliolus versicolor* or *Lenzites trabea* on malt extract agar in plates. Eighty blocks were tested, 40 from stain-free sapwood and 40 from bacterial-stained heartwood. Data on weight loss was obtained after incubation for three months at 22 C. Weight loss of sapwood exceeded that of heartwood ($P=0.05$). The data indicate that stained heartwood is more resistant to decay than unstained sapwood.

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ANTAGONISM OF FUNGAL GROWTH BY ACTINOMYCETE ISOLATES FROM *ULMUS AMERICANA*. J.G. O'Brien and R.A. Blanchette, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Nearly 400 actinomycete isolates recovered from the bark and xylem of *Ulmus americana* were tested in culture for antagonistic effects on *Ceratocystis ulmi*. Each actinomycete isolate was streaked down the center of a petri dish containing mycological agar (Difco). Four days later, a mycelial plug of an aggressive isolate of *C. ulmi* was placed at the edge of each plate, perpendicular to the actinomycete streak. Radial growth of the *C. ulmi* was compared to controls at 2-day intervals. At 10 days, inhibition of radial growth ranged from 48 to 95%, averaging 74%. Each of the actinomycete isolates tested inhibited the growth of *C. ulmi*. *Coniophora puteana*, *Schizophyllum commune* and *Ceratocystis fagacearum* were also inhibited. Actinomycetes may play an important role in the saprophytic survival of *Ceratocystis ulmi*. Further research involving the potential of actinomycetes in biological control of Dutch elm disease is warranted.

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THE DISTRIBUTION AND RELATIONSHIP TO WOUNDING OF WETWOOD IN AMERICAN ELM. C.W. Murdoch and R.J. Campana, N.E. Plant, Soil and Water Laboratory and Department of Botany and Plant Pathology, University of Maine, Orono, ME 04469.

A study was made to evaluate the location and distribution of wetwood columns in elm stems, including their relation to the injection of chemicals. Data were taken by inspection from 35 large, injected, 35 large, noninjected, and 40 small, noninjected trees on: cross-sectional area, volume and surface to volume ratio of wetwood columns; number, type, depth and location of bleeding and nonbleeding wounds; and relationship of tree age to visible wetwood development. One year injections did not significantly increase cross-sectional area or volume of wetwood. Increased depth of injection wounds was associated with higher frequency of bleeding. Regression line analysis showed that the number of wetwood-free rings increased with stem age in large injected ($r=0.88$) and noninjected ($r=0.92$) trees of 60 years or younger.

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FOMES FRAXINOPHILUS STEM DECAY OF GREEN ASH IN NEBRASKA WOODLANDS. Jerry W. Riffle, Edward M. Sharon, and Mark O. Harrell. Rocky Mountain Forest and Range Exp. Stn., Forestry Sciences Laboratory, Univ. Nebr., Lincoln, 68583; USDA Forest Service, Region 2, Lakewood, Colorado 80225; and Mark O. Harrell, Dept. Forestry, Fisheries, and Wildlife, Univ. Nebr., Lincoln 68583, respectively.

Incidence of *Fomes fraxinophilus* stem decay of *Fraxinus pennsylvanica* in native woodlands in Nebraska was determined by examination of 7066 living trees in 360 plots in 10 multi-county forest inventory units from April 1979 to June 1981. Based on occurrence of sporocarps, infected trees were found in 56% of the plots and in 86% of the counties. Incidence of infected trees ranged from 3.3% in southeastern Nebraska to 21.9% in northwestern Nebraska. Incidence of infected trees in diameter classes 2.5 to 12.6, 12.7 to 45.7, and over 45.7 cm was 3.8, 14.8, and 42.3% respectively. We conclude that 16.2% of an estimated population of 473,000 living *F. pennsylvanica* in Nebraska woodlands are infected with *F. fraxinophilus*.

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CANKER DISEASE OF HONEY LOCUST CAUSED BY *Nectria cinnabarina*. P.J. Bedker, R.A. Blanchette and D.W. French. Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Nectria cinnabarina causes a serious canker disease of honey locusts in Minnesota. Bark wounds were made on eight honey locusts 5 to 8 cm DBH; each tree received 4 control and 4 inoculated wounds that were spiraled up the main stem of the tree. Eleven weeks after inoculation all inoculated wounds had formed cankers whereas, none of the control wounds developed cankers. Surveys throughout the St. Paul - Minneapolis metropolitan area indicated that 20% of boulevard honey locusts had cankers caused by *N. cinnabarina*. *Thyronectria austro-americana*,

a common canker-causing pathogen of honey locust, was isolated only once and does not appear to be important in Minnesota.

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OPTIMAL IN VITRO CONDITIONS FOR GROWTH, AND SPORE RELEASE AND GERMINATION OF *THYRONECTRIA AUSTRO-AMERICANA*. W. R. Jacobi, K. J. DeMott, P. A. Moon, and T. S. Naumann. Dept. of Botany and Plant Path., Colo. State Univ., Fort Collins, Colorado 80523.

Thyronectria austro-americana causes basal, stem, and branch cankers alone or in conjunction with a nectria-like canker fungus resulting in mortality of honeylocusts. Little is known of the pathogen's means of dispersal and infection in Colorado. Effects of various temperatures and carbon and nitrogen sources on the growth of single conidium cultures were assessed by liquid-shake and agar culture. Growth occurred between 10-40 C with an optimum of 25-30 C and over a wide range of carbon and nitrogen sources. Spore release from pycnidia was affected more by moisture than temperature and occurred within seconds after application of free water. The effects of environmental parameters on conidial germination were assessed by an agar coated microscope slide procedure. The data from this study suggest that *T. austro-americana* is capable of surviving, re-releasing spores and germinating under a wide range of nutritional and environmental conditions.

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INCIDENCE OF *GLOEOSPORIUM PLATANI* AND *PHOMOPSIS SCABRA* IN ASYMPTOMATIC SYCAMORE TISSUE. Vernon Ammon and Stephen Vann. Department of Plant Pathology and Weed Science, Mississippi State University, Mississippi State, MS 39762.

Incidence of *Gloeosporium platani* and *Phomopsis scabra* in asymptomatic tissues removed from 5 to 7-yr-old American sycamore trees growing in upland and delta plantations was determined for two growing seasons. In 1980, the incidence of *G. platani* in the delta plantation ranged from 3% in April to 52%, 42%, and 53% in June, August, and September respectively. Incidence in the upland plantation was 61% in April followed by 28%, 22%, and 22% in June, August, and September respectively. *Phomopsis scabra* incidence for the same growing seasons averaged 75% and 86% in the delta and upland plantations respectively. In 1981, incidence of *G. platani* decreased to an average of 12% in the delta location and to 7% in the upland site, whereas *P. scabra* was isolated from an average of 69% of trees sampled at both locations.

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DECAY FOLLOWING THINNING SWEETGUM SPROUT CLUMPS. F. I. McCracken, Southern Forest Experiment Station, P. O. Box 227, Stoneville, MS 38776.

Butt rot in living sweetgum (*Liquidambar styraciflua*) stems was observed 5, 10, 15 and 21 years after the twin was cut in a 50-year-old sprout origin stand near Tallulah, LA. There was no decay in the standing stems 5 years after thinning and decay was insignificant 10 and 15 years after thinning. Twenty-one years after thinning, extensive decay had developed in the standing stems from cut stubs which had longitudinal split down the side during thinning. However, decay was insignificant when clean horizontal cuts were made. Clean cut stubs had grown over and contained water, decay and anaerobic bacteria. Split stubs did not retain water and the decay had spread into the remaining stem. A succession of decay fungi was observed. Decay risk associated with thinning sprout clumps can be eliminated by undercutting and care to prevent splitting during felling.

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PROGRESSION OF BUTTERNUT CANKER IN A BUTTERNUT PLANTATION. N. Tisserat and J. E. Kuntz. Dept. Plant Pathology, University of Wisconsin, Madison, WI 53706.

Progression of butternut canker, caused by *Sirococcus clavignenti-juglandacearum*, was followed in an isolated, 400-m-long, 10-year-old butternut (*Juglans cinerea* L.) plantation. Between 1976 and 1981, 60 trees at one end of the stand were branch inoculated. Cankers with hyphal pegs and pycnidia were produced on all inoculated trees. Of the 939 non-inoculated trees, 10.2% were diseased by October 1980. Trees with natural cankers increased to 19.4% by October 1981 and were located throughout the plantation. Most of the naturally infected trees had only branch mortality, but 38% also had one or more stem cankers within 30 cm of ground level. These basal cankers, often originating at branch stubs, progressed into the root collar and tap root. Natural infection of black walnuts (*J. nigra* L.), scattered in the plantation, was not detected. However, in another plantation, *S. clavignenti-juglandacearum* was isolated from a basal canker on a declining black walnut tree.

AN AUXOTROPHIC MUTANT OF *COCHLIOBOLUS HETEROSTROPHUS* CAN CAUSE A CONDITIONAL EPIDEMIC ON SUSCEPTIBLE CORN. Robert C. Garber, William E. Fry and O.C. Yoder, Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Growth of an induced auxotrophic mutant of *C. heterostrophus* (*Helminthosporium maydis*) race T was conditional, both *in vitro* and in the field. If supplied with its required nutrient (histidine), the auxotroph grew normally under laboratory conditions and caused an epidemic in field plots of T-cytoplasm corn; if the nutrient was withheld there was no growth on defined medium and no field epidemic. The severity of the epidemic caused by the auxotroph, as measured by area under the disease progress curve, was approximately 25% of that caused by a near-isogenic strain (7 backcrosses) of wild-type *C. heterostrophus* race T. The auxotrophic strain was readily recovered from field plots of corn 56 days after inoculation by sampling foliar lesions and plating onto minimal vs complete medium to verify auxotrophy. Auxotrophy may be useful both as a field marker in epidemiological research and as a tool to experimentally manipulate population growth in the field.

PATHOGENIC VARIATION IN ISOLATES OF *SEPTORIA GLYCINES* IN THE FIELD. T. A. Kamicker and S. M. Lim, Department of Plant Pathology, University of Illinois, Urbana, IL 61801.

Field plots of Williams 79 soybean were inoculated with 12 isolates of *Septoria glycines* from 12 different geographical locations. Fungicide sprayed and uninoculated plots served as controls. Significant differences in disease severity were found among the treatments. Control plots had the lowest area under the disease progress curve (AUDPC) but were significantly different from each other and all 12 isolates. Significant differences in AUDPC were noted for some isolates. Although the uninoculated plot remained relatively disease-free during the early growing season, it had the highest apparent infection rate and differed significantly from 11 of the isolates. There were no differences in apparent infection rates among the isolates. At the R6-R7 growth stage, several of the isolates differed in vertical disease progress and defoliation. There were no differences in yield, but differences in 300 seed weight occurred between 2 isolates.

THE RELATION OF VERTICILLIUM DAHLIAE STRAINS AND COTTON PLANTINGS TO THE EPIDEMIC OF WILT DISEASE IN PISTACHIO NUT TREES. W. C. Schnathorst, USDA, SE, ARS, University of California, Davis, CA 95616.

Almost 40,000 acres (16,200 hectares) of pistachio nut trees (*Pistache vera*) are planted in California. Over 600,000 trees have been lost to verticillium wilt. The incidence of wilt caused by *Verticillium dahliae* in trees planted in virgin soil is usually low (0-3%). However, if cotton is planted several years before pistachio trees are planted, disease incidence can be 40% or more. Isolations were made from diseased trees sampled at random in several orchards. The *V. dahliae* isolates obtained were identified to strain using several laboratory and greenhouse procedures. The mild SS-4 and severe T-1 cotton strains of *V. dahliae* were equally involved in the epidemic of wilt in pistachio. Greenhouse inoculations verified the virulence of SS-4 and T-1 in several *Pistache* spp. These results substantiate the role of cotton plantings and cotton strains of *V. dahliae* in wilt epidemics in pistachio nut trees.

RELATIVE IMPORTANCE OF SOURCES OF VERTICILLIUM WILT INFESTATION IN ALFALFA. A. A. Christen and R. N. Peadar, Wash. State Univ. and USDA, Prosser, WA 99350

Alfalfa field plots in a series of experiments were infested with *Verticillium albo-atrum* by 1) injecting conidia in sprinkler-applied irrigation water; 2) disseminating conidia within the canopy; 3) incorporating infested hay in the soil before planting; 4) mowing the hay with an artificially contaminated cutter bar; 5) planting seed which were lightly coated with dark mycelia and conidia; and 6) scattering infested plant material over the plot immediately after a cutting. Mean differences between treated and check plots in the number of plants per 4.1 m² plot showing disease symptoms were 0.25, 0.25, 6, and 10 for the first four methods, respectively by the third harvest; 86 for treatment No. 5 by the fourth harvest; and 66 for No. 6 within 35 days after treatment. Yields were significantly ($P=0.05$) reduced in methods five and six by the fourth harvest but not in the other methods.

APPLE POWDERY MILDEW DISEASE PROGRESS ON INTERVALS OF SHOOT GROWTH: AN ANALYSIS OF LEAF MATURATION AND FUNGICIDAL EFFECTS. Lalancette, N. and K.D. Hickey, The Pennsylvania State University Fruit Research Lab, Biglerville, Pennsylvania 17307

The independent and combined effects of leaf maturation and fungicidal control were examined by monitoring disease progress on fixed intervals of shoot growth. Six Rome Beauty trees were sprayed to run-off at two week intervals with the fungicide bitertanol at 150 mg a.i./liter; another six unsprayed trees were used as controls. On each tree ten vegetative terminals were randomly chosen for observation. During each of five disease assessments, the youngest, most completely unfolded leaf was tagged; thus, by the end of the season, four intervals of growth were demarcated. Analysis of disease incidence on each interval revealed an epidemic pattern that was separated into a disease increase phase followed by a disease decrease phase on both sprayed and unsprayed trees. Although bitertanol significantly reduced disease throughout the epidemic, further reduction in disease incidence during the decrease phase was attributed to leaf maturation.

THE EFFECT AND SIGNIFICANCE OF 'LATE SEASON' APPLE POWDERY MILDEW. L. P. Berkett and K. D. Hickey, Department of Plant Pathology, The Pennsylvania State University Fruit Research Laboratory, Biglerville, PA 17307

The number of 'late season infection sites' was determined for 'Rome Beauty' apple trees in September 1981. 'Late season infection sites' were considered to be terminals whose buds had opened in the latter part of the growing season producing a new flush of leaves which became infected with *Podosphaera leucotricha*. The following spring data were collected on the growth and development of these terminals. 'Late season infection sites' had a significantly lower survival rate than other terminals. When they did survive, a greater proportion produced vegetative buds than fruit buds. 'Late season sites' were similar in appearance to primary infection sites and, whereas primary infection sites served as an important source of inoculum for secondary spread, 'late season infection sites' possibly served as a source of inoculum for infection of overwintering sites.

ASCOSPORE RELEASE PATTERN OF *GNOMONIA NERVISEDA*, INCITANT OF PECAN VEIN SPOT. R. S. Sanderlin, Louisiana State Univ. Pecan Research and Extension Station, Shreveport, LA 71105.

Over a 3 yr period (1978-'80), vein spot was the most frequently observed foliar disease on several pecan cultivars in North Louisiana. Infected leaves usually abscised in late summer or early fall. Infection was primarily by ascospores produced in leaf debris. Release of *G. nerviseda* ascospores was monitored with a Burkard spore trap during the 1980-81 growing season. Ascospores were released following rainfall. Spore discharge could be induced by as little as 0.25 cm of rain, although spores were not caught after every rainfall. Spores were caught in every month from April through August. The largest and most frequent catches were in May and June of both years. Number of days of spore catches were 12 and 5 in May and 10 and 4 in June, 1980 and 1981, respectively. The average number of spores trapped per 14.3 m² of air/day in discharge periods was 12 and 18 X 10³ in May and June, 1980, respectively. In 1981 the average was 9 and 3 X 10³ for May and June, respectively, although the trap was off for most of June.

EVALUATION OF WIND PENETRATION EFFECTS ON GRAPEVINE CANOPY MICROCLIMATE WITH REFERENCE TO BOTRYTIS BUNCH ROT. S. D. Savage and M. A. Sall, University of California, Davis.

The restriction of air movement by plant canopies is an important aspect of the microclimate in which disease develops under marginal conditions; however, the direct characterization of this parameter is difficult. A mode of analysis is presented which allows inferences concerning the differences in wind penetration of two canopy types which exhibit differences in infection by *Botrytis cinerea* Pers. An analysis of the diurnal pattern in the difference between fruit cluster temperatures and of the rate of change of temperature with respect to wind speed at 3 meters suggests that one canopy type (two wire trellis) is more easily penetrated by wind than the other (cross arm trellis). The cross arm trellis is associated with greater disease development. The implications of the wind penetration difference for development of the pathogen is discussed.

INTERACTION OF BARLEY STRIPE MOSAIC VIRUS AND SPOT BLOTCH ON DICKSON BARLEY. F. W. Nutter, Jr., V. D. Pederson, and R. G. Timian. Dept. of Plant Pathology, and USDA-ARS, North Dakota State Univ. Fargo, ND 58105

Field plots were established to quantify the effects of barley stripe mosaic virus (BSMV) and spot blotch (*Cochliobolus sativus*) on yield components of 'Dickson' barley. Seed infected with BSMV was blended with healthy seed to produce several levels of seed infection. Seed infection levels constituted whole plots of a factorial experiment planted in a randomized complete block design. Subplots were sprayed either with a spore suspension of *C. sativus*, sprayed with fungicide, or untreated. BSMV had the greater effect on reducing yield/plot, 500-kernel weight, test weight and % plump kernels. Spot blotch had less effect on yield components as the proportion of BSMV-infected plants per plot increased. There was no difference in the sporulation or disease efficiency of *C. sativus* on healthy vs. BSMV-infected plants; however, the latent period was longer and lesion size was smaller on BSMV-infected plants. Spot blotch and BSMV are less than additive in their effect on yield components. Analysis of resistance components may explain their non-additivity.

WHEAT STREAK MOSAIC OF WHEAT IN MISSOURI-1981. Einar W. Palm, Department of Plant Pathology, University of Missouri, Columbia, MO 65211.

Wheat streak mosaic (WSM) made a dramatic appearance in Mo. in the spring of 1981, with infected wheat fields in 40 counties, both north and south of I-70. The only area relatively untouched was in the Delta counties of southeast Mo. Field responses varied from traces to complete losses. It is estimated that Missouri farmers lost \$30 million to the disease. Although the wheat curl mite vector (*Aceria tulipae*) has been present in Missouri in the past, and the virus has been transmitted to corn and wheat incidentally, there was never a serious epidemic until 1981. It is believed that an unusual combination of environmental conditions in 1980 were responsible for the outbreak: 1) Dry weather and prevailing winds aided massive wind-borne movement of mites into Missouri. 2) Large acreages of early planted wheat. 3) Considerable volunteer wheat in or near wheat fields. 4) Extended fall of ideal weather conditions for disease development.

EPIDEMIOLOGIC SURVIVAL OF *PSEUDOMONAS SYRINGAE* PV. *TOMATO* ON TOMATO TRANSPLANTS SHIPPED FROM GEORGIA, U.S.A. TO ONTARIO, CANADA. W. G. Bonn and R. D. Gitaitis, Agriculture Canada, Harrow, Ontario NOR 1G0, and Dept. of Plant Path., University of Georgia, Tifton, GA 31793.

The survival of *Pseudomonas syringae* pv. *tomato* on tomato transplants shipped from Georgia to Ontario was studied using an antibiotic-resistant strain (G13) of the pathogen. New Yorker tomato transplants were inoculated with 3.3×10^8 cfu/ml of G13 and assayed 1 hr post-inoculation for the presence of the organism by leaf washings which revealed a population of 7.5×10^2 cfu/leaf. Following incubation at 12 C for 18 hr and shipment to Ontario (10 hr) the transplants were again assayed and 1.3×10^6 cfu/leaf were detected on the leaves. Plants were then stored at 10 C for 60 hr, assayed for the presence of G13 and then transplanted into the field. The second leaf assay showed a population of 3.6×10^7 cfu/leaf. Bacterial speck symptoms developed on the tomato plants 4 days after transplanting into the field. It is concluded that *P. syringae* pv. *tomato* can survive as an epiphyte on tomato transplants to cause disease in the field.

ETIOLOGY AND EPIDEMIOLOGY OF BACTERIAL LEAF SPOT AND STEM BLIGHT OF SAFFLOWER IN MONTANA. D.L. Jacobs, J.W. Bergman and D.C. Sands, Dept. of Plant Pathology, Montana State University, Bozeman, MT 59717.

During the 1978 growing season, a severe leaf necrosis and stem blight of safflower (*Carthamus tinctorius*) occurred. Consistent isolation of fluorescent pseudomonads from infected tissue, pathogenicity tests and identification of the bacterium determined that *Pseudomonas syringae* was the incitant of bacterial leaf spot and stem blight of safflower. *P. syringae* was isolated from 7.3% and 3.6% of Montana produced seed of the varieties S-208 and Sidwill, respectively. *P. syringae* was recovered from the aerial portions of 2.6% of S-208 seedlings grown from non-surface sterilized seed. During the 1978 and 1979 growing season, weeds, plant debris and soil yielded isolates of *P. syringae*. Results from seed transmission studies indicated that seeds, weeds, plant debris and soil are possible sources of primary inoculum.

DISEASE PROGRESS OF PEA ROOT ROT AT VARIOUS *APHANOMYCES* INOCULUM LEVELS. W. F. Pfender and D. J. Hagedorn, Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Epidemics of pea root rot incited by *Aphanomyces euteiches* were studied during three seasons in field plots on sandy soil. Disease progress data, obtained by plating sampled roots on agar at weekly intervals to determine % plants infected by *Aphanomyces*, were compared for different soil inoculum levels of the pathogen. In one year, rate of disease increase (slope of the regression line for disease incidence vs time) increased with increasing inoculum level (.01, .1, .5, or 4 prop/g). In the other 2 years, with inoculum levels of .02 and .2 prop/g, slopes were not affected by inoculum level. There was a correlation between inoculum level and number of days from planting to 50% disease incidence in all 3 years. Midway through the epidemic, the % plants infected was higher in plots receiving more frequent irrigation. In experiments conducted on noninfested soil, infected plants were transplanted into a stand of healthy peas. The pathogen spread from infected to healthy plants, demonstrating that this is not a monocyclic disease.

CONIDIAL SURVIVAL OF *SIROCOCCUS CLAVIGINENTI-JUGLANDACEARUM* IN A SIMULATED AIRBORNE STATE. N. Tisserat and J. E. Kuntz. Dept. Plant Pathology, University of Wisconsin, Madison, WI 53706.

Conidia of *Sirococcus claviginenti-juglandacearum*, the causal agent of butternut canker, are liberated from pycnidia by rain-splash impactation. Survival of the hyaline conidia ($9-17 \times 1-2 \mu\text{m}$) in a simulated airborne state was studied by impacting spores on fine spider threads ($<1.5 \mu\text{m}$ diam.) wound around rectangular frames. Frames were exposed to ambient conditions for periods up to 32 hours. Spore survival was measured as a percentage of germination on 2.0 % water agar after 48 hours incubation at 25 C. Survival of conidia after 32 hours in still air at 35 % RH was greater at 13 C (82 %) than at 25 C (18 %). At both temperatures, viability was greater at 35 % RH than at >95 % RH. In the forest, spore survival varied greatly with weather conditions. Highest survival (35 % after 8 hours) was found on a cool, humid day with overcast skies. Survival of rain-splashed conidia disseminated in aerosols allows for long distance dispersal of the pathogen.

SOYBEAN YIELD IN IPM SYSTEMS INOCULATED WITH *CERCOSPORA KIKUCHII*. T.S. Abney, Dept. Botany & Plant Pathology, Purdue University and USDA, ARS, West Lafayette, IN 47907.

Wells II soybeans in Integrated-Pest-Management systems inoculated with conidial suspensions of *Cercospora kikuchii* developed foliar blight in all treatments (soybean-soybean, S-S, and corn-soybean, C-S, rotations; conventional, chisel and minimum tillage systems; and minimum, moderate and maximum weed management treatments). *Cercospora* seed infection due to inoculation averaged 45% and 58% in the S-S and C-S rotations, respectively; 62%, 57% and 42% for the conventional, chisel and minimum tillage systems, respectively; and 57%, 53% and 50% for the minimum, moderate and maximum weed management levels. Foliar blight and seed infection occurred in less than 5% of non-inoculated soybeans. Yield reduction due to inoculation averaged 20% in the S-S and C-S rotations. Foliar blight reduced yields more with conventional and chisel tillage (23%) than with minimum tillage (12%). Yield reduction in minimum weed management treatments (24%) was higher than in moderate or maximum management treatments (17%).

INACCURACY IN VISUAL ASSESSMENT OF ORCHARDGRASS PURPLE LEAFSPOT AREA. R.T. Sherwood, K.E. Zeiders, and C.C. Berg. U.S. Regional Pasture Research Laboratory, USDA-ARS, University Park, PA 16802.

Two groups of five experienced scorers used published area reference diagrams to aid in scoring *Dactylis glomerata* leaves for percentage of leaf area infected by *Stagonospora arenaria*. There were significant differences among groups and scorers within groups, and a significant leaf x scorer interaction. The number of spots per leaf significantly biased estimates by five scorers. All but one scorer usually overestimated the true area of spotting. The overestimation was greatest when the total spotted area was least. Regression analysis showed that overestimation was inversely proportional to log area and directly proportional to the number of spots. Use of these visual scores in the Vanderplank equation for predicting disease increase would underestimate true rate of disease increase.

Role of within-field inoculum sources in forecasting gray mold of snap beans. K.B. Johnson and M.L. Powelson, Dept. of Botany & Plant Pathology, Oregon State University, Corvallis, OR 97331.

Observations in five commercial snap bean fields in Oregon's Willamette Valley revealed that senescing cotyledons were the initial plant tissue colonized by *Botrytis cinerea*. Young stem tissues also became diseased and served as the most durable within-field inoculum source. The number of these inoculum sources before bloom varied between fields and ranged from 14.8 to 0.2 per 5 m row. The number of spores per plant at bloom initiation and the incidence of *B. cinerea* on blossoms at full bloom was positively correlated with the number of prebloom inoculum sources within a field. Using the number of inoculum sources prior to bloom as a predictive variable, 50% of the variation in the incidence of pod rot between fields could be accounted for. A multiple regression model, which included the number of inoculum sources before bloom, frequency of irrigation, duration of leaf wetness due to irrigation and rain, and canopy size accounted for 82% of the variation in percent pod rot between fields.

DEVELOPMENT AND SPREAD OF LATE BLIGHT, *PHYTOPHTHORA INFESTANS* (MONT.) DEBARY IN SOLE AND INTERCROP POTATO PLOTS. S. A. Raymundo and J. Alcazar, International Potato Center, Apartado 5969, Lima, PERU

In attempts to evolve sound and viable control methods against late blight in the mid elevation, humid tropics, various approaches including intercropping were tried. Results showed that potato plants in potato-sorghum, potato-peanut and potato-cowpea intercrop plots had remarkably less blight infection when compared with the infection on sole potato plots. Potato plants in the potato-sorghum plots were clearly the best developed and most vigorous. Conversely, potato plants in the potato-tomato and potato-sweet potato plots were severely damaged by the disease. Possible explanation on the differences in infection spread in the various crop association are presented. Implications of the findings on the choice of what crops to interplant with potato in order to reduce late blight damage are discussed.

QUANTIFICATION OF ADAPTATION OF *PHYTOPHTHORA INFESTANS* POPULATIONS TO DIFFERENT POTATO CULTIVARS. R. V. James and W. E. Fry, Cornell University, Ithaca, NY, 14853.

We assessed potential adaptation of natural and mutagenized populations of *Phytophthora infestans* to potato cultivars differing in rate-reducing resistance. Infection efficiency, sporulation, and rate of epidemic development in field plots, were used to measure adaptation. Subpopulations were derived from each initial population of *P. infestans* by repeated culture on two cultivars. Changes in subpopulations were not differential for increased ability to cause disease on the cultivar on which the subpopulation was derived, suggesting no adaptation had occurred. In the field, disease progress was assessed on two cultivars in separate or mixed plots. The lack of differences in rate of disease development indicated there was no adaptation. We conclude that rapid adaptation of *P. infestans* populations is unlikely to endanger the use of cultivars with rate-reducing resistance.

POTATO LATE BLIGHT - EPIDEMIOLOGY: AN INSTRUCTIONAL FILM. R. V. James and W. E. Fry, Cornell University, Ithaca, NY 14853.

This film was made to illustrate the epidemiology of *Phytophthora infestans* on potatoes and to show the impact of plant disease at the population level. Views of symptoms on individual plants introduce the disease. Observations of field plots at three-day intervals illustrate the dynamics of disease development in populations. Adjacent plots of a susceptible cultivar and one with rate-reducing resistance demonstrate the influence of resistance on rate and extent of disease development. Disease progress curves superimposed over views of the plots "grow" with time, allowing disease development to be assessed visually and graphically. The need for knowledge and understanding of epidemiology to formulate logical disease control programs is emphasized. Current control practices in commercial potato production are illustrated.

DETECTION OF POTATO SPINDLE TUBER VIROID IN TRUE POTATO SEED. M. E. Grasmick and S. A. Slack, Dept. of Plant Pathology, Univ.

of Wisconsin-Madison, Madison, WI 53706

Since potato spindle tuber viroid (PSTV) is transmitted through true potato seed (TPS), information on detection in TPS is of interest to breeding programs and germ plasm collections. PSTV-infected TPS were obtained from controlled crosses utilizing infected parental stock. Progeny were indexed by bioassay on 'Rutgers' tomato (*Lycopersicon esculentum* Mill.) or by polyacrylamide gel electrophoresis (PAGE). PSTV could not be detected in ungerminated TPS by either indexing procedure. Bioassay plants inoculated with expressed sap from composite samples of 2-week-old TPS seedlings only developed symptoms at high but not low serial sap dilutions. PAGE of 2-week-old composite TPS samples but not single seedling samples detected PSTV. PSTV detection in single TPS apices by bioassay was best before the first true leaves were fully expanded and decreased with plant age. Data suggest that bioassay is more sensitive than PAGE and that assays should be of 2-week-old TPS seedlings.

AN UNIQUE INTERACTION OF SPINDLE TUBER VIROID AND VIRUS Y IN POTATOES. R.P. Singh, Agriculture Canada, Research Station, Fredericton, N.B., Canada E3B 4Z7

A severe necrotic disease was observed on 'Kennebec' potato plants in an experimental field plot. Symptoms consisted of necrotic rings and spots on foliage near the top of the plant, severe drop of the lower leaves, and severe stunting of the entire plant. Identification of causal agents showed the presence of both potato spindle tuber viroid (PSTV) and potato virus Y (PVY), in infected plants. Virus and viroid-free plants of 'Kennebec' potatoes were mechanically inoculated with PSTV or PVY alone or in various combinations. The necrotic symptoms were only reproduced when the potato plants were infected with PSTV prior to the infection of PVY. No necrosis was observed either with simultaneous inoculation with PSTV + PVY or PVY prior to PSTV infection. Thirteen potato cultivars with different types of PVY susceptibility were tested for their necrotic reaction to PSTV - PVY combinations. The cultivars Keswick, Sebago and Saco reacted similarly to Kennebec, while others did not show any increased symptoms.

INTERFERENCE BETWEEN POTATO VIRUS Y AND PEPPER MOTTLE VIRUS. Matthew D. Alegbejo and Merritt R. Nelson, Department of Plant Pathology, University of Arizona, Tucson, Arizona 85721.

Potato virus Y (PVY) and pepper mottle virus (PeMV) are potyviruses unrelated serologically. Potato virus Y induces local lesions followed by systemic necrosis and death of a hypersensitive selection from Anaheim chili (Special Pepper - SP). The pattern of symptom development of PeMV on tabasco is near identical to PVY on SP. Potato virus Y and PeMV are systemic to tabasco and SP, respectively. When SP and tabasco were inoculated respectively with PeMV and PVY, and challenged with the reciprocal virus at one day intervals, local lesions were produced up to 6 (PVY) and 9 (PeMV) days. When inoculum from Anaheim chili infected with both viruses was used to inoculate SP and tabasco, local lesions were produced only on tabasco. Neutralization of infectivity with antiserum to PeMV resulted in no lesions on either host. Local lesions were produced on both SP and tabasco when the mixed inoculum was prepared from separately infected Anaheim chili.

TRANSLOCATION OF POTATO VIRUS S IN INOCULATED POTATO PLANTS IN THE FIELD AND GREENHOUSE. G.D. Franc and E.E. Bantari, Research Associate, Department of Botany and Plant Pathology, Colorado State University, Ft. Collins, CO 80521; Professor, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Potato cultivars Norland, Kennebec and Russet Burbank were mechanically inoculated with a Minnesota isolate of potato virus S (PVS). The amount of tuber infection in field inoculated plants as measured by enzyme-linked immunosorbent assay (ELISA), indicated that Russet Burbank was significantly more susceptible than Norland or Kennebec and Kennebec was significantly more resistant than Norland or Russet Burbank ($P=0.01$). In plants inoculated in the greenhouse PVS spread from inoculated middle-leaves within a 24-hr post-inoculation period and detectable amounts of PVS occurred in upper and lower leaves at 13 and 20 days, respectively. Translocation of PVS from an inoculated leaf downward in the stem below the soil line occurred within 13 days for Russet Burbank and Norland and within 20 days for Kennebec. Mature plant resistance to this PVS isolate was not observed in the field.

IDENTIFICATION OF VECTOR-SPECIFIC BARLEY YELLOW DWARF VIRUS (BYDV) ISOLATES AND THEIR APHID VECTORS IN CALIFORNIA. F. E. Gildow and W. F. Rochow. Plant Pathology Dept., Univ. of California, Berkeley, CA 94720 and Plant Pathology Dept., Cornell University, Ithaca, N. Y. 14853.

Cereal grains collected from eight counties in California were tested by aphid transmission and enzyme-immunosorbent assay for luteoviruses that cause barley yellow dwarf. California isolates were compared to the previously characterized MAV, PAV and RPV isolates of BYDV. Of 128 plants sampled, 75% were infected by luteoviruses similar to PAV, 19% by viruses similar to MAV and 6% by RPV-like isolates. No difference in vector competence occurred among California and New York clones of *Rhopalosiphum padi*, which transmitted PAV- and RPV-like isolates. California clones of *Metopolophium dirhodum* transmitted MAV and PAV, but not a California isolate similar to PAV (CA-PAV). California and New York clones of *Sitobion avenae* transmitted MAV and PAV. Several varieties of oats and barley differed in tolerance to infection by three California luteovirus isolates.

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BARLEY YELLOW DWARF VIRUS IN EASTERN WASHINGTON: THE SUSCEPTIBILITY OF CORN TO DIFFERENT ISOLATES. D.W. Hazelwood and S.D. Wyatt. Dept. of Plant Pathology, Washington State University, Pullman, WA 99164-6430.

Corn in the Columbia Basin of Eastern Washington is an oversummering host of barley yellow dwarf virus (BYDV). In Okanogan County, which is a non-corn growing region geographically excluded from aphid migrations from the Columbia Basin, endemic BYDV isolates rarely contact corn. The susceptibility of corn to isolates from Columbia Basin and isolates from Okanogan County were compared. *Rhopalosiphum padi* from both regions and non-viruliferous greenhouse-reared *R. padi* were used to transfer BYDV to test hosts in the greenhouse. All isolates infected *Hordeum vulgare* cv. 'Luther' and *Triticum aestivum* cv. 'Daws'. The Columbia Basin isolates readily infected *Zea mays* cv. 'Rainbow' while those from Okanogan County were rarely detected on test corn assayed by symptoms on 'Luther' barley or 'Rainbow' corn, or by ELISA using PAV-specific antiserum. Aphid transmission tests indicate isolates from both regions are aphid-nonspecific PAV types. Although corn is a host for both BYDV and *R. padi*, not all isolates tested are adapted to corn.

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APHID AND WHITEFLY TRANSMITTED CUCURBIT VIRUSES IN IMPERIAL COUNTY, CALIFORNIA. J. A. Dodds, S. T. Nameth, J. G. Lee, and F. F. Laemmlein. University of California, Riverside, CA 92521 and *Cooperative Agricultural Extension, University of California, El Centro, CA 92243.

Incidence of mosaic approached 100% in some cantaloupe fields by June 1981. Watermelon mosaic virus-1 (WMV-1), WMV-2, squash mosaic virus (SqMV) and cucumber mosaic virus (CMV) were surveyed by ELISA. The most common virus detected was aphid transmitted WMV-2 (95% of samples with symptoms, 0-72% of samples collected at random). Two fields had a low incidence of SqMV. WMV-1 and CMV were not detected. Virus diseases associated with whiteflies (*Bemisia tabaci*) caused severe losses and reached epidemic levels for the first time in several Imperial Valley crops in late summer to winter, 1981. Geminiviruses typical of whitefly-transmitted geminiviruses have been detected by electron microscopy in leaf dips and partially purified virus preparations from squash, watermelon and cantaloupe.

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LETTUCE INFECTIOUS YELLOW -- A NEW WHITEFLY TRANSMITTED VIRUS OF THE DESERT SOUTHWEST. James E. Duffus, Dennis E. Mayhew and Robert A. Flock, USDA-ARS, U.S. Agricultural Research Station, Salinas, CA 93915, California Department of Food and Agriculture, Sacramento, CA 95814, Imperial County Agricultural Commissioner, El Centro, CA 92243.

A new infectious yellowing disease of lettuce, sugarbeet, carrot and other crop and weed hosts has been found in the desert areas of Southwestern U.S.A. The causal virus, lettuce infectious yellow [LIYV] is transmitted by the whitefly *Bemisia tabaci*. Losses of from 50 to 75% in lettuce production occurred in desert areas during the 1981-1982 growing season. The virus particles, as visualized from leaf dips of field plants, whitefly induced infections in the greenhouse, and partially purified preparations, are rod-shaped (11 x 1000-2000 nm). The host range, particle size and properties of LIYV appear to be distinct from previously described whitefly transmitted viruses.

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BEETLE REGURGITANT AS A DETERMINANT OF SPECIFICITY OF PLANT VIRUS TRANSMISSION. R.C. Gergerich, H.A. Scott, and J.P. Fulton. Dept. of Plant Pathology, PS 217, University of Arkansas, Fayetteville, AR 72701.

Regurgitant from leaf-feeding beetles (*Cerotoma trifurcata*, *Epilachna varivestis*, and *Diabrotica undecimpunctata*) contains a factor which selectively inhibits infection by viruses not transmitted by beetles, but which does not affect infection by beetle-transmissible viruses. When beetle feeding was simulated by gross wounding of leaf tissue and purified virus was applied during wounding, high levels of transmission of both beetle-transmissible and non-beetle transmissible viruses were recorded. When inoculum was mixed with beetle regurgitant, however, there was a high level of transmission of beetle-transmissible viruses and a very low level of transmission of non-beetle transmissible viruses. The regurgitant factor is heat labile, stable to freezing, and has a molecular weight greater than 8000 daltons. The regurgitant factor does not irreversibly inactivate non-beetle transmissible viruses.

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ATTEMPTS TO TRANSMIT WHEAT STREAK MOSAIC VIRUS USING BANKS GRASS MITE. J. P. Hill and B. D. Congdon. Dept. of Botany & Plant Pathol. and Dept. of Zool. and Entomol., Colorado State University, Fort Collins, Colorado 80523.

Wheat Streak Mosaic Virus (WSMV) causes significant localized losses of winter wheat in Colorado and also occurs in field corn. Severely WSMV infected wheat fields are often located adjacent to sprinkler irrigated corn which can serve as the WSMV inoculum source. The Banks grass mite, *Oligonychus pratensis*, parasitizes both corn and wheat. The similarities in geographical distribution and host range of *O. pratensis* and WSMV suggest that this mite, if capable of transmission, could be an important WSMV vector. The grass mite is wind dispersed and an active crawler, especially when the food source is depleted, which would facilitate movement between corn and wheat. Greenhouse studies were conducted to determine if the Banks grass mite could transmit WSMV from wheat to wheat, wheat to corn, corn to wheat, and corn to corn. WSMV was mechanically transmitted in all host combinations, but no grass mite virus transmission was observed.

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ISOLATION, IDENTIFICATION, AND WHEAT VARIETY INTERACTIONS OF WHEAT STREAK MOSAIC VIRUS IN COLORADO. J. Shahwan and J. P. Hill. Department of Botany & Plant Pathol., Colorado State University, Fort Collins, Colorado 80523.

Wheat Streak Mosaic Virus (WSMV) was isolated from six commercial wheat fields in Eastern Colorado. Positive identification of the WSMV isolates was made on the basis of disease symptoms, host range, electron microscopy, and leaf dip serology. The effects of a Colorado and Nebraska WSMV isolate were compared on nine commonly grown winter wheat cultivars in Colorado and four spring wheat varieties. All mechanically inoculated plants developed typical WSMV symptoms and had significantly reduced plant height, fertile tillers, and grain yield. The number of tillers and 100 seed weight was not consistently significantly reduced. Yield reduction ranged from 87% in Wichita to 28% in Super "X". No significant differences in these traits could be attributed solely to isolate differences. The most yield loss occurred in the cultivars Wichita, Lugimi, Centurk, Baca, and Duke while the least occurred in Super "X", Arz, Vona, and Eagle.

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NUMBER OF GENES FOR RESISTANCE TO MAIZE DWARF MOSAIC VIRUS IN 4 MAIZE INBREDS. Eugen Rosenkranz and Gene E. Scott. USDA, ARS, Dept. Plant Path. & Weed Sci. and Dept. Agronomy, Miss. State Univ., Mississippi State, MS 39762.

Using a new method, we determined the number of genes conditioning resistance to maize dwarf mosaic virus strain A (MDMV-A) in inbreds Ark361, Mp71:222, Pa405, and T232. The method utilizes 2 classes of plants, diseased and symptomless, in 3 segregating generations- (resistant-Rx susceptible-S)F₂, (RxS)xS, and (RxS)xR - to determine the number of alleles for resistance that allow symptom expression at various times after inoculation. The observed ratios of diseased to symptomless plants are then compared to the expected ratios by calculating chi-square values for goodness-of-fit. Data from 15 and 19 days postinoculation showed 2 resistant genes in Ark 261, data from 9 and 17 days postinoculation showed 3 resistant genes in Pa405, and data from 5 and 8 days postinoculation showed 2 and 3 resistant genes in Mp71:222 and T232, respectively. Previously, we found agreement in results between this method and the chromosomal translocation technique for determining the number of MDMV-A-resistant genes.

RELATIONSHIP OF SYMPTOMLESS INFECTION TO MAIZE WHITE LINE MOSAIC INCIDENCE. Raymond Louie, D. T. Gordon, L. V. Madden, and J. K. Knoke. USDA-SEA-ARS, Dept. of Plant Pathology, Ohio Agricultural Research and Development Center, Wooster, OH 44691 and The Ohio State University, Columbus, OH 43210.

In Ohio, symptomless infection of sweet corn (*Zea mays* L.) by maize white line mosaic virus (MWLMV) was detected by enzyme-linked immunosorbent assays (EIA) in 13/13, 11/13, and 7/13 consecutive plants from three fields with a known history of MWLM. In addition, MWLMV was detected by EIA in 2/2, 2/2, and 2/2 plants with MWLM symptoms in these three fields and in 4/15, 1/15, and 0/15 plants from three fields where MWLM had not been previously found. In test plots where Seneca Chief sweet corn was planted at four different dates, all correlations between incidence of MWLM by symptoms, EIA of roots, and EIA of leaves were nonsignificant ($P > 0.05$). Frequency of symptomless infection in corn was higher in 1981 than in 1980. Data indicated MWLM leaf symptoms alone underestimated disease incidence.

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A NEW CUCUMBER SOIL-BORNE VIRUS COMPARED WITH TOMBUS-DIANTHO- AND OTHER SIMILAR VIRUSES. R. Koenig, D.E. Lesemann, V. Huth, and K.M. Makkouk. Institut Fur Viruskrankheiten der Pflanzen, BBA, Braunschweig, Federal Republic of Germany and Lebanese National Council for Scientific Research.

A virus isolated from cucumber roots grown south of Beirut produced local lesions on a number of plant species, but no systemic infection. Particles c.31 nm in diameter had a sedimentation coefficient of 120 S. The virus migrated towards the anode at pH 7. The A280:A260 value of 0.66 indicated 14% RNA. Coat protein and major RNA species were 4.14×10^4 d and 1.5×10^6 d, respectively. A minor RNA species was 0.16 - 0.19 $\times 10^6$. The base composition of the RNA was C22, A23, G32, U23. Virus aggregates were scattered or in small aggregates in the cytoplasm only. The virus did not induce cytopathic effects typical of tombusviruses or dianthoviruses, nor did it react with antisera to known tombusviruses, dianthoviruses or 39 other isometric viruses. The name cucumber soil-borne virus is proposed for this apparently new virus.

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DIRECT DEMONSTRATION OF LIGHT MODIFICATION OF TOBACCO MOSAIC VIRUS CONCENTRATION IN TOBACCO. G. B. Olson, Department of Plant Pathology, University of California, Riverside, CA 92521.

Lower leaves of tobacco (*Nicotiana tabacum* L. cv Xanthi) were inoculated with Tobacco Mosaic Virus, strain U-1, and placed in a differential temperature inoculation system to achieve synchronous virus replication (Dawson, W. O. and Schlegel, D. E., 1973. Virology 53: 476-478). After 10-16 days, 11 mm discs were cut from newly expanding leaves, floated upside down on DD H₂O, and placed under different fluorescent-incandescent light conditions with varying red/far red ratios. After four days, the discs were frozen at -15°C. Quantitation of TMV concentration was measured by rocket immunoelectrophoresis at 7°C using 0.08 M Tris-HCl, 0.024 M Tricine, 0.3mM Magnesium Acetate, buffer pH 8.6 (TTM Buffer). Ten leaf discs (Avg. wt. 0.36 gm) were ground in liquid nitrogen, resuspended in 1 ml TTM buffer, centrifuged at 4300 X G for 10 min, and the supernatant used directly for quantitation. Results indicate distinct differences in virus concentration under different red/far red ratios.

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PERINUCLEAR ASSEMBLY OF AMERICAN WHEAT STRIATE MOSAIC VIRUS IN ZEA MAYS. Gardner, Wayne S. Dept. of Plant Science, SD State Univ., Brookings, SD 57007.

Bacilliform and bullet-shaped rhabdovirus particles of American wheat striate mosaic virus (WSMV) accumulated in or near perinuclear spaces of leaf bundle-sheath and parenchyma cells of July 3, 1979-planted and naturally-infected N28 inbred corn. Although individual and clusters of virions appeared to be free in cytoplasmic and nuclear inclusions, the predominant appearance of virions was in single-membrane bound clumps that accumulated in cytoplasm or nuclei and were suspended in material that resembled nucleoplasm. Nuclear heterochromatin was scarce or absent in cells containing virions regardless of whether the virions accumulated in cytoplasm, nuclei or both. It is concluded that WSMV was assembled in corn leaf tissue between the lamellae of the nuclear envelope and either moved into the cytoplasm in clumps surrounded by the outer lamella of the nuclear envelope, or moved into the nucleus in clumps surrounded by the inner lamella of the nuclear envelope. The large accumulation of virus caused displacement of normal components of nuclei and cytoplasm.

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FLUORESCENCE MICROSCOPY OF LEAF TISSUE AND ISOLATED CELLS FROM BEANS INFECTED BY BEAN GOLDEN MOSAIC VIRUS. Narceo B. Bajet and Robert M. Goodman, Dept. of Plant Pathology, University of Illinois, Urbana, IL 61801.

Leaf sections and enzymatically-isolated cells from trifoliolate leaves of Top Crop beans at 6 and 20 days after sap inoculation (DAI) with bean golden mosaic virus (BGMV) were fixed and examined after indirect fluorescent antibody staining. Leaf sections showed fluorescence in both the mesophyll cells and the sieve elements and associated parenchyma. Eight to 12% of the cells isolated from leaves at 6 DAI showed bright fluorescence confined to the nuclei. At 20 DAI both generalized cytoplasmic fluorescence (22% of the cells) and fluorescence only from the nuclei (8-10% of the cells) were observed. No fluorescence was seen in healthy tissues treated with IgG from preimmune serum or IgG against BGMV or in sections and cells from infected leaves treated with IgG from preimmune serum. These results confirm that in acute stage infections viral antigen is concentrated in the nuclei of phloem cells. In later stages of infection, viral antigen is detected in mesophyll cells and is not confined to the nucleus.

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BEAN GOLDEN MOSAIC VIRUS ANTIGEN AND VIRAL SPECIFIC DNA SEQUENCES IN PHASEOLUS VULGARIS DETERMINED BY ELISA AND DNA-DNA HYBRIDIZATION. K.M. Franklin, Dept. of Biological Sciences, University of Alabama in Huntsville, Huntsville, AL 35807 and Robert M. Goodman, Dept. of Plant Pathology, University of Illinois, Urbana, IL 61801

Uppermost expanded trifoliolate leaves of *Phaseolus vulgaris* plants infected with bean golden mosaic virus (BGMV) were harvested at 8, 10-20, and 24 days after inoculation with the virus. Enzyme linked immunosorbent assay (ELISA) and DNA-DNA hybridization on nitrocellulose were used to detect viral antigen and DNA sequences in the tissue from each day of harvest. Antigen concentration was at its peak in leaves harvested 14 days after BGMV inoculation, consistent with results of previous experiments in which virus titer was measured by purification. DNA-DNA hybridization, however, gave consistent and strong signals from leaves harvested throughout the time course. The presence of virus specific sequences in leaf tissue after actual antigen concentration has decreased suggests that viral sequences are stable in cells in some form other than intact virions.

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INFLUENCE OF PLANTING DATE ON INCIDENCE OF THE GOLDEN MOSAIC DISEASE OF PHASEOLUS VULGARIS. J. A. M. Rocha, A. Sartorato, and C. A. Rava, CNPA/EMBRAPA, Golanla, Go., Brasil.

The bean golden mosaic virus often incites a devastating disease in Brasil. At the Santa Helena Research Station 20 selected bean cultivars (10 susceptible and 10 tolerant) were planted 11/1/78, 12/1/78, 1/4/79 and 2/5/79 to determine the effect of planting date on disease incidence. Disease data were recorded after 45 and 65 days, and yields at cultivar maturity. Disease incidence increased strikingly with each successive planting date. Yields for 20 cultivars averaged 940, 717, 76 and 0 kg/ha for the 1st, 2nd, 3rd and 4th planting, respectively. The 2nd planting date is considered most appropriate because harvest is more likely to occur in dry weather.

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FIELD BIOLOGY OF TOBACCO STREAK VIRUS IN EASTERN WASHINGTON. W. J. Kaiser*, S. D. Wyatt**, and G. R. Pesho*, *USDA, ARS, Regional Plant Introduction Station and **Department of Plant Pathology, Washington State University, Pullman, WA 99164.

Tobacco streak virus (TSV) naturally infects cowpeas (*Vigna unguiculata*) at Central Ferry, WA. TSV-infected cowpeas exhibited yellow mosaic symptoms or were symptomless. White sweet clover (*Melilotus alba*) is the primary reservoir and overwintering host of TSV in this region of eastern Washington. The virus was not isolated from 40 other wild species growing at Central Ferry. TSV was symptomless in *M. alba* and the virus was transmitted through <3% of the seeds of naturally infected *M. alba*. Virus isolates from white sweet clover and cowpeas were divided into two pathotypes on the basis of reactions in agar-gel double-diffusion tests and symptoms produced in *Chenopodium quinoa*, *Phaseolus vulgaris* 'Bountiful', and *Vicia faba*. TSV was transmitted from naturally infected *M. alba* to *C. quinoa* and *M. alba* by thrips (*Thrips tabaci* and/or *Frankliniella occidentalis*), but not by pea aphids (*Acyrtosiphon pisum*) or pea leaf weevils (*Sitona lineata*).

A SNOWMOLD DISEASE OF MOUNTAIN BIG SAGEBRUSH ARTEMISIA TRIDENTATA VASEYANA. D. L. Nelson and D. L. Sturges, USDA Forest Service, Intermtn. For. and Ra. Expt. Sta., Provo, UT 84601 and Rocky Mtn. For. and Ra. Expt. Sta., Laramie, WY 82070.

An unreported and unidentified fungus is causing a severe snowmold of mountain big sagebrush in late-lying snowpacks in Wyoming, Colorado and Utah. Patches of killed leaves and shoots on affected plant crowns expand annually, as the fungus invades, until after one to several winters entire plants may be killed. Fruiting structures of the causal fungus have not been found on diseased plants nor has the fungus been induced to sporulate in culture. A fungus with tough, hyaline, septate hyphae has been isolated that reproduced field symptoms on potted plants in coldroom inoculation tests at $1 \pm .5$ C. In southern Wyoming, temperatures in snowpacks in the sagebrush crown zone ranged from -4 to -16 C in early winter; however, in late winter the snowpack warms and becomes isothermal at 0 C. In temperature growth studies the isolate grew from a low of slightly below 0 C, had an optimum of near 12 C and grew little above 20 C.

DISEASES OF TREES IN WINDBREAKS IN OKLAHOMA. Kenneth E. Conway, Lou S. Morrison, and Mark W. Andrews. Department of Plant Pathology, Oklahoma State University, Stillwater, OK 74078.

Many of the windbreaks planted in Oklahoma during the Prairie States windbreak program (1940-45) are now in a state of decline. The six windbreaks selected for investigation in Kingfisher and Blaine counties in West Central Oklahoma were surveyed for diseases and wood rots at three month intervals from December 1980 - September 1981. Each windbreak differed in the number of structural components and tree species. The most severe diseases identified were; *Cytospora* dieback of Cottonwood, *Thyronectria austro-americana* canker and dieback on Honeylocust, *Phellinus robiniae* heart rot of Blacklocust, *Phellinus pini* root rot of Austrian and Ponderosa pines, and *Ganoderma* root rot of Honeylocust. Woodrotting fungi represented 45% of the total fungi observed and were indicative of the advanced state of decline of the windbreaks in this area.

RUSTS OF ACACIA IN HAWAII. C. S. Hodges and D. E. Gardner, USDA For. Serv., Honolulu HI 96813 and Natl. Park Resour. Stud., Dept Botany, Univ. of Hawaii, Honolulu HI 96822, respectively.

Three rusts infect endemic species of *Acacia* in Hawaii. *Uromyces koae*, first described in 1925, apparently is limited to the Hawaiian Islands and in its host range to *Acacia koa*. Spermogonia and aecia are associated with witches' brooms or hypertrophy of various tissues, including buds, flowers, seed pods, and phyllodes. Urediniospores and teliospores are produced in the same sorus on true leaves and phyllodes of *A. koa* and are not associated with hypertrophy. *Uromyces digitatus*, which also occurs in Australia, New Zealand and Java, is reported from Hawaii for the first time and affects *A. koa* and *A. koaia*. Spermogonia and telia are associated with sparsely branched witches' brooms up to one meter in height. Scattered uredinia and telia, but no spermogonia, are produced on normal phyllodes. A third rust on *A. koa* and *A. koaia* is tentatively identified as *Uraecium* sp. Aecia accompanied by spermogonia are produced on witches' brooms. Seven months after inoculation with aeciospores, small brooms with aecia and spermogonia were produced.

SEVERITY OF A GALL DISEASE OF RHIZOPHORA MANGROVES IN FLORIDA. H. J. Teas and J.B. Reark. Biology Dept., University of Miami, Coral Gables, FL 33124 and 6870 SW 75th St, South Miami, FL 33143.

Olexa and Freeman (Pl. Dis. Rep. 62:283-285) described a widespread disease of *Rhizophora mangle* in Florida, U.S.A., caused by *Cylindrocarpus didymum*. We have defined five severity stages of the disease: (1) trees healthy, no disease; (2) galls on prop roots, trunk or branches; (3) galls on more than one part of the tree; (4) trees heavily galled, top or at least one large branch dead; and (5) trees dead with obvious large galls. Although in some areas none of the trees show galls, in other areas 100% of the trees bear galls (stages 2 and 3) and we have found some areas where half the large trees are stage 5. The disease appears to be more lethal than had been recognized previously.

THE PINE WOOD NEMATODE ON BALSAM FIR IN MINNESOTA. M.J. Wingfield and R.A. Blanchette, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108; and E. Kondo, Faculty of Agriculture, Saga University, Saga, Japan.

The pine wood nematode (*Bursaphelenchus xylophilus*) has been found in balsam fir (*Abies balsamea*) in Minnesota. Trees from which nematodes were extracted had *Armillariella mellea*-infected roots and were infested with *Pityokteines sparsus* (Coleoptera: Scolytidae) and *Monochamus* spp. (Coleoptera: Cerambycidae). Adult female nematodes (Bx-b) from balsam fir have tails bearing a distinct mucro, are morphologically distinct from the nematode (Bx-p) from pine in Minnesota and more similar to *B. mucronatus* which occurs in Japan. Nematodes from isolate Bx-b mated with those from Bx-p while neither mated with *B. mucronatus*. Bx-p were reared on cultures of *Ceratocystis ips*, while Bx-b did not multiply on this fungus. Bx-b killed balsam fir seedlings but not red pine (*Pinus resinosa*) or Scots pine (*P. sylvestris*) in greenhouse inoculations, and Bx-p killed only pine seedlings.

THE EFFECT OF SOIL TEMPERATURE ON THE INVIVO GROWTH AND DEVELOPMENT OF VERTICILLADIELLA WAGNERII IN DOUGLAS-FIR ROOTS. P.F. Hessburg and E.M. Hansen. Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, Oregon 97331.

Roots of seedling and pole-sized Douglas-fir were artificially inoculated with the "black-stain" fungus *Verticilladiella wagnerii* and were sampled at one and two week intervals, respectively. Maximal fungal growth in roots was measured in axial, radial, and circumferential directions from the known point source of inoculum and mean growth rates were computed for each direction in each successive sample period. Soil temperatures were monitored continuously at both greenhouse and field sites. In a simple linear regression model of the form $log Y = a + b log X$ using temperature and fungal growth rate, temperature accounted for 98% of the variation in fungal growth observed between sample periods when temperatures were within the optimal range for growth (15-18° C). Outside of the optimal range (> 18.5°), temperature accounted for 82% of the variation and the slope of the regression line was significantly reduced ($p = .01$). Fungal growth-temperature regressions were derived from greenhouse seedling experiments.

DISTRIBUTION AND SEVERITY OF SWISS NEEDLE CAST IN DOUGLAS-FIR CHRISTMAS TREE PLANTATIONS. E. Michaels and G.A. Chastagner, Dept. of Plant Pathology, Wash. State Univ., Puyallup, WA 98371

In 1981, 53 Douglas-fir Christmas tree plantations in western Washington and Oregon were surveyed to determine the incidence and severity of Swiss Needle Cast caused by *Phaeocryptopus gaeumannii* (Robde) Pefrak. Presence of pseudothecia, loss of needles, and associated disease symptoms were tabulated for 50 randomly selected trees in each plantation. Of 6 plantations judged to be the worst, 71% (213) of the trees sampled had only current year's needles. The 42 remaining plantations had trees with varying degrees of needle loss ranging from 10% of needles up to 3 years old, to trees which had lost almost all 3- and 4-year-old needles, but retained most of their 2-year-old needles and virtually all of their current year's needles. Based on Christmas tree grading criteria, trees bearing only current year's needles, or current year's needles plus a portion of 2-year-old needles, would be unmerchantable. Trees with a lesser degree of needle loss would be merchantable, but may be reduced in grade.

DEHYDRATION AND NEEDLE LOSS ON CUT DOUGLAS-FIR CHRISTMAS TREES WITH SWISS NEEDLE-CAST. G. A. Chastagner, R. S. Byther, and E. Michaels, Department of Plant Pathology, Washington State University, Puyallup, WA 98371.

Swiss needle-cast (SNC), caused by *Phaeocryptopus gaeumannii* (Robde) Pefrak., commonly occurs on Douglas-fir Christmas trees grown in the Pacific Northwest. Needles become infected soon after bud break and normally discolor and drop 1 to 3 years later. In 1981, trees with symptomless diseased and healthy needles were compared for needle loss and dehydration after cutting. Treatments were: SNC controlled in both 1980 and 1981; SNC controlled in 1980; SNC controlled in 1981; SNC not controlled. Trees were cut Nov 12, stored outdoors, placed indoors at 20 C, 40-60% RH, and continuous light on Dec 7. Trees were either placed in water, or left dry. Controlling SNC in 1980, or in 1980 and 1981, reduced the rate of dehydration by 55 and 72%, respectively, on dry trees, and 29 and 53%, respectively, on wet trees. The rate of loss of 1980 needles was 80% higher for dry trees when SNC was not controlled during 1980. This was not true for 1980 needles on wet trees or the 1981 needles on dry or wet trees.

WETWOOD FORMATION AS A HOST RESPONSE IN WHITE FIR.

J. J. Worrall and J. R. Parmeter, Jr., Dept. Plant Pathology, University of California, Berkeley, CA 94720

Wetwood in white fir (*Abies concolor*) occupies the central cylinder of heartwood and is also associated with branch gaps, wounds, insect scars, and root and butt decay. In two inoculation experiments, the bacterium associated with wetwood did not cause wetwood, and wetwood formed while external water was excluded. *Fomes annosus*, large wounds, and treatment with $HgCl_2$ resulted in the formation of wetwood. Wetwood formed in seedlings which survived inoculation with *F. annosus* but not in those which were killed by the fungus or which received control inoculations. Large variation was observed between trees in quantity and quality of wetwood formed in response to various treatments. The results indicate that wetwood may be a host response to parenchyma death rather than a bacterial disease or an accumulation of external water.

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ETIOLOGY AND DISTRIBUTION OF ROOT DISEASES ON NORTHERN ROCKY MOUNTAIN CONIFERS. J. W. Byler, R. L. James, and S. H. Dubreuil. USDA Forest Service, Forest Pest Management, Northern Region, P.O. Box 7669, Missoula, Montana 59807.

Conifer root diseases are widespread throughout forests of the northern Rocky Mountains. Centers of infection and scattered mortality occurs over large areas. Although Douglas-fir and true fir are most susceptible, other conifers are also affected. *Armillaria mellea* is common throughout Idaho and Montana on many hosts. It often attacks trees previously infected with other root pathogens and also commonly kills regeneration. *Phellinus weirii* infects primarily grand fir and Douglas-fir in northern Idaho and northwestern Montana, often causing large mortality centers. *Phaeolus schweinitzii* infect all conifer species but is most frequent on Douglas-fir. *Fomes annosus* occurs on ponderosa pine and subalpine fir. *Verticicladiella wagnerii* has been found on Douglas-fir and several pine species. Bark beetles frequently attack infected trees. Root diseases severely limit management options in many forest areas.

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ROOT DISEASE MORTALITY OF NORTHERN ROCKY MOUNTAIN CONIFERS. R. L. James, C. A. Stewart, R. E. Williams, and J. W. Byler. USDA Forest Service, Forest Pest Management, Northern and Inter mountain Regions, P.O. Box 7669, Missoula, MT 59807, and 305 N. 5th St., Boise, ID 83702.

Root disease impact surveys in northern Idaho and western Montana show disease mortality centers occupy 1.9%, 0.4%, 1.0%, and 1.2% of the commercial forest area of the Idaho Panhandle, Clearwater, Nezperce, and Lolo National Forests, respectively. This represents more than 30,000 ha of nonproductive forest. Actual area infected is probably much greater. Root disease associated mortality losses are estimated at 0.4 and 2.2 trees/ha/yr (0.3 and 0.8 m^3 /ha/yr) for the Clearwater and Nezperce National Forests, respectively. About 30% of the annual conifer volume loss on these two Forests is associated with root diseases. Using USFS regional timber mortality figures, we estimate that 0.3 and 0.2 m^3 /ha/yr are lost annually to root diseases in northern Idaho and Montana, respectively. This represents about 2.3 $MM m^3$ /yr for the entire region, which amounts to 40% of the average annual timber harvest.

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HOST-SPECIFICITY WITHIN VERTICICLADIELLA WAGNERII, CAUSE OF BLACK STAIN ROOT DISEASE OF CONIFERS. T. C. Harrington, and F. W. Cobb, Jr., Dept. Plant Pathology, University of California, Berkeley, CA 94720.

Three host-specific groups within *V. wagnerii* (1) on pinyons, (2) on other *Pinus* spp., and (3) on Douglas-fir can be identified by mycelial characters but not by size and shape of conidiophores or conidia. Each of 89 isolates of *V. wagnerii* was morphologically assignable to the group that was predicted on the basis of host of origin. Host specificity of these groups was investigated by inoculating seedlings and mature trees of ponderosa pine and Douglas-fir. Inoculation of seedlings in the greenhouse resulted in significant differences in percent infection and disease severity among the groups. Stronger host preferences were shown with wound-inoculation of mature trees than with inoculation of seedlings. In both experiments, pine isolates infected a few Douglas-fir and vice versa, but such cross-overs are apparently rare in nature. Designation of special forms within *V. wagnerii* appears to be warranted.

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CONIFERS VARY IN SUSCEPTIBILITY TO GREMMENIELLA ABIETINA. George W. Hudler, Guy R. Knudsen, and Mary Ann Beale, Dept. of Plant Pathology, Cornell University, Ithaca, NY 14853.

In May 1979, seedlings of *Pinus resinosa*, *P. sylvestris*, *P.*

strobus, *Picea glauca*, and *P. abies* were planted in a field in northern New York. Four wk later they were sprayed to runoff with water or with a suspension of conidia of *Gremmeniella abietina* ranging in concentration from 10^2 - 10^5 conidia/ml. Seedlings showing symptoms of infection were counted and rogued in 1980. Remaining seedlings plus some replacements were reinoculated in 1980 and examined in 1981. In both yr, disease occurrence was high on *P. resinosa* and was moderate on *P. sylvestris* and *P. strobus*. On all *Pinus* spp., a positive correlation between inoculum concentration and disease incidence was observed. *G. abietina* was rarely found on *Picea* spp. and then was only on dead, lower branches where it appeared to be saprobic. Our results indicate that New York law restricting shipment of most conifers grown within 1000 ft of trees infected with *G. abietina* is unduly conservative with respect to *Picea* spp., and we suggest that it be amended.

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VARIATION IN ISOLATES OF DIPLODIA PINEA IN THE NORTH CENTRAL UNITED STATES. Marguerita A. Palmer and Elwin L. Stewart, North Central Forest Exp. Station, St. Paul, MN 55108; and Dept. of Plant Pathology, Univ. of MN, St. Paul, MN 55108.

Shoot blight and canker caused by *Diplodia pinea* (Desm.) Kickx are important problems in conifer plantations and nurseries in north central United States. Many conifer species are affected, although the most serious losses occur in red pine (*Pinus resinosa* Ait.). Isolates of *D. pinea* from red and jack pine (*P. banksiana* Lamb.) were found to differ in several characteristics. The "red pine type" (RP) isolate produces fluffy, gray-green mycelium when grown on PDA. The "jack pine type" (JP) produces gray to black mycelium closely appressed to the agar. JP isolates sporulate readily in daylight at 23° C, whereas RP isolates sporulate only if sterile host tissue is added to the culture. The RP isolate spores are longer and narrower than those of the JP isolate. In greenhouse studies, pathogenicity of isolates differed on red and jack pine seedlings, depending on the inoculation method used. The differences in these isolates suggest the presence of a new strain of the organism.

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SEVERITY OF ARMILLARIA MELLEAE IN SOIL WITH LOW PH AND NUTRIENT DEFICIENCY. Pritam Singh, Newfoundland Forest Research Centre, Canadian Forestry Service, St. John's, Nfld. Canada.

Armillaria mellea is a facultative parasite but can be an aggressive pathogen of trees growing in adverse site and soil conditions. Investigations were conducted to determine the combined effects of soil nutrients and pH on the susceptibility of four softwood species. Seedlings of Norway, black and Sitka spruces, and Scots pine, growing on nutrient deficient and non-deficient soils in a greenhouse, were inoculated with *A. mellea* and development of the root rot studied. The deficient soil had a lower pH, and trees growing on it were smaller and less vigorous. A higher percentage of roots and trees were infected and more trees died on deficient soil than on non-deficient soil. Non-vigorous trees also contacted infection earlier than the vigorous trees. Roots of vigorous trees showed more resinosis and callus formation, and less mycelial growth, indicating resistance during the establishment of the fungus on the host. Alterations in the levels of nutrients in vigorous and non-vigorous, infected trees were generally similar.

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BASAL AREA LOSS OF BLACK SPRUCE DUE TO MORTALITY BY EASTERN DWARF MISTLETOE IN THE KOOSCHICHING STATE FOREST. Paul Scherman, William Livingston, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108; Michael Albers and Gerald Hecht, Minnesota Department of Natural Resources, Grand Rapids, MN 55744.

Black and white aerial photography (scale 1:15,840) for one township in the Koochiching State Forest was interpreted to locate mortality centers and determine basal area losses caused by eastern dwarf mistletoe (*Arceuthobium pusillum* Peck). Due to difficult access, suspected mortality centers were checked by helicopter. Seven of the 32 stands checked were infested with *A. pusillum*, of which five were correctly identified and two were not identified from the aerial photos. Of the total area checked, 89 acres were out of production, and 85 of the 89 acres were in one stand. Basal area lost was 5% of the total basal area.

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MYCOSPHAERELLA LARICINA, THE CAUSE OF A NEEDLE CAST OF EUROPEAN LARCH IN WISCONSIN PLANTATIONS. Robert F. Patton and Russell N. Spear, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Mycosphaerella laricina, first described on larch in Europe, was

identified as the cause of a needle-cast disease that has appeared recently on European larch (*Larix decidua*) in a few Wisconsin plantations. This is the first report of this fungus in the United States. Tips or sections of needles turn yellow then rapidly become necrotic and reddish-brown. Symptoms appear in early June and continue to develop and intensify throughout the summer. Needles are cast prematurely and trees may be completely defoliated. Black acervuli bearing hyaline needle-like conidia, 1-3 septate, 24-45 μ m long soon appear in the necrotic portions. Spermagonia appear in late fall and mature in mid-December on dead, cast needles. Perithecia mature in May in a submerged to erumpent stroma. Hyaline ascospores are equally 2-celled, narrowed and rounded at each end. Both conidia and spermagonia form in cultures, which range from white to black.

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MYCOSPHAERELLA NEEDLE CAST OF EUROPEAN LARCH. M. Ostry, T. Nicholls, M. Palmer, and K. Robbins, USDA Forest Service, 1992 Folwell Ave., St. Paul, MN 55108.

Intensive management of European larch, *Larix decidua*, is being considered in the United States. Rapid growth and good wood properties and pulping characteristics have increased the interest of industry in growing this exotic species. R. F. Patton recently identified *Mycosphaerella laricina* as the cause of premature defoliation of European larch in Iowa, Michigan, and Wisconsin. *M. laricina*, previously thought to occur only in Europe, has caused growth losses and is potentially a serious threat to European larch in this country. Primary infection of needles in Wisconsin and Iowa by ascospores begins in late April and secondary infections resulting from conidia begin in June. Symptoms first appear in early June and heavily infected trees are defoliated by early July. Disease severity varies by seed source of European larch. Japanese larch, *L. leptolepis*, appears to be immune, and hybrid larch, *L. eurolepis*, moderately susceptible. Planting resistant species and larch trees from resistant seed sources will minimize the impact of this disease.

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SYMPTOMS AND DISTRIBUTIONS OF PHAEOLUS SCHWEINITZII AND ARMILLARIA MELLEAE IN ROOT SYSTEMS WITHIN MIXED CONIFER STANDS. S. H. Dubreuil and N. E. Martin, USDA Forest Service, Forest Pest Management, P. O. Box 7669, Missoula, MT 59807 and Intermountain Forest and Range Experiment Station, 1221 S. Main St., Moscow, ID 83843.

Patterns of infection by *Phaeolus schweinitzii* and *Armillaria mellea* were studied in 29 tree root systems within two mixed stands. Douglas-fir and grand fir were most extensively infected and ponderosa pine and western larch were least affected. *P. schweinitzii* infections originated in root tips and spread toward the root crowns. Root contacts and wounds were inconsequential as sources of *P. schweinitzii* infection. Root galls were frequent on Douglas-fir. Most *A. mellea* infections in live Douglas-fir were limited to tissue within root gall stubs. In grand fir roots they were in the outerwood of main roots in and near the root crown.

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RESISTANCE TO PHOMOPSIS JUNIPEROVORA AMONG PROGENIES OF JUNIPERUS VIRGINIANA. Glenn W. Peterson. Rocky Mountain Forest and Range Exp. Stn., Forestry Sciences Laboratory, Univ. Nebr., Lincoln, 68583.

Progenies from 86 *Juniperus virginiana* trees selected from within the Great Plains were evaluated for resistance to *Phomopsis juniperovora*. Seedlings with new (yellowish-green) foliage were inoculated with spore suspensions from a single isolate, incubated at 24 C for 24 hr, and then placed in a greenhouse. Ten seedlings of each progeny were inoculated in each of 7 tests. After 2 wk, 25 branchlets on each seedling were examined for symptoms. Among progenies, the average percentage of infected branchlets ranged from 5 to 38 and the average percentage of infected trees ranged from 40 to 100. Among the 7 least severely infected progenies, the average percentage of infected branchlets ranged from 3 to 13 and the average percentage of infected trees ranged from 40 to 80. Seed of the least severely infected progenies were from trees in South Dakota (1), Nebraska (2), Kansas (3), Oklahoma (1).

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PEROXIDASE ACTIVITY IN RELATION TO ADULT PLANT RESISTANCE OF OATS TO ERYSIPIHE GRAMINIS F. SP. AVENAE. S.M. Douglas, *R.T. Sherwood, and F.L. Lukezic. Dept. of Plant Pathology, The Pennsylvania State University and *U.S. Regional Pasture Research Laboratory, USDA-ARS, University Park, PA 16802.
Soluble peroxidase activity was significantly higher in 4th-

formed leaves (resistant) than 2nd-formed leaves (susceptible) of Maldwyn (adult plant resistant). Reversible heat shocking was used to manipulate resistance. Leaves heat shocked (50 C, 2 min) before inoculation were readily penetrated and lacked cytoplasmic aggregates, papillae, and autofluorescence. Peroxidase activity increased significantly in 4th-formed leaves 24 h after inoculation, but no significant change in activity was found in 2nd-formed leaves. Heat shocked 4th-formed leaves had significantly lower peroxidase activity than their non-heat shocked counterparts 24 h after inoculation. However, heat shocked and non-heat shocked 2nd-formed leaves showed no significant difference in peroxidase activity 24 h after inoculation. Peroxidase activity in 4th-formed leaves of Mariner (susceptible) and Maldwyn was compared to evaluate variety response to infection.

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THE ROLE OF GLYCEOLLIN IN SOYBEAN ROOT TOLERANCE TO PHYTOPHTHORA ROOT ROT. A. F. Olah, A. F. Schmitthenner, and A. K. Walker, Ohio Agricultural Research and Development Center and The Ohio State University, Wooster 44691.

Soybeans with *Rps1^a*, *Rps1^c* or no resistance and either high or low tolerance to root rot incited by *Phytophthora megasperma* f. sp. *glycinea* (Pmg) were challenged with compatible and incompatible races of Pmg by root or hypocotyl inoculation. Glyceollin accumulation at the inoculation site was measured. Maximum glyceollin accumulation in the hypocotyl was higher than in the root and occurred earlier. In hypocotyls of incompatible combinations glyceollin levels were >300 μ g/g fr wt, whereas little was detected in roots. In hypocotyls of compatible combinations glyceollin levels were less than 100 μ g/g fr wt, with highly tolerant cultivars accumulating twice as much as slightly tolerant cultivars. Twice as much glyceollin accumulated in roots of compatible combinations than in the hypocotyls, but the levels reached did not reflect differences in root tolerance. Glyceollin accumulation does not appear to explain soybean tolerance to *Phytophthora* root rot.

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MEASUREMENT OF PEROXIDASE ACTIVITY AS A QUANTITATIVE ASSAY FOR ELICITOR ACTIVITY OF CLADOSPORIUM FULVUM GLYCOPEPTIDES. H.J. Sugiyama and V.J. Higgins, Dept. of Botany, University of Toronto, Toronto, Canada M5S 1A1.

Glycopeptides produced *in vitro* by the tomato leaf mold pathogen *Cladosporium fulvum* have previously been shown to elicit callose deposition, necrosis, and phytoalexin synthesis when injected into tomato leaves. Infection of tomato by *C. fulvum* resulted in increased leaf peroxidase activity. This effect could be duplicated by injection with the glycopeptides. Elevated levels of peroxidase activity were detected 12 hr after injection and increased with glycopeptide concentration over the range 5 to 25 μ g glucose equivalents/ml. Peroxidase activity peaked 18 to 30 hr after treatment and then declined sharply and the activity appeared to be correlated to the severity of the visible necrosis. Stimulation of peroxidase activity by glycopeptides was not cultivar specific. The ease of measuring peroxidase activity on small amounts of injected tissue provides a useful routine assay for activity of these glycopeptides.

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CORRELATIVE PATTERNS OF NECROSIS AND FUNGAL GROWTH IN TWO EXAMPLES OF CULTIVAR RESISTANCE TO THE COWPEA RUST FUNGUS. Michèle C. Heath, Botany Department, University of Toronto, Toronto, Ontario, Canada M5S 1A1.

The growth of one strain of the cowpea rust fungus was compared in two resistant cowpea cultivars using light and electron microscopy. In cultivar TVu 4552, intercellular fungal growth usually ceased after the rapid necrosis of the first cell invaded by a haustorium. A delay in this necrosis in ca. 25% of infection sites correlated with continued intercellular growth but such growth soon ceased during a second wave of necrosis involving host cells with and without haustoria. In cultivar Purple Hull Pinkeye, the first cell invaded by a haustorium seemed to remain healthy for up to 4 days. Intercellular growth continued for ca. 7 days but slowed as the number of necrotic host cells per unit of intercellular mycelium increased. These results suggest that in the two cowpea cultivars examined, there is a close inverse relationship between the patterns of host necrosis and fungal growth.

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PRESENCE OF WILT INDUCING FACTOR(S) IN TRACHEAL FLUID OF PHYMATOTRICHUM-INFECTED COTTON PLANTS. M.W. Olsen, I.J. Misaghi, and D. Goldstein, Department of Plant Pathology,

Resistance to water flow in roots and lower stems of cotton plants infected by *Phymatotrichum omnivorum* was found to increase significantly in early stages of disease development. Xylem elements in roots and lower stems of infected plants are discolored and appear to be occluded. To test the possibility of the presence of a factor in tracheal fluid in infected plants which might be responsible for occlusion of xylem elements and increased resistance to water flow, the cut-end of stems of excised cotton seedlings were placed in xylem sap collected from healthy and diseased plants. Xylem sap from infected cotton plants wilted seedlings while sap from healthy plants did not. These preliminary results suggest that a substance produced in infected roots may move upward in the plants and cause xylem occlusion and increased resistance to water flow.

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PHYMATOTRICHUM OMNIVORUM PRODUCES WILT INDUCING FACTORS IN VITRO. P.J. Cotty and I.J. Misaghi, Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

In our search for the mechanism of wilt development in plants infected with *Phymatotrichum omnivorum* we have considered the possibility that wilting might be due to the occlusion of xylem elements with a fungus-produced, high-molecular-weight substance(s). Among different fungal products tested, a water-soluble, hydrochloric acid and methanol insoluble, non-dialyzable, pigmented substance, produced *in vitro*, was found to exhibit wilt inducing activity in excised cotton seedlings at the two-leaf stage. A partially purified preparation of this wilt inducing factor (2.0 mg/ml) caused a 40 to 55% reduction in transpiration rate compared to controls. A highly pathogenic isolate of the fungus exudes large quantities of the factor in droplets on the surface of fungal colonies. This native exudate, collected aseptically, was highly active with a dilution end-point of 1:100 and caused wilting of excised cotton seedlings at the cotyledon stage within 2 hr. The chemical nature of the wilt inducing factor is being investigated.

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STUDIES ON THE ADHERENCE OF AGROBACTERIUM TUMEFACIENS CELLS TO POTATO TUBER TISSUES. J. A. Bluepfel, S.G. Pueppke, Department of Plant Pathology, University of Florida, Gainesville, FL 32611.

We developed a radiolabeled bacteria-binding assay to assess the adherence of *A. tumefaciens* to potato tuber tissue. During a 10 min incubation period, between 300,000 and 500,000 oncogenic bacterial cells from an inoculum of 2×10^7 cells adhere firmly to tuber tissue. Four polygalacturonides, which are from 0 to 89% methylated, reduce tumor number from 55 to 70%, but the inhibitory activity is unrelated to the degree of methylation. At the same concentration (1 mg/ml) the polygalacturonides have essentially no inhibitory effect on the adherence of bacteria to plant tissues (average reduction is 3%). In contrast, brief exposure of oncogenic bacteria to proteolytic enzymes causes marked changes both in the ability of the bacteria to adhere to potato tissues and in tumor formation.

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CARBOHYDRATE METABOLISM IN BLUE MOLD INFECTED TOBACCO LEAVES. W. E. McKeen and A. M. Svircev
Plant Sciences Dept., The University of Western Ontario,
London, Ontario, Canada. N6A 5B7

Tobacco leaves during the first four days after infection with *Peronospora hyoscyami* f.sp. *tabacina* synthesize starch at the same rate as healthy tobacco leaves when they are maintained at 18°C and in a light intensity of 1500 micro Einsteins per $M^2 \text{ Sec}^{-1}$. By the fifth day starch formation is retarded in the infected leaves. Starch degradation is retarded four hours after inoculation and thereafter in diseased leaves. Extracts from healthy and infected leaves degraded commercial starch at different rates. This physiological change may insure the pathogen a supply of carbohydrate during cloudy periods.

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ALTERED HOST RHYTHMS INDUCED BY BIOTIC AND ABIOTIC STRESS. B.W. Kennedy, W.L. Koukkari and F.M. Guillaume, Department of Plant Pathology and Department of Botany, University of Minnesota, St. Paul, MN 55108.

Stress in plants induced by biotic and abiotic agents was analysed by a cosine curve-fitting technique to measure the

amplitude, period and phase of circadian (~24 hr) and ultradian (<20 hr) leaf movement rhythms. The effects of pathogens on rhythms were examined in several host-pathogen combinations, including *Cossypium hirsutum* inoculated with *Verticillium albo-atrum* and in *Phaseolus vulgaris* inoculated with *Xanthomonas campestris* pv. *phaseoli* and *Pseudomonas syringae* pv. *phaseolicola*. The presence and characteristics of rhythms were dependent upon leaf age and the specific combination of host and pathogen. Sometimes susceptibility of plants to injury depended upon time of day of exposure to a given pathogen or environmental agent (eg. heat).

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THE POSSIBLE ORIGIN OF ETHYLENE IN VERTICILLIUM WILT OF TOMATO. H. Mussell, P. Stilwell & S. Peck, Boyce Thompson Institute, Cornell University, Ithaca, NY 14853.

Cell walls prepared from foliage and stems of tomato plants contained an enzyme system capable of generating ethylene from ACC. This enzyme complex could be solubilized from particulate cell wall preparations by incubating the cell walls with highly purified endopolygalacturonase from *Verticillium dahliae*. The solubilized enzyme system was more active than when associated with the particulate fraction. The enzyme system required oxygen and an oxidizable substrate to generate ethylene from ACC. Although several substrates would trigger the appearance of ethylene, maximum rates of ethylene evolution were observed when the soluble enzyme system was provided with indoleacetic acid, Mn²⁺, and p-coumaric acid. The above information, and the fact that ethylene generation was inhibited by a competitive inhibitor of indoleacetic acid oxidase, provide the first direct evidence for a metabolic link between regulation of the levels of these two important plant hormones.

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TECHNOLOGY OF MEASURING RHYTHMIC LEAF PATTERNS IN DISEASED PLANTS. F.M. Guillaume, W.L. Koukkari and B.W. Kennedy, Dept. of Botany and Dept. Plant Pathology, University of Minnesota, St. Paul, MN 55108.

A computer-assisted system permitted simultaneous monitoring of leaf rhythms of healthy and diseased plants. The measuring device was an angular position sensor, a fulcrum, and a lever connected to the leaf by a silk thread. Leaf movements were converted to voltage variations. The data acquisition system consisted of a 48k computer, disk drive, 16 channel 8 bit A/D converter, and an electronic clock-calendar. Sampling intervals ranged from 1 sec to 20 min. Data processing was based on discrete Fourier transform methods and double complex demodulation. During a 20 day test of the device, leaf movements of *Glycine max* (L.) Merr. were recorded by an analog method. Ultradian (<20 hr) oscillations with periods averaging 54 min. were detected. The ultradian oscillations had a complex, but day to day reproducible sequence, of transitory and steady state patterns which may have value in diagnosis.

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SEQUENTIAL USE OF LIGHT MICROSCOPY, SCANNING ELECTRON MICROSCOPY, AND X-RAY MICROANALYSIS TO STUDY POWDERY MILDEW INFECTION. T.L.W. Carver, R.J. Zeyen, and G.G. Ahlstrand, Welsh Plant Breeding Station, Aberystwyth, U.K.; and Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Light microscopic details of *Erysiphe graminis hordei*-host cell interactions were observed in acetic-ethanol fixed barley leaves stained with Coomassie Brilliant Blue R-250. Encounter sites were mapped and details photographed. Tissues were then critical point dried, encounter sites relocated by SEM, and insoluble silicon (Si) was located by energy dispersive X-ray microanalysis. Silicon was found in papillae and throughout the area subtended by the cytoplasmic aggregate, regardless of fungal success or failure. High levels of Si accumulated throughout epidermal cells in certain dead cells characterized by dense staining cytoplasm. This sequential procedure allows for accurate interpretation of X-ray microanalysis through prior knowledge gained by light microscopy, and can be extended to include procedures other than bright field observations.

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X-RAY MICROANALYSIS OF PAPILLAE ISOLATED FROM BARLEY COLEOPTILES. *H. Kunoh, *J. R. Aist, and *H. W. Israel, *Faculty of Agriculture, Mie University, Tsu-city, 514 Japan, and *Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Our previous studies indicated that: i) normal papillae (NP's) induced in barley coleoptiles by *Erysiphe graminis hordei* were unable to stop the fungus; ii) oversize, preformed papillae

(PFP's), formed in the presence of $\text{Ca}(\text{H}_2\text{PO}_4)_2$, were resistant to penetration; and iii) $\text{Ca}(\text{H}_2\text{PO}_4)_2$ may have been directly inhibitory to penetration. The potential role of $\text{Ca}(\text{H}_2\text{PO}_4)_2$ in PFP's and an earlier report of Si in barley leaf papillae prompted us to microscopically isolate individual papillae and determine their elemental composition by electron microprobe analysis. NP's contained Si, P, Cl, S and Ca as major peaks and Mg, Al and K as minor peaks. By contrast, PFP's contained Ca and P as major peaks and S and Cl as minor peaks. The peaks of Ca and P were considerably higher for PFP's than for NP's. It is inferred that resistance of PFP's is more likely related to elevated levels of Ca and P [possibly as $\text{Ca}(\text{H}_2\text{PO}_4)_2$] than to Si content.

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EFFECTS OF HEAT-SHOCK ON VARIETAL AND NONHOST RESISTANCE IN CUCUMBERS. B.A. Stermer and R. Hammerschmidt, Dept. of Botany and Plant Pathology, Michigan State Univ., East Lansing, MI 48824

Cucumbers resistant and susceptible to *Cladosporium cucumerinum* were heat treated for 40 s at 50 C. The heat-shock temporarily induced susceptibility in both varieties to *C. cucumerinum* and nonpathogen *Helminthosporium carbonum*. Delayed inoculations showed that normal resistance was regained within 48 h. Heat-shocked but uninoculated plants did not appear to suffer any permanent effects. Heat-shocked plants inoculated immediately after treatment developed less lignin-like material in their cell walls compared to inoculated unshocked controls or shocked plants inoculated 24 h after treatment. The heat-shocked plants also supported greater spore germination and hyphal growth when inoculated immediately after treatment, but germination and hyphal growth was the same as unshocked controls when inoculated 24 h after treatment. Suppression of lignin deposition was always correlated with susceptibility. Heat-shock seemed to delay the dynamic responses of the plant, including those activities essential for disease resistance.

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EFFECT OF TEMPERATURE ON PENETRATION OF WHEAT LEAVES BY BIPOLARIS SOROKINIANA. C.A. Stockwell and R.T. Sherwood, Dept. of Plant Pathology, The Pennsylvania State University and U.S. Regional Pasture Research Laboratory, USDA-ARS, University Park, PA 16802.

Spring wheat cv. Thatcher was inoculated with a compatible isolate of *B. sorokiniana* and incubated in a dew chamber at 12, 20, or 25 C. At 48 hr, we assessed events that accompanied successful and unsuccessful penetration attempts on third-formed leaves. Increased temperature resulted in increased frequency of appressoria that were multicellular, increased frequency of successful penetration, and enlarged area of induced autofluorescence. Location of penetration attempts and frequency of papilla formation were not temperature-dependent. Significantly greater numbers of successful penetrations occurred at guard and accessory cells than in long cells at all temperatures. Resistance to penetration did not appear to be determined by papilla formation.

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INHIBITION OF THE HYPERSENSITIVE REACTION OF BARLEY TO *ERYSIPHE GRAMINIS* F. SP. *HORDEI* BY CYTOCHALASIN B AND HEAT SHOCK. Beth E. Hazen, Department of Botany, University of Minnesota and W.R. Bushnell, Department of Plant Pathology, Cereal Rust Laboratory, USDA, ARS, St. Paul, MN 55108.

Cytochalasin B (CB) and heat shock were tested for their ability to inhibit the hypersensitive reaction (HR) of barley to powdery mildew without inhibiting the fungus. The treatments were applied to epidermal tissue partially dissected from barley coleoptiles with the *Mla* gene for resistance before or after inoculation with *E. graminis* f. sp. *hordei*, race 3. HR was suppressed but the fungus was not by 10 µg/ml CB applied at 13 h or 50 µg/ml CB at 17 h after inoculation and by a preinoculation heat shock at 55 C for 45 sec. Both treatments partially or completely inhibited cytoplasmic streaming in host cells. These results suggest that HR depends on processes or structures related to cytoplasmic streaming.

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HEAT SHOCK BREAKS DISEASE RESISTANCE IN PEAS BY BLOCKING THE FORMATION OF RESISTANCE-RESPONSE-SPECIFIC mRNAs. L.A. Hadwiger and Wendy Wagoner, Dept. of Plant Pathology, Washington State University, Pullman, WA 99164-6430.

As pea pod tissue at 22°C resists *Fusarium solani* f.sp. *phaseoli*, the rate of synthesis of a specific group of 20 proteins is

simultaneously enhanced. The resistance of f.sp. *phaseoli* can be blocked with inhibitors of RNA or protein synthesis or by simple heat shock for 2h at 40°C. The 2h-40°C heat shock treatment causes the formation of mRNAs which preferentially code (in a rabbit reticulocyte *in vitro* protein synthesis system) for 5 or more major proteins which are not coded by mRNAs from non-treated tissue. Pea tissue after being challenged 4 additional h at 22°C following heat shock with f.sp. *phaseoli* do not accumulate the mRNAs required to enhance synthesis of the resistance-response-specific proteins. Messenger RNA from heat shocked non-inoculated tissue, following a 4h recovery period at 22°C codes for lower levels of the 5 heat shock proteins. Thus heat shock appears to program an mRNA synthesis response which cannot be superseded by the programmed response required for disease resistance until after a 4h recovery period.

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TWO DIMENSIONAL ELECTROPHORETIC SEPARATION OF PROTEINS CODED BY mRNAs ISOLATED FROM *FUSARIUM SOLANI* SPORES WHICH WERE: GERMINATED, UNGERMINATED, OR GERMINATED/RESISTED. L.A. Hadwiger and Wendy Wagoner, Dept. of Plant Pathology, Washington State University, Pullman, WA 99164-6430.

The growth of *F. solani* f.sp. *phaseoli* (bean pathogen) and f.sp. *pisi* (pea pathogen) macroconidia are completely terminated and temporarily inhibited, respectively, within 4-24 h after being applied to pea pods. Growth reduction occurs in association with accumulations of chitosan, an antifungal compound present in fungal walls. Translatable mRNA (per 5µg extracted RNA) was extremely low in ungerminated f.sp. *phaseoli* spores and low in resisted sporelings removed from pods. A qualitative difference in synthesis was detected in the 2-dimensional electrophoretic patterns of proteins coded by mRNAs from the two f.sp. Quantitative differences were detected within each f.sp. between germination/resisted growth on plant and germination/growth in Vogel's medium. Thus the two f.sp. do not synthesize the same spectra of proteins and their genetic expression appears to be quantitatively changed when in contact with host tissues. Is growth on host tissue limited by an inadequate supply of mRNA?

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EFFECT OF VERTICILLIUM WILT ON LEAF WATER POTENTIAL AND PROLINE ACCUMULATION IN COTTON PLANTS DURING DISEASE DEVELOPMENT. Dean D. Tzeng and J. E. DeVay, Department of Plant Pathology, University of California, Davis, CA 95616

Greenhouse-grown cotton plants (*Gossypium hirsutum* cv. Acala SJ-2) were stem-puncture inoculated with conidial suspensions of T9 (defoliating) and SS4 (non-defoliating) isolates of *Verticillium dahliae*. Both T9- and SS4-inoculated plants had slightly higher midday leaf water potentials (Ψ_L) than healthy controls for 4-6 days after inoculation. Midday Ψ_L values in T9-inoculated plants continued to increase with time after inoculation in contrast to Ψ_L in SS4-inoculated plants which decreased sharply 8 days after inoculation. Reduction of Ψ_L in SS4-inoculated plants paralleled an abrupt increase in proline accumulation. Proline accumulation in T9-inoculated plants was less than in SS4-inoculated plants, but it was accompanied by an increase of free amino acids and a decrease in chlorophyll. In contrast to SS4-inoculated plants, senescence phenomena in T9-inoculated plants appear to be associated more with defoliation than with water stress.

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WATER RELATIONS OF CITRUS SEEDLINGS INOCULATED WITH *FUSARIUM SOLANI*

S. Nemec, USDA, ARS, Orlando, FL 32803, and J. Syvertsen, Univ. of Florida, Lake Alfred, FL 33850

Fusarium solani infection in fibrous roots of citrus seedlings causes root rot and wilt symptoms. Wilted leaves on diseased plants exhibited lower water potential and water content, higher osmotic values (mOsm), and a higher diffusive resistance than leaves on healthy plants. Transpiration and root conductivity were lower in diseased plants compared to healthy plants. Visible symptoms of root rot and wilt occur as early as 24 to 48 h after inoculation. During this early phase of infection, cortical cells are quickly colonized and destroyed, little infection is present in the xylem, and vessel plugging has not yet formed. Wilting appears to be due to the inability of roots to supply water to the leaves. In the field, *F. solani* is associated with fibrous root rot symptoms and can be present in wood of larger roots.

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EFFECTS OF CAPTAN ON GERMINATION, DEVELOPMENT, AND NITROGEN FIXATION IN FIELD PEAS. A. W. Helton and Richard Dilbeck, Dept.

When Captan-80 (Stauffer) was applied to cv. Alaska seeds at 63.5, 179, or 610% of recommended field rate (0.7 g a.i./kg seeds), significant reductions occurred in radicle length only at high concentrations (179=44% reduction in comparison with controls; 610=47%) in germination plates, N-fixation rate (610=32%; measured with the acetylene-ethylene assay) at the onset of flowering (28 days from planting), and in root volume (179=44%; 610=47%) and root dry weight (179=40%; 610=40%) at maturity (66 days from planting) in the greenhouse. When Captan-30 (Gustafson) was applied to Alaska seeds at 19.7, 76.6, and 123% of field rate (0.5 g a.i./kg seeds), a significant high-concentration reduction occurred only in radicle length (123=20%) and in root volume (123=37%) at maturity. When Captan-30 was applied to cv. Garfield seeds at 19.5, 71.8, or 123% of recommended field rate, significant increases occurred in N-fixation rate at all treatment levels (19.5=39.5% increase; 71.8=20.6%; 123=26.3%) at the onset of flowering (43 days from planting).

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RHIZOCTONIA AND SEED DECAY CONTROL IN POTATOES BY DIRECT APPLICATION OF FORMALIN IN THE FURROW AT PLANTING. Terry Miller and Richard Wilkinson. Miller Research Inc. Rupert, Id. 83350.

For a number of years, we have been concentrating efforts on resurrecting formalin as a potato seed treatment to control disease. Our efforts have generally met with success and we have been able to control disease and improve potato crops (Potato Grower of Idaho 10:16, 1981). Because of some difficulties in treating seed with formalin, we experimented with treatment of the soil with formalin during planting. To do this, an Acme cup-type planter was fitted with 4" nozzles behind the opening shoe and various concentrations of formalin were metered into the furrow. The data generated during the 1981 season showed that the incidence of both *Rhizoctonia* and decay were reduced with this method. We had conservatively started at a low formalin concentration (5%) and progressed as high as 65%. Disease control was not apparent except at higher concentrations on silt loam soil. There were no signs of phytotoxicity or of delayed emergence. Our studies during 1982 will expand this work to higher concentrations and at injection sites closer to the seed. SUPPORTED IN PART BY THE IDAHO POTATO COMMISSION

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THE EFFECTS OF FUNGICIDE TREATMENTS ON YIELD AND QUALITY OF TOMATO PLANTS INFECTED WITH *LEVEILLULA TAURICA*. Wayne B. Jones and Sherman V. Thomson, Department of Biology, UMC 45 Utah State University, Logan, Utah 84322.

Field trials were conducted with 5 fungicides to determine the effect of tomato powdery mildew (*Leveillula taurica* (Lev.) Arn.) on yield and quality of tomatoes. Tilt (438.5 ml/ha, 3.6 EC), Bayleton (140 gm/ha, 50% W) and Benlate (1.12 kg/ha, 50% W) were sprayed biweekly and Karathane (840.6 gm/ha, 20% W) and That Big 8 Flowable Sulfur (2.52 l/ha, 64% flowable) were sprayed weekly for 7 weeks. Fruit yields (total weight) in the Tilt, Bayleton, and Sulfur plots were significantly greater ($P=0.01$) by 38, 40 and 41% respectively than the check plots. The Benlate and check plots had 2 to 5.6 times more sunburned fruit than the Tilt, Bayleton or Sulfur plots. Tilt gave highly significant control of foliar symptoms over all other treatments and Bayleton and Sulfur gave highly significant control over the check, Karathane, and Benlate plots. Benlate is noted for control of tomato mildew in other countries but our results suggest that the pathogen may be resistant in Utah.

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APPARENT MOVEMENT OF ALIETTE OR RIDOMIL IN *PERSEA INDICA* AND ITS EFFECT ON ROOT ROT. Donald E. Munnecke. Department of Plant Pathology, University of California, Riverside, CA 92521.

Persea indica was grown with roots divided into two pots and the effect of soil drenches of Ridomil (metalaxyl) or Aliette (fosetyl) on *Phytophthora cinnamomi* was studied. Direct effects were measured by applying fungicides to the pot of soil inoculated with the pathogen. Indirect effects were studied by applying fungicides to soil in the uninoculated pot and measuring the effects on roots and pathogen in the inoculated pot. When fungicides were applied directly to inoculated pots, plants usually were symptomless and propagules of *P. cinnamomi* were scarce. After indirect treatments, plant heights were as great as those of uninoculated controls. With Aliette treatments, roots in the inoculated pot sometimes were healthy, and numbers of *P. cinnamomi* were low. Applications made prior to inoculation with *P. cinnamomi* were as effective as combined applications made before as well as after inoculations. Extracts of roots treated with Aliette were inhibitory to *P. cinnamomi*.

EFFICACY OF SULFUR FOR CONTROL OF SUGARBEET DAMPING-OFF AND ROOT ROT CAUSED BY *RHIZOCTONIA SOLANI*. E. G. Ruppel and R. J. Hecker, U. S. Dept. of Agriculture, ARS, Crops Research Laboratory, Colorado State Univ., Fort Collins 80523.

In greenhouse studies, sulfur (92% WP) applied at 11, 22 and 44 kg/ha to soil did not control sugarbeet damping-off by *Rhizoctonia solani* (AG¹). Two flowable sulfurs (50 & 52% S) sprayed on soil (36 ml flowable/946 ml water) increased seedling survival only in nonautoclaved soil, indicating that their effect may be on another biotic system with indirect effects on *R. solani*. In field sites heavily infested with *R. solani* (AG²), two flowables broadcasted and incorporated at 34 kg S/ha significantly reduced root rot in mature beets in 1980; however, in a test of one flowable at three rates (7, 13 and 34 kg S/ha) and two methods of application (broadcast and banded) in 1981, no control was obtained. If the action of sulfur is on some other biotic system, conditions were not conducive for such a system to be operative in 1981.

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EFFICACY OF METALAXYL/MANCOZEB TANK MIXTURES FOR CONTROL OF POTATO LATE BLIGHT (*Phytophthora infestans*). L. D. Houseworth, J. Snow, and T. Young. Agricultural Division, CIBA-GEIGY Corporation, P. O. Box 18300, Greensboro, NC 27419.

Metalaxyl [N-(2,6-dimethylphenyl)-N-(methoxyacetyl) alanine methyl ester] has been evaluated for control of potato late blight in the United States since 1976. Because of potential for *Phytophthora infestans* to develop resistance to metalaxyl, a project was initiated in 1981 to evaluate the efficacy of combinations of low rates of metalaxyl plus mancozeb. Metalaxyl plus mancozeb mixtures were evaluated at 0.18 plus 0.79 kg ai/ha, and 0.22 plus 1.16 kg ai/ha on a 14-day protective schedule. At the same spray intervals, combination treatments provided better control than either fungicide applied alone at equal rates. Post infection activity was not adequate under high disease pressure.

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VAPAM[®] APPLICATION THROUGH A CENTER PIVOT IRRIGATION SYSTEM FOR CONTROL OF VERTICILLIUM WILT OF POTATO. Gene D. Easton and Michael E. Nagle. Washington State University, IAREC, Prosser, WA 99350.

In the fall of 1980, Vapam[®] (467.5 l/ha) was applied in 2.46 cm of water in a center pivot circle that had been cropped the previous two years to field corn and in 1.68 cm of water in a circle that had been cropped in 1979 to field corn and to potatoes in 1980. Treatments were replicated in 6-4 ha pie-shaped plots. DD-PI[®] (280.5 l/ha) and Telone C-17[®] (257.1 l/ha) were shanked into the soil in plots 16.5 m wide x 106.8 m long in the center of six untreated plots in the circle that had been cropped the previous year to potatoes. Vapam[®] neither decreased Verticillium wilt nor increased yield or tuber quality in the circle cropped previously two years to corn. Vapam[®] and both shank-applied fumigants reduced wilt and increased yield and tuber quality in the circle cropped the previous year to potatoes. Vapam[®] appears equal to shank-applied fumigants for control of Verticillium wilt.

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EVALUATION OF BENOMYL AND TRIAZOLE IN REDUCING AFLATOXIN B₁ CONCENTRATIONS IN FIELD CORN. R. K. Jones and J. R. Mulkey, Texas A&M University Agricultural Research and Extension Center, Uvalde, Texas 78801.

Field experiments were designed to evaluate the efficacy of systemic fungicides in reducing infection and subsequent aflatoxin production in corn (*Zea mays*) inoculated with *Aspergillus flavus*. Granular applications of a triazole derivative (CGA 64250 2.5G) were applied over the whorl and shanked into the soil 5 weeks after planting at a rate of 1.12 kg a.i./ha. Silk applications of the triazole derivative (CGA 64250 3.6 EC) at 0.50 kg a.i./ha and benomyl (Benlate 50W) at 1.12, 0.56 and 0.28 kg a.i./ha were applied with a ground sprayer at 90% silk emergence. Silks were atomized with 5X10³ spores/ml of *A. flavus* (NRRL-3357) one week after fungicide applications. Significant ($P=0.05$) reductions in percent infection and aflatoxin B₁ concentrations were observed in benomyl treatments. Triazole derivative spray and whorl applications significantly reduced aflatoxin B₁ concentrations; however, soil applications were ineffective.

PHYROXYFUR SEED TREATMENT FOR INTEGRATED CONTROL OF PHYTOPHTHORA ROOT ROT OF SOYBEAN. A. F. Schmitthenner and M. E. Kroetz. Departments of Plant Pathology and Agronomy, respectively, The Ohio State University, Columbus 43210.

Phytophthora megasperma f. sp. *glycinea* (Pmg) damping-off of soybean has become increasingly prevalent in Ohio on root rot tolerant cultivars resistant to race 1, but susceptible to the predominant races 3 and 7. Pyroxyfur, 2-chloro-6-(2-furanylmethoxy)-4-(trichloromethyl) pyridine, seed treatment at .63 g/kg of seed improved stands and yields of cv. Corsoy (no resistance, little tolerance), Beeson (race 1 resistance, slight tolerance) and Voris 295 (race 1 resistance, high tolerance), but not of Vickery (Corsoy with race 1-3 and 6-9 resistance). Yields of treated, highly tolerant Voris 295 were equal to multirace resistant Vickery but yields of lesser tolerant Corsoy and Beeson were 9.1 and 6.4 q/ha below. Seed treatment of highly tolerant soybean cultivars improved stands and yields (0.8 to 8.0 q/ha) in 29 fields with *Phytophthora* root rot. It was concluded that pyroxyfur seed treatment of highly tolerant cultivars is a good alternative to multirace resistance for control of Pmg root rot.

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CONTROL OF PYTHIUM DAMPING OFF AND PHYTOPHTHORA ROOT ROT OF SOYBEANS WITH METALAXYL. H. V. Morton, C. L. Kern, and T. D. Taylor. Agricultural Division, CIBA-GEIGY Corporation, P. O. Box 18300, Greensboro, NC 27419.

In the heavy soils of the Midwest, soybean yields are frequently limited by *Pythium* damping off (*Pythium* spp.) and *Phytophthora* root rot (*Phytophthora megasperma*, var. *sojae*). During 1980/81, several field trials were conducted in the upper Midwest to compare the efficacy of seed dressing and soil applications of metalaxyl. Metalaxyl was applied as a seed dressing at rates of 0.15 and 0.30 g ai/kg seed. Soil applications were made in furrow, and banded over the row at rates of 0.07 to 0.56 kg ai/ha. Metalaxyl seed treatments provided control of *Pythium* damping off and early season *Phytophthora* root rot. Under heavy disease pressure, 0.30 g ai/kg seed was superior to 0.15 g ai/kg seed. Soil applications were necessary for season long control of *Phytophthora* root rot. Yield increases averaged 67 kg/ha across 18 treatments. Further research with metalaxyl seed and soil treatments are in progress during 1982.

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Effect of formulation on leaf penetration and translocation in soybean of foliar applied Thiabendazole. E. J. Butterfield and R. J. Pochiari. Boyce Thompson Institute for Plant Research, Cornell University, Ithaca, New York 14853.

Changes in the formulation of Thiabendazole (TBZ) significantly alter the extent to which foliarly applied TBZ penetrates soybean leaves. Leaf penetration of TBZ, as determined by assessment of post-infection activity against *Colletotrichum lagenarium*, phytotoxicity to soybean leaves and monitoring of ^{14}C -TBZ, was greatest from acid solubilized formulations. Oil based flowables, water based flowables, and a wettable powder formulation gave progressively poorer penetration. Even with the solubilized formulations, more than 90% of the TBZ remained on the leaf surface. In soybean, most of the TBZ which penetrated the leaf remained in the same leaf with some acropetal movement. Less than 1% of the applied TBZ could be recovered from roots, stems, flowering parts and leaves other than the site of application. Plant age did not significantly alter the pattern of translocation.

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FUNGICIDES FOR PROTECTION AGAINST TOBACCO BLUE MOLD. P. B. Shoemaker, NCSU, Rt. 2, Box 249, Fletcher, NC 28732.

Twenty fungicides were compared as protectants against blue mold (*Peronospora tabacina*) on tobacco (*Nicotiana tabacum* 'Burley 21') in the greenhouse. Fungicides tested included 16 currently approved for Phycomycetes and 4 nonregistered materials. Each was tested at 1/2, 1 and 2X standard rates or rates suggested by the manufacturer. Treatments were applied to 6-wk-old plants with a compressed air sprayer the day before inoculation. Inoculum consisted of fresh conidia of *P. tabacina* adjusted to 5,000 conidia per ml and applied to 5 single plant replicates. Protection against blue mold was determined 10 days after inoculation. Materials providing complete protection at one or more rates included benalaxyl, mancozeb WP, mancozeb F, maneb, maneb plus zinc, polyram, chlorothalonil, tribasic copper sulfate, metalaxyl, difolatan, captan, folpet, ferbam, UC-55248 and zineb. Materials not giving complete protection at any rate tested included propamocarb, streptomycin sulfate, sodium hypochlorite, cymoxanil, Terrazole and the control.

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SYSTEMIC FUNGICIDES FOR CONTROLLING DAMPING-OFF OF COTTON. A. O. Paulus, J. Nelson and F. Shibuya. Department of Plant Pathology, University of California, Riverside, CA 92521.

Rhizoctonia solani Kuhn and *Pythium ultimum* Trow can cause reduced emergence and cotton stands in the interior valleys of southern California. Studies have evaluated combination and single seed treatments and row-furrow granular fungicides in greenhouse and field trials. Metalaxyl (N-2,6-dimethylphenyl-N-methoxyacetyl alanine methyl ester) gave excellent control of *Pythium ultimum* seed or root decay in numerous tests. Furmecyclox (N-cyclohexyl-N-methoxy-2,5-dimethyl-3-furan-carboxamide) was outstanding for control of *Rhizoctonia solani* and gave more healthy plants per replicate than any other treatment. A combination of metalaxyl + furmecyclox was effective for control of both fungi. Row-furrow granular applications of PCNB + Terrazole was effective for control of both fungi when disease conditions were severe for damping-off.

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FUNGICIDAL CONTROL OF A FUNGAL ENDOPHYTE IN SEED AND ESTABLISHED PLANTS OF TALL FESCUE. Mary J. Williams, P. A. Backman, and M. A. Crawford. Department of Botany, Plant Pathology and Microbiology, Auburn University, Alabama, 36849.

Cattle performance on the 14 x 10⁶ ha of tall fescue (*Festuca arundinacea*) in the U. S. has been poorer than expected; an unidentified fungal endophyte of fescue has been implicated in this 'summer syndrome' of cattle. Seed treatment fungicides were evaluated for the control of the endophyte; 80% of the seedlings established from infected seed had signs of the endophyte, while seed treated with triadimefon (0.3g ai/kg seed) had < 8% and triadimenol reduced incidence to < 50%. In mature, established, greenhouse plants averaging 91% infection, three foliar applications of triadimefon (1.1kg ai/ha), CGA-64250 (0.56kg ai/ha) or nuarimol (0.56kg ai/ha) at ten day intervals, reduced the incidence of the endophyte to 41, 65, and 47% respectively. If the same treatments were supplemented with an application of CGA-64250 granules (1.1kg ai/ha) at date 1, the incidence of endophyte was 0, 8, and 21%, respectively; plants treated with the granule alone were 68% infected.

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THE INFLUENCE OF CARBOXIN ON SEEDLING VIGOR AND GROWTH OF BARLEY PLANTS FROM HEALTHY AND *USTILAGO NUDA* INFECTED SEED. V.D. Pederson. Plant Pathology Department, North Dakota State Univ., Fargo, ND 58102

Increases in yield of barley from different seed lots containing various percentages of kernels infected with *Ustilago nuda* Jens. Rostr. treated with 5,6-dihydro-2-methyl-1,4-oxathiin-3-carboxanilide (carboxin) usually have not equalled expected yield increases despite 100% control of loose smut. However, yields from non-infected seed lots have not been adversely affected by seed treatment. The interaction between *U. nuda*-infected seed and carboxin was studied by making nondestructive determination of scutellar infection and measuring growth and yields of barley from treated and untreated seed. Smutted seed, whether treated or untreated, germinated slower than healthy seed. Plants from treated smutted seed grew slower, were smaller and had fewer tillers and heads than did plants from healthy seed. In the field, the per plant yield from treated smutted seed was significantly less than from healthy seed. Differences in yield from treated and untreated healthy seed were not significant.

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CONTROL OF COMMON ROOT ROT (*COCHLIOBOLUS SATIVUS*) IN SPRING WHEAT WITH SYSTEMIC SEED TREATMENT FUNGICIDES, P. R. Verma, Can. Agr. Res. Stn. Saskatoon, Saskatchewan, Canada S7N 0X2

The efficacy of baytan, imazalil and nuarimol for common root rot (*Cochliobolus sativus*) control in spring wheat was tested in 1979, 1980 and 1981 with similar results. Results in 1981 showed that baytan at 0.125, 0.175 and 0.250; imazalil at 0.100, 0.150 and 0.175; and nuarimol at 0.075, 0.100 and 0.125 g active ingredient/kg seed resulted in significant reduction in root rot severity in both Cypress and Neepawa wheats. With all three fungicides, a rate response was apparent in both cultivars at three test locations. The degree of disease control was better in Neepawa than in Cypress despite higher disease ratings in the latter. Nuarimol provided better control than baytan or imazalil. A significant disease control, however, was not reflected in the yield data. The overall yields of both cultivars were slightly reduced by all treatments. At the seedling stage, the mean dry weight per plant was slightly lower in fungicide-treated than in nontreated plants. Most treatments did not affect plant stands.

TRIADIMENOL SEED TREATMENT REDUCES FALL INFECTIONS OF WINTER WHEAT BY *GAEUMANNOMYCES GRAMINIS* VAR. *TRITICI*. W. W. Bockus, Department of Plant Pathology, Kansas State University, Manhattan, KS 66506.

Take-all root and crown rot of wheat is caused by the soil-borne fungus *Gaeumannomyces graminis* var. *tritici* (Ggt). Spring wheat or late-planted winter wheat sustains less take-all injury than early-planted winter wheat, indicating the importance of fall infections. The systemic seed treatment fungicide triadimenol reduced fall Ggt incidence 42% in artificially-inoculated field experiments. Yield loss due to take-all was reduced 61%. In a naturally-infested field with a moderate level (35% whiteheads) of Ggt, triadimenol increased yield 38% over the nontreated control. In the greenhouse, triadimenol protected wheat seedlings from Ggt for 8 wk. Thus, reduction in take-all severity with triadimenol seed treatment appears to be the result of protection of winter wheat plants from fall Ggt infections.

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CHEMICAL CONTROL OF STRIPE RUST AND LEAF RUST OF WHEAT IN THE PACIFIC NORTHWEST. Roland F. Line. ARS-USDA, WSU, Pullman, WA 99164

Epidemics of stripe rust (SR) and leaf rust have become more frequent in the Pacific Northwest (PNW) primarily because management practices now provide continuous hosts for the rusts, warmer winters since 1958 favored rust survival, and new, virulent races attacked previously resistant cultivars. Based on the changes; distribution, prevalence and pathogenicity of the rusts in the fall of 1980; and a predictive model for SR, severe epidemics of both rusts were predicted for 1981. Since 1973, Bayleton has controlled rust in local studies. Using data from the studies, guidelines for chemical control of the rusts based on host resistance and stage of growth and rust type and intensity were developed. Consequently, emergency registration of Bayleton was obtained for the PNW. More than 100,000 acres (40,000 ha) were sprayed in 1981 resulting in a production increase of 1,000,000 bu (27,000 metric tons). This is the first extensive use of a fungicide for control of wheat rust in the USA.

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MANAGING PLANT DISEASES WITH SUB-LETHAL DOSAGES OF SYSTEMIC FUNGICIDES. R. D. Schein and R. R. Nelson, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Managing plant diseases at levels below yield loss thresholds by rate-reducing resistance was prompted by the periodic demise of major, race-specific resistance genes designed to provide essentially complete control. The fact that recommended dosages of systemic fungicides, also designed to give nearly complete control, often prompt the occurrence of tolerant fungal populations persuaded us to determine whether plant diseases could be managed with sub-lethal dosages. Working with triadimefon (Bayleton) and powdery mildew and stem rust of wheat and blast of rice, we found that fungal isolates inoculated onto plants treated with sub-lethal dosages have greatly reduced disease efficiencies and sporulation capacities and induce smaller infection sites. Sub-lethal dosages of triadimefon mimic rate-reducing resistance and should reduce selection pressure towards tolerance much as is proposed for rate-reducing resistance, thus increasing the effective life of the compound.

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ABSORPTION, TRANSLOCATION AND CHEMICAL MODIFICATION OF FUNGAL MACROCYCLIC TRICHOECENES BY *BACCHARIS MEGAPOTOMICA*. G. A. Bean, Dept. of Botany and B. B. Jarvis, Dept. of Chemistry, Univ. of Maryland, College Park, MD 20742.

Roots of seedlings of *Baccharis megapotamica* approximately 15 cm tall were immersed in a 10^{-5} M CaSO_4 solution containing roridin A and verrucarins A. After 1, 2, and 3 day intervals, the roots and top portions of the seedlings were separated, washed and analyzed. No toxins were detected in the root tissue at any time. After 24 hours, roridin A, verrucarins A and their hydroxy forms were found in the leaves. Although the *Baccharis* plants had no phytotoxic symptoms, seedlings of tomatoes and peppers were dead after 3 days even though roridin A had been absorbed and translocated. Since the hydroxy form of roridin and verrucarins is normally found in *B. megapotamica* under field conditions, these studies indicate that the origin of these compounds is probably fungi present in the soil.

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TRICHOECENES PRODUCED BY *Fusarium roseum* (Alaska 2-2) GROWN ON SOLID RICE SUBSTRATE. Yin-Won Lee and C.J. Mirocha, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

An isolate of *Fusarium roseum* obtained from overwintered barley in Alaska, was grown on 300g autoclaved rice for 2 weeks at 21-24°C, followed by 2 weeks at 10°C. The cultures were extracted with methanol-water (1:1, v/v), concentrated in vacuo and partitioned with petroleum ether. The aqueous phase was applied to XAD-2 column, washed with water and eluted with chloroform-methanol (3:1, v/v). The XAD-2 eluate was concentrated, applied to a Florosil column and eluted with chloroform-methanol (3:1, v/v). Two major and one minor trichoecenes were separated and purified on TLC. The NMR, GC-MS, and hydrolysis data identified diacetoxyscirpenol (DAS) and 7-hydroxy-diacetoxyscirpenol (7-OH-DAS); the third minor trichoecene remains unidentified. DAS and 7-OH-DAS are responsible for the mouth lesions in chicks and also the low hatchability of fertile eggs caused by feeding cultures of this fungus. This culture also causes tibial dyschondroplasia in chickens but the toxin involved is not a trichoecene.

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A TOXIC METABOLITE OF *Fusarium roseum* (Alaska 2-2) WHICH REDUCES HATCHABILITY AND CAUSES TIBIAL DYSCHONDROPLASIA IN POULTRY. Yin-Won Lee, C.J. Mirocha, Department of Plant Pathology, and N.K. Allen, Department of Animal Science, University of Minnesota, St. Paul, MN 55108.

A rice culture of *Fusarium roseum* (Alaska 2-2) caused tibial dyschondroplasia (TDP) and reduced hatchability of chicken and turkey eggs. The toxic metabolites were not extractable with various organic solvents but were soluble in water. The concentrate was applied to an XAD-2 column, and eluted with methanol. The methanol eluate was concentrated, applied to a Silica gel column and eluted with ethylacetate-methanol (5:1, v/v), followed by chloroform-methanol (3:1, v/v), and finally methanol. Each fraction was dissolved in dimethylsulfoxide (DMSO) and their toxicity measured in a chick embryo test. The components causing death were found in the chloroform-methanol (3:1, v/v) fraction; this fraction also caused tibial dyschondroplasia in feeding tests. The active component is water soluble and is fluorescent under ultraviolet.

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DESTRUCTION OF AFLATOXIN WITH BISULFITE: ISOLATION OF THE REACTION PRODUCT AFLATOXIN B₁S. W. M. Hagler, Jr., and P. B. Hamilton, Dept. of Poultry Science, N.C. State U., Raleigh, NC 27650

Reaction of aflatoxin B₁ with sodium bisulfite yielded a water-soluble addition product, aflatoxin B₁S, in about a 98% yield. The product was isolated by reverse-phase high-pressure liquid chromatography and partially characterized. B₁S was identical to B₁ in color of fluorescence under 365 nm ultraviolet (UV) light on thin-layer chromatography. Spectral characteristics of B₁S were similar to those of other aflatoxins (B₁, B₂, G₁, and G₂). UV spectrum of B₁S had maxima identical to those of B₁. Infrared spectrum of B₁S showed loss of the furofuran double bond, an intact cyclopentenone system and maxima consistent with a sulfonyl moiety. Nuclear magnetic resonance spectra also indicated an addition across the furofuran double bond as well as thirteen protons in the molecule. Aflatoxin G₁ was also susceptible to bisulfite while aflatoxins B₂ and G₂ did not react. B₁S is apparently a sulfonate derivative of B₁. Ease of reaction suggests its candidacy for detoxification purposes.

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EFFECT OF WATER STRESS ON INFECTION AND AFLATOXIN PRODUCTION BY *ASPERGILLUS FLAVUS* IN CORN. G. A. Payne, D. L. Thompson, and E. B. Lillehoj. Depts. Plant Pathology, Crop Science, N. C. State Univ., Raleigh, NC 27650; and SRRC, USDA-ARS, New Orleans, LA.

Corn plants grown under day/night temperatures of 34/30°C were water stressed by withholding water until wilting. Leaf water potential was measured at 0830 each day with a thermocouple psychrometer. Plants were stressed beginning 14 or 19 days after silking and leaf water potential before rewatering was -7.2 bars (unstressed = -5.8 bars) and -10.2 bars (unstressed = -7.6 bars) respectively. Plants were inoculated with *A. flavus* by spraying spores on silks or wounded kernels 19 days after silking while kernels were in the dough stage. Aflatoxin B₁ levels in wound inoculated kernels stressed before inoculation (41,397 ug/kg) were higher than in unstressed kernels (26,507 ug/kg). Aflatoxin B₁ levels in kernels stressed after inoculation (33,600 ug/kg) were not significantly different from levels in unstressed kernels or kernels stressed before inoculation. Water stress had no effect on the % infected kernels in silk inoculated ears.

STORAGE MOLD PREVALENCE IN MINNESOTA FARM STORED CORN AND SOYBEANS. L.L. Castor, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Farm storage structures were surveyed in seven central and south central Minnesota counties. Fifty shelled-corn storage structures, containing 13333t (525,000 bu), and 27 soybean storage structures, containing 4762t (175,000 bu), were sampled using a 1.8 m grain probe in December 1981. Data were taken on grain temperature, moisture content, visible mold, germination, fungal incidence, and grading characteristics. Four percent of the corn (in 24% of the structures) and 0.4% of the soybeans (in 15% of the structures) were visibly molded. Moisture contents of corn and soybeans were above those considered safe for long term storage in 46% of the corn and 85% of the soybean storage structures. The predominant storage fungi identified from sample sterilized corn and soybeans were in the Aspergillus glaucus and Aspergillus flavus groups. This survey, which continues, indicated that storage conditions on many farms were inadequate to maintain optimum corn and soybean quality on a long term basis.

OCCURRENCE OF VOMITOXIN IN SWINE FEED. Ronald F. Vesonder, Northern Regional Research Center, ARS, USDA, Peoria, IL 61604.

Specimens of feed obtained from herd episodes of swine disorders, i.e., refusal to eat, vomiting, abortion, hyperestrogenism, and bloody stools, were investigated from 1972-1981 for causative principles. In many of these samples, 3,7,15-trihydroxy-12,13-trichothec-9-ene-8-one (vomitoxin, 4-deoxynivalenol) was detected in substantial quantities (2-40 µg/g). From January through July 1980, midwest swine operations were confronted with lack of growth of feeder pigs and reproductive problems with sows. Many of the 57 feed samples representative of the feed batches used at these pig co-ops for lactation/farrowing and finishing during this 6-month period were found to contain vomitoxin and zearalenone. The highest level of vomitoxin detected was 40 µg/g. Microbiological examination of these 57 feed samples indicated a high number of propagules of Fusaria in vomitoxin-positive feed samples.

SEROLOGICAL REACTIVITIES OF ISOLATES OF EPICHLÖE TYPHINA. Mark C. Johnson, USDA, AR, Dept. of Plant Pathology, University of Kentucky, Lexington, KY 40546

The reactivity of an antiserum prepared to mycelial homogenates of a Kentucky tall fescue isolate of the endophytic fungus, Epichloe typhina (= Acremonium coenophialum), was tested with seven other isolates of the fungus. The fungi were grown in liquid medium for 10-13 weeks, washed, ground, and freeze dried. Ten-fold dilutions of mycelial extracts ranging from 10 mg/ml to 1 µg/ml were tested by ELISA. Extracts of tall fescue isolates, from Alabama, Georgia, and Tennessee, as well as an isolate of Epichloe typhina from bent grass, produced absorbances comparable to the Kentucky isolate. However, one Alabama tall fescue isolate and a wedgegrass isolate were found to be at least 1,000X less reactive than the Kentucky tall fescue isolate. The possibility of differences in serological reactivity of E. typhina isolates should be considered when using ELISA in certifying tall fescue seed for endophyte content.

STRUCTURAL CHARACTERIZATION OF THE ALTERNARIA CITRI TOXINS HOST-SPECIFIC FOR ROUGH LEMON. John M. Gardner*, James L. Templeton and Roy W. King. *AREC, University of Florida, Lake Alfred, FL 33850 and University of Florida, Gainesville, FL 32611.

Alternaria citri isolates produce at least three closely related host-specific toxins which were separated and purified by high pressure liquid chromatography and TLC. Coincident with a loss in activity, the isotoxins (active at 0.02 µg/ml) partially convert to a relatively nonpolar and weakly toxic (active at > 2 µg/ml) yet host-specific compound which is structurally related to each toxin. Toxin structure has been partially elucidated by GLC-mass spectrometry, NMR and IR. The basic structural component of the isotoxins is a highly saturated hydrocarbon having a molecular weight of 292 and is characterized by derivatizable ketone and alcohol groups. It appears to be structurally unrelated to other previously characterized Alternaria toxins.

FUNGICIDE SUPPRESSION OF CORN HEAD SMUT. W.C. Stienstra, Erik Stromberg, Thor Kommedahl and Carol E. Windels, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Head smut (Spacelotheca reiliana) of corn, reported in four counties in Minnesota in 1980, has been reported to be controlled by chemical seed treatment. Susceptible corn hybrids, planted in artificially infested soil at three planting dates in 1981 (28 April, 12 and 27 May), were treated with four fungicides either as seed treatments (ST), or granules (G) in furrow or surface band, or as a foliar spray. The following chemicals eliminated or reduced smut incidence: Baytan (ST), Ciba Geigy 88531 (ST) and CG 64250 (G). Chemical seed treatment with vitavax generally did not reduce the incidence of head smut. The experimental seed treatments (Baytan and CG 88531) and granular in-furrow or surface-band applications (CG 64250) could become part of a head smut management system.

HISTOLOGICAL DEVELOPMENT OF SORI AND SYMPTOM VARIATION OF Sphacelotheca reiliana ON Zea mays. C. A. Matyac and T. Kommedahl, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Sori of head smut of corn on ears and tassels develop over a system of intercellular hyphae with haustoria, which occur in nodal tissue but not in adjacent parenchyma. The hyphae grow abundantly between columns of highly vascularized host tissue and as the sorus matures, round up to form teliospores. A young sorus has a peridium consisting of a dense layer of hyphae beneath a 2-celled layer of host tissue. When the ears and tassels emerge the sorus ruptures, exposing a mass of teliospores and host vascular elements. The ears or tassels may be totally or partially replaced, depending on the distribution of hyphae in the primordial tissue. Besides such sori, there may be phyllody (<1%) in tassels and ears, stunting, and multiple ears at nodes. Of leaves resulting from phyllody, 28% contained spores. Sori are found infrequently on normal vegetative leaves.

FUSARIUM ROSEUM 'GRAMINEARUM' AS A LATE-SEASON STALK ROT PATHOGEN OF CORN. Thor Kommedahl and C.E. Windels, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Corn stalks have been sampled for Fusarium spp. during October and November (1973-81) in southern Minnesota, and stalk rot incidence seldom averaged more than 10%. Green stalks were sampled using a bark-increment hammer whereas rotted stalks were sampled by tweezer-extraction of infected pith. From 1973 through 1980, up to five Fusarium spp. were isolated from green (symptomless) stalks by season's end. In this period, Graminearum occurred in 1 to 24% of stalks, varying with field and year, and averaged 14%. However, in 1981, 50% of green stalks yielded Graminearum at near-harvest time; 90% of fallen, rotted stalks and 81% of standing, rotted stalks yielded Graminearum. Earlier infections with other Fusarium spp. caused little or no damage based on observations and field inoculations. Stalk rot is caused by Graminearum late in the season as the stalks are ripening.

CONDITIONS INFLUENCING GROWTH, SPORULATION, AND LESION DEVELOPMENT OF CERCOSPORA ZEAE-MAYDIS. P. M. Beckman and G. A. Payne, Department of Plant Pathology, N. C. State University, Raleigh 27650.

Sporulating cultures of Cercospora zeae-maydis were obtained by homogenizing a culture (either freshly isolated or from storage) in water, dispensing the homogenate on V-8 juice agar, and incubating the plates under a diurnal light (fluorescent) regime for 13 days. Sporulation was also good on decoction media made from green or senescent corn leaves but was poor on potato dextrose agar. Constant light inhibited spore germination, germ tube growth, and sporulation but stimulated conidiophore production. Optimum temperature range for germination and germ tube growth was 22-30°C. Cultures of the fungus could be stored successfully at 4°C for at least 23 months on V-8 juice or decoction media slants. Lesion development on corn plants was obtained by misting inoculated plants for 3 sec every 4 min between 8 PM and 10 AM for two weeks. Lesion development was greatest during June-August in a greenhouse kept at 22-28°C by wet pad cooling.

FUNGAL SPECIES ISOLATED FROM ROOTS AND STALKS OF SYMPTOMLESS SORGHUM PLANTS DURING THE GROWING SEASON. J.E. Reed, J.E. Partridge, & P.T. Nordquist, Dept. of Plant Pathology and Dept. of Agronomy, University of Nebraska, Lincoln, NE 68583-0722.

Stalks and roots of field-grown grain sorghum [*Sorghum bicolor* (L.) Moench] were examined for fungal colonization throughout the 1980 and 1981 growing seasons. Fungi were found to colonize apparently healthy tissue as early as 3 weeks after planting. They became increasingly abundant after anthesis, during the period of grain development. Of the several species recovered from stalks and roots, *Fusarium* species were found to occur most frequently. *Fusarium moniliforme* appeared to predominate in stalk tissue, whereas in roots, no single species was observed to predominate. Evidence from this study indicates that fungal colonization of sorghum stalk and root tissue occurs routinely, in the absence of any symptoms of stalk rot. Therefore, the critical factors in the development of this disease appear to be related to the interaction between fungal activity and the physiological condition of the plant, rather than simply to the presence of fungi within plant tissue.

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YIELD LOSSES ASSOCIATED WITH SEVERITY OF BEAN RUST (*UROMYCES PHASEOLI*) ON PINTO BEANS (*PHASEOLUS VULGARIS* UI-114). J. R. Venette and D. A. Jones. Plant Pathology Department, North Dakota State University, Fargo, ND 58105.

Results from five experiments indicated yield losses of harvested dry beans were directly related to the logarithm of severity of bean rust. Yield was calculated from hand harvested beans taken from over 200 plots grown under North Dakota dryland conditions in 1980 and 1981. Rust severity ranged between 0.7 and 31.6 pustules/sq cm. Yield losses ranged from 336 to 1233 kg/ha and represented potential losses of 13 to 54%. Experimentally derived losses compared favorably with yield loss estimated by county agents, consultants, and others closely associated with bean production.

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EFFECTS OF FOLIAR FUNGICIDE AND FERTILIZER TREATMENTS ON SEED QUALITY AND YIELD RESPONSE IN SOYBEANS. F.E. Wright and R.D. Gipson. P.O. Box 549 State University, AR 72467

Fifteen treatments of various foliar fungicides and fertilizer materials were applied to the soybean cultivar, 'Forrest', at three different growth stages, R3, R5 and R7. A significant improvement in seed quality was found for several of the treatments over the control. Eight different pathogenic fungi were isolated from soybean seed obtained from plants in the study. Numbers of seed-borne pathogens were significantly reduced with several of the foliar fungicide treatments compared to the control. Yields were significantly increased in the fungicide treated plots over the control. No significant reduction in seed-borne pathogens or increase in yield was found for the plots treated more than twice.

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PREEMPTIVE FUNGAL INFECTION OF SOYBEAN SEED. J. P. Ross, Plant Pathology Department, N. C. State University, Raleigh 27650.

During a current program of breeding for resistance to *Phomopsis* seed rot of soybean, relationships have appeared among fungi commonly found in soybean seed after exposure to conditions favoring seed infection. Some lines with low percentage of seed infected with *Phomopsis* (resistant) had high percentages of seed infected with *Alternaria*, *Fusarium*, or *Cercospora*. Isolations on acidified PDA from fragmented testas of surface disinfested seed grown in Fla. and N.C. usually yielded only one fungus per seed; this indicates that infection by one fungus preempts infection by other fungi. Although antagonism by *C. kikuchii* toward other seed infecting fungi has been recorded, the general effectiveness of preëemptor fungi in soybean seed has not been recognized. In breeding programs selecting for resistance to *Phomopsis*, consideration should be given to whether the lines are either more susceptible to seed-invading fungi which prevent *Phomopsis* infection or are genetically resistant to *Phomopsis*. Under optimum conditions for seed infections, lines resistant to *Phomopsis* may be invaded by other fungi.

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EFFECTS OF BEAN POD MOTTLE VIRUS INFECTION ON SOYBEANS. M. T. Windham and J. P. Ross, Department of Plant Pathology, N. C. State University, Raleigh, NC 27650.

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The objectives of this study were to determine the effects of bean pod mottle virus (BPMV) on soybeans and how they relate to yield reduction. Three soybean cultivars each planted in 8 sets of 3-row paired plots were mechanically inoculated with BPMV at the V-2 to V-3 stage. Sevin was applied twice weekly to control vectors. Chlorotic leaf area (CLA) (top 4 nodes of 8 plants), plant height, and canopy were measured. CLA was greatest within 1 wk prior to flowering and could be used to identify cultivars as tolerant, moderately tolerant, and intolerant. Virus infection reduced plant height and canopy and caused yield reductions for Centennial, Ransom, and Davis of 9, 7, and 2% respectively, compared with noninoculated plants. The correlation between CLA and yield reduction occurred at various times but was greatest 1 wk prior to flowering ($r=0.999$). Reductions in plant height and canopy were also correlated with yield loss ($r=0.999$ and 0.890, respectively).

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A NEW BIOTYPE OF *PERONOSPORA MANCHURICA* IN SOYBEAN DISEASE MONITORING PLOTS IN ILLINOIS. S. M. Lim, R. L. Bernard, C. D. Nickell, and L. E. Gray, USDA-ARS, Department of Plant Pathology and Department of Agronomy, University of Illinois, Urbana, 61801.

A soybean-monitoring program was established in 1977 to determine the incidence and severity of foliar diseases, detect new races of current pathogens, and identify areas of high disease risk. Over the five-year period, incidence and severity of foliar diseases differed by location, year and cultivar. At all the locations throughout the growing season, one of the prevalent foliar diseases was downy mildew caused by *Peronospora manchurica*. Prior to 1981, downy mildew developed on all the soybean cultivars in the monitoring plots except the resistant cultivar Union which carries the gene *Rpm*. In 1981, however, downy mildew lesions on the leaves of Union plants were first observed at two southern locations in Illinois in early August. By early September, downy mildew was observed on Union plants at all locations.

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INFECTION OF SOYBEAN BY *CERCOSPORA KIKUCHII* AS AFFECTED BY DEW TEMPERATURE AND DURATION OF DEW PERIODS. Keith F. Martin and H. J. Walters, Dept. of Plant Pathology, University of Arkansas, Fayetteville, AR 72701.

Soybean seedlings were inoculated with *Cercospora kikuchii* and placed in a dew chamber for 24, 48, or 72 hr at temperatures ranging from 16 to 36 C. Amount of infection was determined from disease ratings made 12 days after inoculation. Maximum infection occurred from 20 to 24 C. Infection increased as the length of the dew period increased. A cycle of 12 hr darkness followed by 12 hrs light in the dew chamber resulted in significantly more infection than continuous darkness. A dew period of at least 8 hr was required to establish a significant amount of infection when inoculated plants were exposed to 1, 2 or 3 dew periods (24 C) ranging from 0 to 24 hr with one dew period every 24 hr. The greatest amount of infection was obtained with 24-hr dew periods. Disease ratings increased with an increase in exposure to a given dew period. These results establish procedures to screen for resistance to *C. kikuchii*.

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INCREASE OF *SEPTORIA NODORUM* INOCULUM DURING WINTER. Barry M. Cunfer, Dept. of Plant Pathology, University of Georgia, Georgia Station, Experiment, GA 30212.

Latent infection by *Septoria nodorum* was monitored during the winter by plating the lower leaves of wheat and barley on a medium containing fentin hydroxide, chloramphenicol, and paraquat. The number of infection foci was determined by observing sporulating pycnidia in the leaves on the agar plates. Foci were very few in December, slightly more in January, then significantly greater at the beginning of March in three experiments. In a study in which the inoculum source was seed, the number of infection foci in leaves in March was proportional to the level of seed infection. Infection foci on barley leaves increased from 0.05 per leaf in December to 2.7 per leaf in March. Foci ranged from 0.3 to 2.7 per leaf among eight wheat cultivars on March 3. Cultivars with the most foci in March generally were those rated most susceptible near maturity. Inoculum buildup of *S. nodorum* during winter can be significant in the Southeast.

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INFLUENCE OF SEED SOURCE ON PERFORMANCE OF SPRING WHEAT SEED TREATMENTS. H. W. Johnston and H. G. Nass, Agriculture Canada,

Research Station, P. O. Box 1210, Charlottetown, Prince Edward Island, Canada C1A 7M8.

The influence of 5 fungicide seed treatments on performance of spring wheat, cv. Opal, produced in Eastern and Western Canada, and differing in seed size, germination, vigor and level of contaminating fungi was evaluated in a field trial. The seeding rate of the Eastern seed was increased to compensate for its lower germination level. Growth characteristics as measured by emergence and vigor were superior for the Western seed but there was no difference between sources for seeds/head, head wt., and 1000 kwt. Tillering rates increased in those treatments where emergence was low. Overall yield was highest for Western seed but neither fungicide treatment nor the fungicide treatment x source interaction were significant. Yield correlations were highest with vigor ratings taken at Growth Stage 9 (Feekes).

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THE EFFECTS OF ATRAZINE ON YIELD COMPONENTS AND DISEASES OF WINTER WHEAT. G. R. Watson, H. Cole, Jr., and J. A. Frank, Dept. of Plant Pathology and USDA-ARS, Center for Cereals Research, The Pennsylvania State University, University Park, PA 16802.

The effects of triazine herbicides on diseases and yield components of winter wheat were studied in field experiments at Rock Springs, PA in 1981 and 1982. Wheat cultivars currently recommended for use in Pennsylvania were planted into soil previously treated with various rates of the corn herbicide, atrazine, as a preplant incorporated treatment. Several atrazine rates were used, including phytotoxic and nonphytotoxic levels (2.197 and 0.02197 kg/ha, respectively). Wheat disease assessments were made for Septoria Leaf and Glume Blotch, Powdery Mildew, and a root rot complex. There was no detectable effect of atrazine treatments on Septoria Leaf and Glume Blotch or root rot severity. However, atrazine treatments increased severity of Powdery Mildew. Certain yield components were increased with nonphytotoxic levels of atrazine indicating possible stimulatory effects of the herbicide.

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SEED-BORNE INCIDENCE OF SEPTORIA NODORUM AND THE EFFECTS OF ENVIRONMENTAL CONDITIONS ON WHEAT SEEDLING INFECTION. M. Babadoost and T. T. Hebert, Dept. of Plant Pathology, NC State Univ., Raleigh, NC 27650

Septoria nodorum (Berk.) Berk., the causal organism of glume blotch of wheat, survived and remained pathogenic in stored seed over 2 years. Seed infection did not affect germination, but did produce seedlings with infected coleoptiles and significantly reduced growth. In growth chambers seedlings became infected at temperatures of 10, 15, and 20°C. There was no significant difference in seedling infection at planting depth of 2.5, 5, and 7.5 cm. High soil moisture significantly reduced seedling disease. Numerous pycnidia were produced on infected coleoptiles 3-4 weeks after planting that could serve as a source of inoculum for plant foliage during favorable environmental conditions. Results of 2 years of field experiments showed that even when foliage disease was not severe, significant yield reduction, up to 31%, of the plants grown from infected seeds occurred.

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THE EFFECTS OF INOCULATION OF WHEAT PLANTS AT DIFFERENT GROWTH STAGES WITH SEPTORIA NODORUM ON DISEASE SEVERITY AND YIELD. M. Babadoost and T. T. Hebert, Dept. of Plant Pathology, NC State University, Raleigh, NC 27650

The objective of this study was to assess yield loss due to glume blotch disease of wheat caused by *Septoria nodorum* (Berk.) Berk. Inoculum consisting of fungal spores and mycelium was prepared by homogenizing cultures of 10 isolates of *S. nodorum* grown on potato Dextrose Agar (PDA). Plants of cultivar 'Coker 747' were inoculated one to six times at the growth stages of 9-11.2 on the Feekes' scale. Check plants in each treatment were sprayed with a mixture of water and PDA. Plants inoculated at growth stages of 9 to 10.5 became severely diseased; disease severity on these plants was significantly higher than on plants inoculated at later growth stages or on check plants. Yields of severely affected plants were reduced significantly, up to 35%. Moreover, early infection of plants resulted in higher percentages of seed infection.

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FALL INFECTIONS OF WINTER WHEAT BY *ERYSIPHE GRAMINIS* F. SP. TRITICI AS INFLUENCED BY PLANTING DATE. J. A. Frank and H. Cole, Jr. USDA-ARS and Dept. of Plant Pathology, Center for Cereals Research, The Pennsylvania State University, University Park, PA.

The soft red winter wheat cultivar 'Hart' was planted on four

different planting dates in field plots at Rock Springs, PA. The dates ranged from one week prior to the recommended planting date to five weeks after that initial planting. Each wheat planting was separated by a 30.5 m wide planting of barley to reduce interplot interference. Three weeks after the final planting, 100 plants from each plot were transplanted into flats, placed into a mist chamber, and evaluated for mildew severity after three days. There was a significant decrease in mildew severity between the first and second planting dates, with reductions of greater than 60% in the second planting. This same reduction was evident between the second and third dates, although severities were much lower. The most mature leaves had the greatest mildew severities for each planting date.

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EFFECTS OF WINTER WHEAT MANAGEMENT PRACTICES ON POWDERY MILDEW AND SEPTORIA LEAF BLOTCH SEVERITY. S. C. Broschous, J. A. Frank, H. G. Marshall. Department of Plant Pathology and USDA-ARS, Center for Cereals Research, The Pennsylvania State University, University Park, Pennsylvania 16802.

To determine the influence of wheat management practices on disease severity, the variety 'Roland' was grown in field plots in central and southeastern Pa. Seeding depth, seeding rate, row spacing, and spring nitrogen fertilization were evaluated in a factorial arrangement. Severity of infection by *Erysiphe graminis* and *Septoria* spp. was assessed as the percent leaf area affected by each pathogen. Severities ranged from 2-13 and 3-40 percent for powdery mildew and Septoria leaf blotch, respectively. Increased levels of nitrogen resulted in significantly higher severity of both diseases. However, significant interactions between nitrogen and the other factors were dependent on location. Yields generally decreased as nitrogen and row spacing increased. Nitrogen level had the greatest influence on disease severity but the effect was modified by the other crop management practices and the environment.

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STRIPE RUST OCCURRENCE AND CONTROL IN BOLIVIA. Vidal Velasco Rivas and William M. Brown, Jr., Min. of Agric., Cochabamba, Bolivia, and Dept. of Botany and Plant Pathology, C.S.U., Fort Collins, CO 80523.

In 1978 a rust disease of barley was noted for the first time in the Cochabamba Valley of Bolivia. Subsequent identification of the fungus showed it to be race 24 of *Puccinia striiformis* West. Race 24 had not been reported in South America until it was introduced into Colombia in 1974. The fungus then spread to Ecuador and in 1977 was reported in Peru. Initial field trials showed that the fungus could be effectively controlled with two applications of Bayleton (triadimefon) at 500g/ha when applied at first sign of the disease and again 20 days later. This control was possible with manually operated backpack sprayers common to subsistence farming techniques prevalent in the area.

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HOST RANGE AND PATHOGENICITY OF STEM-(RHIZOCTONIA CEREALIS) AND ROOT-ATTACKING (R. SOLANI) ISOLATES IN ARKANSAS. Musa A. Abdel-shife, J.L. Dale, and J.P. Jones, Dept. of Plant Pathology, University of Arkansas, Fayetteville, AR 72701.

Binucleate (BNR) *Rhizoctonia* was isolated from sharp eyespot lesions and multinucleate (MNR) isolates were obtained from roots of cereals. The BNR isolates had optimum growth on PDA at 20-23 C, characteristics typical of *R. cerealis*, and anastomosis occurred between all isolates and with the binucleate tester CAG-1 (*R. cerealis*). Optimal growth of MNR isolates was at 24-28 C; they had *R. solani* characteristics and belonged to anastomosis group AG-4. Pathogenicity tests on seedlings in chambers at 16 and 24 C indicated *R. cerealis* caused pre- and post-emergence damping off of cereals at 16 C, but more sharp eyespot lesions at 24 C. *R. cerealis* was also pathogenic on cotton and soybeans. *R. solani* was not active at 16 C, but was pathogenic on various cereals, cotton, and soybeans causing seed decay, root rot, and death at 24 C. This confirms that *R. cerealis* is highly pathogenic on cereals at lower temperatures, while *R. solani* isolates are most pathogenic at higher temperatures.

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A Gaeumannomyces-like organism associated with diseased bluegrass in Wisconsin. G. L. Worf, R. C. Avenius, and J. S. Stewart, Dept. of Plant Pathology, Univ. of Wisconsin, Madison, WI. 53706

Much of Wisconsin's recently established bluegrass turf is affected by a serious disease of uncertain causation. Chlorotic

or tan rings resembling Fusarium blight frequently occur. However, circular or irregular patches 9 to 45 cm in length or width without living centers are common. Symptoms are sometimes more diffuse, with dead and weak plants interspersed among apparently healthy ones. Symptoms may occur in seeded lawns or golf fairways, but are often dramatically associated with individual rolls of sod laid that same season or a year or two previously. A fungus, was first isolated from crowns of oats and wheat planted into infested soil, and subsequently from diseased bluegrass crowns and roots. Preliminary inoculations indicate it is pathogenic to bluegrass. In vitro growth characteristics and vegetative hyphal comparisons resemble *Gaeumannomyces*, but perithecia were not observed in the field or in the laboratory in vitro cultures.

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OCCURRENCE OF OPHIOBOLUS PATCH OF TURF IN COLORADO. Wong, P.T.W., Cindy Rasmussen-Dykes, L.E. Perotti and W. M. Brown, Jr. Agric. Res. Cen., Tamworth, N.S.W. 2340 Australia, and Dept. of Botany & Plant Path., Colorado State University, Fort Collins, CO 80523.

One hundred thirty two samples of turf showing various disease symptoms were collected in Colorado and examined at C.S.U. in 1981. Only one of these samples was infected by *Gaeumannomyces graminis*, the causal agent of the Ophiobolus patch disease. Other isolations from Colorado turf yielded the fungus *Phialophora graminicola*. Additional samples collected from locations outside Colorado were also found in some instances to contain *P. graminicola*. While Ophiobolus patch is a common and serious problem in Europe and the Pacific Northwest it had not previously been encountered in Colorado. It is possible that the lack of Ophiobolus patch as a problem in Colorado turf may be in part due to natural suppression of the pathogen by indigenous *Phialophora* spp.. Studies to determine the role of naturally occurring *Phialophora* spp. in Colorado turf have been initiated.

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CHARACTERIZATION AND PATHOGENICITY OF RHIZOCTONIA SPP. ASSOCIATED WITH BROWN PATCH OF ST. AUGUSTINEGRASS. B. Hurd and M. P. Grisham. Dept. of Plant Sciences, Texas A&M Univ., College Station, TX 77843.

Rhizoctonia spp. isolated from St. Augustinegrass displaying brown patch symptoms were either binucleate (BNR) or multinucleate (MNR). MNR isolates fit the description of *R. solani* AG 2. MNR isolates produced brown patch symptoms on stolon sections of Texas Common St. Augustinegrass at 24 C in laboratory testing. Representative MNR isolates induced brown patch following field inoculations. At 24 C, MNR isolates also caused postemergence damping-off in two cultivars of *Festuca arundinacea* and pre- and postemergence damping-off in five cultivars of *Lolium perenne*, but not in two cultivars of *Poa pratensis*, one cultivar of *Agrostis palustris*, or one cultivar of *Poa trivialis*. Under similar conditions, BNR isolates were not pathogenic on any of the turfgrass tested. Symptom severity and rate of disease development varied among four cultivars and 11 selections of St. Augustinegrass inoculated with representative MNR isolates.

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A PRELIMINARY INFECTION MODEL FOR ANTHRACNOSE ON POA ANNUA. T. K. Danneberger and J. M. Vargas, Jr., Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824.

Anthracnose caused by *Colletotrichum graminicola* can cause severe damage to *Poa annua* (annual bluegrass) turfs under certain environmental conditions. These conditions were determined in order to enable development of a prediction model for anthracnose infection. Disease ratings and the number of acervuli per leaf were made on a 3-5 day basis at 2 locations. The environmental parameters measured were air temperature, relative humidity, leaf wetness, rainfall and soil temperature. The average air temperatures and leaf wetness were analysed in relation to increased disease severity. A model was developed from field data and supportive greenhouse experiments which related leaf wetness and air temperature to infection severity.

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INTEGRATED CONTROL OF PYTHIUM BLIGHT ON TURF USING METALAXYL AND TRICHODERMA HAMATUM. Cindy Rasmussen-Dykes and William M. Brown, Jr., Dept. Botany & Plant Pathol., Colorado State U., Fort Collins, Colorado 80523.

Pythium aphanidermatum and *P. ultimum* were isolated from a golf

green exhibiting Pythium blight. Studies were conducted using metalaxyl and *Trichoderma hamatum*, an antagonist to *Pythium* spp. (Chet, I., G. E. Harman, and R. Baker. 1981 "Trichoderma hamatum: Its Hyphal Interactions with *Rhizoctonia solani* and *Pythium* spp." Microb. Ecol. 7:29-38) to control the disease. In vitro studies indicate that *Pythium* spp. can be controlled with metalaxyl at lower concentrations than recommended. Three of the four *Pythium* isolates were effectively parasitized by *T. hamatum* (isolate obtained from Ralph Baker, CSU) in vitro. The same concentrations of metalaxyl tested against *Pythium* were used in in vitro tests against *T. hamatum*. The mycelial growth of *T. hamatum* was affected at high but only slightly at the low concentrations of metalaxyl. Further studies are presently underway to determine the combined effectiveness of metalaxyl and *T. hamatum* in controlling Pythium blight.

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RESISTANCE OF SCLEROTINIA HOMEOCARPA TO IPRODIONE. A.R. Detweiler and J.M. Vargas Jr., Botany and Plant Pathology, Michigan State University, E. Lansing, MI 48824

An iprodione-resistant strain of *Sclerotinia homoeocarpa* was isolated from a creeping bentgrass putting green where disease management for dollar spot had failed. The resistant strain grew on potato dextrose agar amended with 1000 µg/ml iprodione while a sensitive strain and a benzimidazole-resistant strain failed to grow on media amended with more than 1 µg/ml iprodione. The iprodione-resistant strain was also resistant to benomyl and maintained its resistance to both fungicides after 4 months in their absence. Although the iprodione-resistant strain was less virulent than the sensitive or benzimidazole-resistant strain in greenhouse trials, spray applications of iprodione and benomyl failed to control this strain under field conditions.

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ISOLATION OF THE BACTERIUM CAUSING A WILT DISEASE OF TORONTO CREEPING BENTGRASS. D.L. Roberts, J.M. Vargas, Jr., and K.K. Baker. Dept of Botany and Plant Pathology, Michigan State Univ., East Lansing, MI 48824.

Rod-shaped bacteria were consistently isolated from Toronto creeping bentgrass plants exhibiting wilt symptoms. Bacteria were grown in nutrient broth for 72 hr, pelleted by centrifugation, resuspended in 0.01 M phosphate buffer (pH 7.2) and used to inoculate individual Toronto creeping bentgrass plants. Plants with injured roots and/or leaves were immersed in the bacterial suspension whereas injured control plants were immersed in 0.01M phosphate buffer. Typical wilt symptoms developed on the inoculated plants after 5-7 days. Isolation and reinoculation showed similar results. Scanning and transmission electron microscopy confirmed the presence of bacteria within xylem vessels of inoculated plants. The isolated bacterium was similar in ultrastructure to those found in diseased plants from the field. These results prove that the isolated bacterium is the cause of "bacterial wilt of Toronto creeping bentgrass".

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SYMPTOM SUPPRESSION WITH OXYTETRACYCLINE OF A WILT DISEASE OF TORONTO CREEPING BENTGRASS. D.L. Roberts, J.M. Vargas, Jr., and K.K. Baker. Dept of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824

Symptom suppression of a Toronto creeping bentgrass disease of presumed bacterial etiology was achieved with 1.0 and 1.5 g/L solutions of oxytetracycline applied to field plots as drench treatments at the rate of 2 L/m². Streptomycin sulfate and cupric hydroxide applied at the same rate did not reduce disease development. Scanning electron microscopy revealed numerous bacteria in the xylem of untreated, streptomycin sulfate-treated and cupric hydroxide-treated plants whereas no bacteria were found in oxytetracycline-treated plants. This evidence supports our previous findings that a bacterium is the causal agent of this wilt disease of Toronto creeping bentgrass.

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A RECENT OUTBREAK OF WITCHES' BROOM OF ROSE IN EASTERN KANSAS AND WESTERN MISSOURI. F. J. Crowe, Department of Plant Pathology, Kansas State University, Manhattan, Kansas 66506.

Incidence of witches' broom or rosette of rose in eastern Kansas and western Missouri gradually increased from no reports prior to 1977 to several hundred reports in 1981. Symptoms were similar to those described for a few isolated wild rose specimens from Canada and Wyoming in 1941 and from northeastern California

in 1941 and 1970, and also for numerous *Rosa multiflora* hedgerow plantings in Nebraska in 1961. Stems elongated rapidly on certain branches followed by breaking of axillary buds, leaflet deformation and wrinkling, bright red pigmentation which failed to turn green, phyllody and increased thorniness. Symptoms gradually spread to all new growth on affected plants. Affected roses died during the season in which symptoms developed or during the following winter. In contrast to previous reports, recent incidence here was high among garden roses located far from wild roses or *Rosa multiflora* hedges. Evidence suggests spread of some undetermined causal agent within this geographical area.

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TREATMENT OF ROSE FLOWERS FOR CONTROL OF GRAY MOLD. K.E. Welch, Mallinckrodt, Inc., St. Louis, Mo. 63147, and A.H. McCain, University of California, Berkeley, California 94720

The major loss from gray mold disease of roses, caused by *Botrytis cinerea*, develops in transit or storage when conidia of the fungus carried on flowers germinate and infect petals. Vinclozolin (Ornalin) was evaluated as a post-harvest treatment of cut flowers to prevent gray mold. 'Samantha' roses, naturally contaminated with *Botrytis* in the greenhouse, were dipped 3 to 4 seconds in various concentrations of the fungicide and incubated under high humidity at 20 to 21°C for 11 days. Three days after treatment the average number of large lesions per flower when flowers were dipped in the fungicide rates of 0.0, 0.45, 0.90, 1.8 and 3.6 g/liter water was 16.8, 0.7, 0.0, 0.0, and 0.0 respectively. The number of small lesions per flower was 4.3, 22.0, 29.3, 15.2, and 2.3, respectively at the above rates. Eleven days after treatment, the flowers not treated with fungicide were completely rotted. The two higher rates of vinclozolin eliminated rot. Lower rates of the fungicide significantly reduced rot. Flecking of flowers, which results from toxins produced by germinating conidia, was reduced, but not eliminated by fungicide treatments.

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A NEW LEAF SPOT DISEASE OF GERANIUM INCITED BY PSEUDOMONAS CICHORII. A. W. Engelhard, H. C. Mellinger, R. C. Ploetz, J. W. Miller. AREC, Univ. of Florida, Bradenton, FL 33508; Glades Crop Care, Ft. Myers, FL 33901; Univ. of Florida, Gainesville, FL 32611; Division of Plant Industry, Gainesville, FL 32601.

A previously undescribed leaf spot disease of geranium has been observed in Florida since 1975. Leaf lesions are the most prominent symptom, ranging in size from 0.5 to 10mm in diameter. In the rain, wet appearing, dark colored irregular shaped necrotic areas 5-10+mm develop. They may enlarge along veins. Coalescing spots may encompass large sections of a leaf. Chlorosis usually develops within two days in the tissue adjacent to lesions. Leaf spots become dark brown to black. Infection along leaf margins and the resultant necrotic tissue causes leaves to curl. Severely affected leaves abscise. Infected buds fail to open. Lesions also develop on peduncles but not on stems. Both seedling and zonal types were susceptible on inoculation. The pathogen produced a fluorescent pigment on medium B of King et al, was oxidase positive, arginine dihydrolase negative and was identified as *Pseudomonas cichorii*.

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EFFECT OF NITROGEN, FLOREL, AND CALCIUM ON BOTRYTIS BLIGHT OF GERANIUM. Molly Niedbalski Cline and B.A. Eisenberg, Department of Plant Pathology and Horticulture, respectively, University of Illinois, 1102 S. Goodwin Ave., Urbana, IL 61801.

Geranium (*Pelargonium x hortorum* cv. Yours Truly) stock plants were irrigated with either 50,200 or 400 ppm nitrogen. Florel ((2-Chloroethyl)phosphonic acid) at 750 ppm, and calcium chloride at 100 kg/ha, were applied as foliar sprays. Cuttings taken from stock plants were inoculated with an isolate of *Botrytis cinerea* (1-5x10⁶ conidia/ml) and placed either under mist for 21 days or in storage at 3°C, 96% RH, and after 14 days placed under mist. Severity and incidence of gray mold on stems and foliage increased significantly as nitrogen levels increased both under mist or in storage. The presence of the pathogen decreased the rate of rooting and root quality. Calcium had no effect on disease development whereas cuttings from Florel-treated stock plants were more susceptible to *B. cinerea*.

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DEVELOPMENT AND CONTROL OF FIRE ON DAFFODILS. G. A Chastagner, Department of Plant Pathology, Washington State University, Puyallup, WA 98371.

Fire, a foliar disease caused by *Botryotinia polyblastis* (Greg.)

Buchw., has been commonly observed in plantings of daffodils in western Washington since 1978. Disease development has been commonly confused with natural senescence. Initial production of apothecia during 1980 and 1981 occurred in early April and mid March, respectively. Apothecial numbers reached a peak after 2 weeks, then gradually declined during the next 3 to 4 weeks. Subsequent foliar infections by conidia from infected flowers appeared in late April during both years. Since 1979, initial foliar infections have been followed by rapid disease spread and foliage death within 3 to 6 weeks. Rapid distal, then proximal yellowing from infection sites coupled with limited lesion development suggests that a toxin is involved in symptom development. Benomyl applications at 0.56 kg a.i./ha in late April and mid May have provided effective disease control.

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INFLUENCE OF ACEPHATE ON ALTERNARIA LEAFSPOT OF SCHEFFLERA AND THE PATHOGEN. A. R. Chase and L. S. Osborne, University of Florida, Agricultural Research Center-Apopka, Rt. 3 Box 580, Apopka, FL 32703.

The influence of acephate (Orthene), an insecticide widely used in the foliage plant industry, on a nontarget organism, *Alternaria* sp. was tested on schefflera (*Brassaia actinophylla* Endl.). Four treatments were applied: 1. plants sprayed with acephate (0.39 g/l, recommended rate) 24 hr before inoculation with the pathogen (1x10⁴ conidia/ml); 2. plants sprayed 48 hr after inoculation; 3. nonsprayed, inoculated plants; and 4. non-sprayed, noninoculated plants. Acephate was applied also as a soil drench in a separate experiment. Disease severity was rated on a scale of 1 (no disease) to 5 (plant death) 1 wk after inoculation. Foliar sprays with acephate prior to inoculation reduced the severity of disease. In some cases, soil drenches increased leafspot severity. In vitro growth and germination of *Alternaria* sp. on media amended with up to 1560 ppm (4x recommended rate) of acephate were not affected significantly. It is possible that acephate affects the host plant resistance but not the pathogen directly.

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LEAF SPOTS AND BLIGHTS OF FOLIAGE PLANTS INCITED BY ALTERNARIA SPECIES. R. A. Atilano, University of Florida, AREC, 3205 SW College Ave., Ft. Lauderdale, FL 33314.

An *Alternaria* sp. was shown to cause leaf spots and blights of ornamental foliage plant species and cultivars of the genera *Brassaia*, *Polyscias*, *Schefflera*, and *Tupidanthus*. Symptoms in artificially inoculated plants were identical to those observed in natural infections. The fungus was readily reisolated on potato dextrose agar from inoculated but not control plants. Mycelial growth and conidial germination were most rapid at 24 and 27°C. Disease severity in *B. actinophylla* and *S. arboricola* was greater following inoculation at 18°C than at 24°C. Conidia from 72 h cultures on V-8 Juice agar averaged 76.9 µm (range: 45 to 105 µm) by 23.8 µm (range: 16.5 to 30.8 µm), and the spore body length averaged 53 µm. Cultivars of aster, cabbage, periwinkle, onion, squash, tomato, and zinnia were not susceptible in inoculation experiments.

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BUD ROT OF DRACAENA DEREMENSIS CAUSED BY ERWINIA CAROTOVORA. J. W. Miller, Florida Department of Agriculture & Consumer Services, P. O. Box 1269, Gainesville, FL 32602.

Soft rot was observed on imported unrooted tip cuttings of *Dracaena deremensis*. This was expressed as a dark green water-soaked and/or soft decay extending up from the bud area. Alternating dark-gray zones were often seen in the rotted tissue. Similar symptoms were observed in established plants of this and other species of dracaena. *Erwinia carotovora* was consistently isolated from these symptomatic plant parts. Inoculated unrooted cuttings and growing tips on *Dracaena deremensis* developed the disease only following wounding of the tissue. Reisolation from symptomatic inoculated tissue consistently yielded *Erwinia carotovora*.

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FOLIAR BLIGHT OF ORNAMENTAL ARTEMESIA IN NEW JERSEY. J. L. Peterson, G. Koslow, and A. S. Green, Department of Plant Pathology, Cook College, New Jersey Agricultural Experiment Station, Rutgers University, New Brunswick, New Jersey 08903.

A foliar blight of ornamental *Artemesia* has been observed in commercial plantings in New Jersey since 1975. Infected leaves turn brownish and are smaller than the silvery leaves of healthy plants. Lower leaves die and plants become stunted rendering

them useless for dry foliage in cut flower arrangements. *Nematostoma occidentalis* produces both ascostroma and pycnidial stages among the leaf hairs. Single ascospore cultures gave rise to pycnidia but no ascostroma on several solid media. Six out of 14 ornamental *Artemisia* species sprayed with a conidial suspension of *Nematostoma* and held in a chamber with intermittent mist for 21 days or until disease symptoms appeared were susceptible. Sporulation was observed on the susceptible species. Disease control in the field was most effective using mancozeb or benomyl sprays at 2-week intervals from mid-June to mid-August. New Jersey Agricultural Experiment Station No. K-11410-1-82.

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LEAF BLIGHT OF *BRASSAIA ACTINOPHYLLA* CAUSED BY *ALTERNARIA PANAX*. J. Y. Uchida and M. Aragaki. Department of Plant Pathology, University of Hawaii, Honolulu, HI 96822.

A severe leaf spot and blight on *Brassia actinophylla*, *Dizygotheca elegantissima*, *Tupidanthus calyptratus*, and *Schefflera arboricola* was shown to be caused by *Alternaria panax*. Leaf spots on *B. actinophylla* were tan to dark brown, surrounded by chlorotic zones, circular to irregular, up to 20 mm in diameter, and frequently expanding into blackish to gray-green blights. Considerable leaflet drop occurred with heavy infection. No host specialization was observed in cross-inoculations among these aralioid species. *Alternaria panax* is highly variable in conidial morphology. Conidia were dark brown, obclavate, and pseudoroseate at 24°C; on vegetable juice agar they averaged $72.5 \times 21.8 \mu\text{m}$ whereas on autoclaved host tissue conidia averaged $105.2 \times 29.4 \mu\text{m}$ or more than 100% larger in volume. Conidia produced at 16°C were slender, obclavate, and symmetrical, while those formed at 28°C were broad, distorted, and asymmetrical. More severely disorganized conidia were produced in abundance on a cellulose-yeast extract-malt extract agar.

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EDAPHIC PARAMETERS ASSOCIATED WITH SHORE JUNIPER DECLINE. D. R. Fravel, D. M. Benson, and R. I. Bruck, Department of Plant Pathology, North Carolina State University, Raleigh 27650.

In the absence of evidence of a biotic agent as the primary cause of decline of shore juniper (*Juniperus conferta* Parl.), abiotic factors were examined to determine their roles in contributing to decline. Six of 20 edaphic components measured in 20 landscape plantings were significantly interrelated to decline index in a multivariate Principal Axis Factor Analysis. These parameters were calcium, clay + silt content, magnesium, nitrate, phosphorus, and zinc. Where soil horizons could be distinguished, parameters of the A+B horizon were a better indicator of decline than those of the C horizon. Supportive evidence for the involvement of these components was provided by tissue nutrient analysis from landscape plantings and from greenhouse studies of nutrient deficiencies and water stress where nitrogen deficiency and in one case, both water excesses and deficiencies, induced decline-like symptoms.

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EFFECT OF PINE BARK AT TWO MATRIC POTENTIALS ON SPORULATION OF *PHYTOPHTHORA CINNAMOMI*. D. M. Benson, Dept. of Plant Pathology, North Carolina State University, Raleigh 27650.

Sporangium formation by *P. cinnamomi* was compared in pine bark and a conducive clay soil at -15 and -100 mbars ψm . Mycelium produced on nylon screens in LBA was rinsed, soaked 2 hr in salt solution then rinsed and placed on a 1-cm deep layer of saturated pine bark or soil in a Buchner funnel tensiometer. A 0.5 cm layer of pine bark or soil was added and saturated from below. Water columns were then adjusted to establish -15 or -100 mbar tensions. Mycelium was recovered after 1 day, washed, stained, and sporangia(s) counted. Mycelium incubated 1 day in water (control) after the 2 hr salt soak formed $11 \pm 7 \text{ s/cm}^2$. Few sporangia ($<1/\text{cm}^2$) were formed on mycelium in pinebark that was dry initially. Pine bark tested after a 7 day period of daily watering in the greenhouse was somewhat stimulating ($39\text{--}48 \text{ s/cm}^2$), but highly stimulatory ($77\text{--}284 \text{ s/cm}^2$) when used after 2 or more wk. Sporangia were formed abundantly ($23\text{--}314/\text{cm}^2$) in clay soil even though it was air dried initially. Sporangia were more abundant in both pine bark and soil at -15 than at -100 mbars.

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HISTOLOGY OF RESISTANT AND SUSCEPTIBLE REACTIONS IN SLASH PINE INOCULATED AT 1 YEAR OF AGE WITH FUSIFORM RUST. F. F. Jewell, Sr., School of Forestry, Louisiana Tech. Univ., Ruston, LA 71272; and C. H. Walkinshaw, Southern Forest Expt. Stn., Gulfport, MS 39503.

New stem growth flushes of potted 1-year-old slash pine (*Pinus*

elliottii var. *elliottii*) were inoculated by a culture of fusiform rust (*Cronartium quercuum* f. sp. *fusiforme*). Symptoms, developing galls or purple stem spots, formed in 9 weeks. Microscopically, resistant reactions were evidenced by small ($1.6 \times 4\text{--}1.9 \times .8 \text{ mm}$) zones of tanninized and necrotic host cells isolated by a periderm in the primary cortex. Hyphae present in the zone were absent in the adjacent normal host cells. Susceptible reactions originated in areas of heavily tanninized cells, but a periderm was lacking. Typically intercellular rust hyphae were prominent in the phloem, cambium, and outer xylem, while atypically large (7μ vs 3.4μ) hyphae were frequent proximal to the epidermis, suggesting early involvement in infection. Haustoria associated with atypical hyphae were rare, indicating the possibility of, at least, a temporary saprophytic relationship of the rust fungi with the host.

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ALTERATION IN GIBBERELLIN LEVELS IN SOUTHERN PINES INOCULATED WITH THE FUSIFORM RUST FUNGUS. Dallas Seifers and Vernon Ammon. Department of Plant Pathology and Weed Science, Mississippi State University, Mississippi State, MS 39762.

Extracts from tissues removed from 3- and 6-mo-old healthy and inoculated susceptible (bulk slash), tolerant (FA-2 slash), and resistant (shortleaf) pine seedlings were assayed for gibberellin-like activity (GLA). GLA in healthy seedlings was higher at 6 mo than at 3 mo and highest in shortleaf pine and lowest in bulk slash pine for both time periods. Activity of gibberellin-like substances in seedlings inoculated 3 mo prior to assay increased in all three reaction types. At 6-mo, GLA decreased in tissue extracts from tolerant FA-2 slash pine and resistant shortleaf pine seedlings. GLA in inoculated bulk slash pine extracts remained higher than in uninoculated controls. Qualitative differences in gibberellin-like substances were observed among the three reaction groups and between healthy and inoculated seedlings.

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EFFECT OF TRIADIMEFON ON SURVIVAL OF OUTPLANTED HEALTHY AND FUSIFORM RUST-INFECTED LOBLOLLY PINE SEEDLINGS. W. D. Kelley, Dept. of Botany, Plant Pathology, and Microbiology, Ala. Agric. Exp. Stn., Auburn Univ., AL 36849.

Results of two field tests showed that triadimefon (Bayleton 5G) applied at planting time may be detrimental to loblolly pine (*Pinus taeda* L.) seedlings depending on rate and method of chemical application. In one test, where both healthy and infected seedlings received either 0 or 100 mg ai of triadimefon/seedling in the planting slit at transplant, survival after one year was significantly less in triadimefon-treated plots for both healthy (50.5%) and infected (5.5%) seedlings compared to non-treated healthy (93.5%) and infected (36.5%). In the other test, where only healthy seedlings were used and triadimefon was applied to the soil surface at rates of 0, 12.5, 25, 50, and 100 mg ai/seedling after the transplant slit was closed, seedling survival after one year was significantly decreased only in plots treated with the 100 mg rate (68%); survival in other triadimefon-treated plots was equal to or better than controls (80%).

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INFLUENCE OF THE TELIAL HOST ON THE VIRULENCE OF *CRONARTIUM QUERCUM* F. SP. *FUSIFORME*. L. David Dwinell. USDA For. Serv., Southeast. For. Expt. Sta., Athens, Ga. 30602

Standardized basidiospore inoculum from seedlings of 20 artificially-inoculated families of water, willow, northern red, blackjack and sawtooth oaks was used to inoculate seedlings of 8 families of loblolly and slash pines. The % galled seedlings was determined after 9 mo and the data were analyzed by nested-factorial ANOVA procedures. The % galled seedlings for both pine species was significantly related to families within oak species and pine families. In loblolly pine, oak species \times pine families interaction was also significant. Inocula from northern red and willow oak caused significantly greater infection on two rust resistant families (9R & 29R) than inocula from the other oaks. Variation in virulence due to the oak host is probably of limited importance in nature; however, the oak host used in evaluating rust resistance in loblolly pine in artificial screening procedures should be given greater consideration.

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MAJOR GENE RESISTANCE TO BLISTER RUST IN *PINUS LAMBERTIANA* IS EXPRESSED IN TISSUE CULTURE. A. M.

Callus cultures were grown from segments of blister rust-susceptible (rr) and hypersensitive-resistant (Rr) sugar pine embryos. Calli were inoculated with vegetative hyphae of *Cronartium ribicola* generated from basidiospores in axenic culture. Growth of the fungal mycelium either on or within the rr calli was 4X as extensive as that exhibited by inoculated Rr calli. The resistant callus surface showed macroscopic necrosis beneath each inoculum. This necrosis was expressed microscopically as densely stained cell layers, and was not reversed by increasing the cytokinin concentration to 110 micromolar in the nutrient medium. This is the first reported expression of genetic resistance to a rust disease by tissue culture and is furthermore exceptional in its insensitivity to reversal by cytokinin.

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METHYL BROMIDE ERADICATION OF THE OAK WILT FUNGUS IN LOGS AND LUMBER. E.L. Schmidt, W.L. MacDonald, M.M. Rutze and N.G. Klag, Dept. of Plant Path., Univ. of Minnesota, St. Paul, MN 55108, Dept. of Plant Path. and Ag. Micb. West Virginia University, Morgantown, W. Va. 26506, Ordinariat fur Holzbiologie, Hamburg, West Germany and APHIS Methods Dev. Ctr., Hoboken, N. J. 07030.

In outdoor trials in Minnesota, Indiana and West Virginia, oak logs and lumber infested with *Ceratocystis fagacearum* were covered with a polyethylene tarp and methyl bromide was introduced at 240g/m³ of space under the tarp. More gas was added after 24 hr to regain the initial concentration. After 2 - 4 days for logs, or 2 - 3 days for lumber, at temperatures as low as 5 C, the fungus could no longer be isolated. Over 20,000 isolation attempts were made from logs, 2,000+ from lumber. Lumber or veneer were neither discolored nor degraded by fumigation, but molds grew better on treated sapwood under warm, humid conditions. Methyl bromide was not detected in air near treated wood within minutes after uncovering, but gas desorption must be monitored when treated wood is enclosed in transit.

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WAVE YEAR INFECTION OF SHELTERBELT PINES IN NORTH DAKOTA BY *ENDOCRONARTIUM HARKNESSII*. T.R. Meyer, J.A. Walla, and R.W. Stack, North Dakota State Univ., Fargo, ND 58105.

We previously reported that most infections by *Endocronartium harknessii* of ponderosa pines in North Dakota shelterbelts appear to occur during wave years. Wave years of infection have been noted in other environments but not in the Northern Great Plains. In 1980, galls on pines in five northeast North Dakota shelterbelts planted in 1939 were examined. Year of infection was based on age of branches on which the perennial galls occurred. For 214 galls found on 51 pines, infections occurred throughout the years 1955 to 1977 but 60% occurred in a single year - 1964. Spore release in North Dakota appears to occur in or near the month of June. June, 1964, was cool and very wet and the preceding July to June seasonal heating degree day accumulation was very low. This combination of unusual climatic conditions was unique in the period 1955-80, and was coincident with the wave year. We suggest the observed infection wave year of 1964 in these shelterbelts is related to the unusual climatic conditions of that year.

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PROPORTION OF TREES INFECTED IN *ARCEUTHOBium AMERICANUM* INFECTION CENTERS IN JACK PINE STANDS. F.A. Baker and D.W. French, University of Minnesota, St. Paul, MN 55108; and Y. Beaubien and K. Knowles, Manitoba Department of Natural Resources, Winnipeg, Manitoba, Canada R3H 0W9.

Arceuthobium americanum reduces volume growth, deforms and eventually kills infected *Pinus banksiana* in Alberta, Saskatchewan, and Manitoba. To obtain information about rates of infection and spread, transects were established through infection centers and into adjacent uninfested areas of the same stand. At 5 m intervals infected and uninfested trees were counted, and height and diameter of each tree were measured. The mean distance from 0 to 100% infection was 16.0 ± 4.2 m. From the point of 100% infection most distant from the center of infestation, percent infection decreased linearly (slope = 5.71, intercept = 96.99, r-squared = .844). Stand density at the point of 100% infection was on the average 23% less than in adjacent uninfested portions of the same stand. Ten m toward the center of infestation from this point, stand density was only 20% of that in uninfested areas.

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FUNGI ASSOCIATED WITH ROOT DISEASE OF SAND PINE IN FLORIDA. E.L. Barnard, J.T. English, R.L. Anderson, and G.M. Blakeslee, Fla. Dep. Agric. and Consum. Serv., Gainesville 32602, USDA Forest Serv., Asheville, NC 28803, and Sch. Forestry, Univ. of Florida, Gainesville 32611.

Evaluation of more than 200 sand pine (*Pinus clausa*) stands indicated that root disease causes substantial damage to the species in Florida. Both the Ocala and Choctawhatchee varieties were found to be affected; disease incidence was greater in the former. Six known or suspected root disease fungi were isolated, singly or in combination, from roots of diseased and apparently healthy trees in 19 stands with high disease incidence. *Phytophthora cinnamomi* and *Inonotus circinatus* were isolated most frequently from roots in young plantations and older natural stands, respectively. *Armillariella tabescens* and *Verticillium dactyloides* were isolated less frequently from trees of various ages in both natural and planted stands. *Heterobasidium annosum* and *Phaeolus schweinitzii* were isolated only rarely.

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PITCH CANKER IN SOUTHERN PINE SEED ORCHARDS. L. David Dwinell and Jane B. Barrows-Broadus. USDA For. Serv., Southeast. For. Expt. Sta., Athens, Ga. 30602

Pitch canker, caused by *Fusarium moniliforme* var. *subglutinans* (FMS), is an important disease in southern pine seed orchards. The disease has been confirmed in over 40 seed orchards in North Carolina, South Carolina, Georgia, Florida, Alabama, Mississippi and Texas, on loblolly, longleaf, shortleaf, slash and Virginia pines. The pitch canker fungus is a wound parasite. Bole cankers on slash pine are usually associated with the use of mechanical tree shakers for cone harvest. In loblolly pine seed orchards, shoot dieback is the most common symptom; it is often associated with wounds caused by twisting and tearing cones off branches. Cankers on Virginia and shortleaf pines in at least two seed orchards are apparently related to damage caused by adverse weather, such as hurricanes or tornadoes. In seed orchards, some clones often are severely affected while others in the same orchard remain disease-free. FMS also causes strobilus mortality and seed deterioration.

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USE OF ANHYDROUS AMMONIA AS A CARRIER FOR AGRICULTURAL CHEMICALS. D.M. Huber and M.A. Ross. Dept. of Botany & Plant Pathology, Purdue University, West Lafayette, IN 47907.

Although aqueous nitrogen solutions have been extensively used as carriers for various agricultural chemicals, little research on NH₃ as a carrier is available. Recently developed "downstream" injectors overcome previously encountered problems of incompatibility or insolubility in NH₃. Our research injecting systemic fungicides, fumigants, nitrification inhibitors, trace elements, herbicides, and insecticides into the NH₃ stream indicate significant advantages of simultaneous application of agricultural chemicals with NH₃. These include enhanced efficacy and dispersion in soil, along with reduced cost of application and non-target effects. By using NH₃ as a carrier, we achieved soil incorporation under minimum tillage programs without the use of large volumes of water or emulsifiable formulations. Production efficiency should also be realized from the simultaneous applications of many agricultural chemicals in the NH₃ band.

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EFFECT ON FUNGICIDES IN FLUID DRILLING GEL MEDIA ON DAMPING-OFF OF *PARTHENIUM ARGENTATUM* G. J. M. Richardson, R. A. Backhaus, and J. C. Stutz. Division of Agriculture, Arizona State University, Tempe, AZ 85287

Captan, Etridiazole, Etridiazole/Thiophanate-methyl, PCNB, and Fenamiosulf were tested for their ability to prevent damping-off of *P. argentatum* seedlings in greenhouse tests. Fungicides were incorporated into a magnesium silicate fluid drilling gel containing pregerminated seeds and sown into vermiculite inoculated with *Pythium* spp. and *Rhizoctonia solani*. Phytotoxic levels of fungicides were determined. Preliminary studies showed enhanced emergence and vigor of seedlings sown in gels containing Etridiazole when inoculated with *Pythium*.

TOLERANCE OF VERTICILLIUM DAHLIAE KLEB. TO BENZIMIDAZOLES
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Verticillium isolates were screened for tolerance to methyl-2-benzimidazolecarbamate phosphate (MBC-P). Twenty-two microsclerotial and dark mycelial isolates were partially or totally inhibited at 1 µg/ml while seven album isolates grew at 5 µg/ml. Microsclerotial isolates of *V. dahliae* were sensitive to other benzimidazoles at 1 µg/ml while album isolates grew at 100 µg/ml. Subsequently, one microsclerotial isolate of *V. dahliae* was found to grow at 500 µg/ml MBC-P. Chemical tolerance remained stable after 40 weeks storage at 2°C or after 13 serial subcultures onto unamended potato dextrose agar. Sectors from tolerant, album isolates grew on 1000 µg/ml of MBC-P and exhibited the same stability of chemical tolerance as the parent isolates. Inoculation studies with a chemically sensitive microsclerotial and two tolerant album isolates from elm indicated that the microsclerotial isolate was more pathogenic.

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REDUCED FITNESS OF BENOMYL-RESISTANT *MONILINIA LAXA*. Victor M. Cañez, Jr. and Joseph M. Ogawa. Department of Plant Pathology, University of California, Davis 95616.

Fitness of benomyl-resistant *Monilinia laxa* isolated from apricot fruit in 1980 was examined. *M. laxa* isolates resistant to 1.0 mg/L benomyl exhibited ED₅₀ values of 0.4 - 0.7 mg/L benomyl for inhibition of germ tube elongation. Benomyl-resistant *M. laxa* isolates exhibited reduced germination percentages and rates compared to sensitive isolates on benomyl-free potato dextrose agar. In field studies, unopened almond and prune blossoms inoculated with various spore concentrations of benomyl-resistant *M. laxa* had less disease than blossoms similarly inoculated with benomyl-sensitive isolates. Longer incubation periods were required for symptom expression in blossoms inoculated with benomyl-resistant *M. laxa* isolates. The ability of benomyl-resistant isolates to compete with benomyl-sensitive isolates also was studied by inoculating unopened blossoms with a mixture containing both benomyl-resistant and -sensitive spores.

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ACTIVITY OF CAPTAN AND PROCHLORAZ ON BENOMYL-SENSITIVE AND -RESISTANT ISOLATES OF *MONILINIA FRUCTICOLA*. J. P. Dijkhuizen, Agric. Univ., Wageningen, The Netherlands; J. M. Ogawa and B. T. Manji, Department of Plant Pathology, University of California, Davis, CA 95616.

Benomyl-resistant isolates of *M. fructicola* grew slower on medium containing either 10 mg/L captan or 1 mg/L prochloraz. Spores exposed to 10 mg/L captan failed to germinate in 8 hr but germinated and grew later when placed onto fungicide-free potato-dextrose agar (PDA) but spores that germinated in 6 hr on 1 mg/L prochloraz failed to grow on PDA. Germ tubes of germinated spores of benomyl-resistant and -sensitive isolates placed on captan amended medium for 16 hr grew normally when transferred to PDA but further growth of germ tubes was restricted when first exposed to prochloraz for 4 hr. In a peach orchard with both benomyl-resistant (4 mg/L) and -sensitive populations, effective blossom blight control was obtained by a single pink-bud spray with prochloraz but not with captan or benomyl. Infected blossoms sprayed with prochloraz produced less spores than with captan.

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BASELINE SENSITIVITY OF *MONILINIA FRUCTICOLA* AND *MONILINIA LAXA* TO VANGARD. B. T. Manji, J. M. Ogawa, and E. A. Bovee, Department of Plant Pathology, Univ. of California, Davis, CA 95616

Baseline sensitivities of *M. fructicola* and *M. laxa* to the fungicide Vanguard (CGA 64251 IOW, Ciba-Geigy Corp., Greensboro, NC) were established by studying the effects of the chemical on conidial germination and mycelial growth. Experiments were conducted using Difco-PDA amended with the fungicide. Conidial germinations were 93% for *M. fructicola* and 96% for *M. laxa* on medium amended with 100 mg/L and 0% for both on medium containing 500 mg/L. ED₅₀ values for mycelial growth inhibition were 0.07 and 0.1 mg/L Vanguard for *M. fructicola* and *M. laxa*, respectively.

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UV IRRADIATION INDUCED RESISTANCE TO CGA 64251 IN *MONILINIA FRUCTICOLA*. R. L. Reese and J. F. Moore, Jr. AMAX Lead Co.,

Boss, MO 65440 and State Fruit Experiment Station, Southwest Missouri State University, Mountain Grove, MO 65711.

UV irradiated conidia of *Monilinia fructicola* were screened for resistance to CGA 64251 on a medium containing 4 µg/ml of the experimental compound. From 1.2×10^6 conidia irradiated, 137 colonies resistant to CGA 64251 were found. Eight resistant isolates were characterized *in vitro* and for pathogenicity on peach fruit. All were pathogenic and all but one sporulated on the fruit. Fruit dipped for 1 min in aqueous solutions of different concentrations of CGA 64251 and allowed to dry were inoculated with conidia of two resistant isolates and one nonresistant isolate. Growth of the resistant isolates was not inhibited on fruit treated with up to 100 µg/ml CGA 64251, whereas growth of the nonresistant isolate was prevented on fruit treated with 50 µg/ml CGA 64251. Resistance to CGA 64251 was lost by *M. fructicola* after three to nine passages from isolation of resistant colonies.

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EVIDENCE FOR HETEROTHALLISM AND MONOGENIC RESISTANCE TO BENOMYL IN *MONILINIA FRUCTICOLA*. R. M. Sonoda, University of Florida, Agricultural Research Center, Fort Pierce, FL 33454, J. M. Ogawa, E. A. Bovee, and P. L. Sholberg, University of California, Davis, CA 95616.

Nine hundred single ascospore isolates of *Monilinia fructicola* (Wint.) Honey, 30 each from 30 ascocarps were collected from a peach orchard in CA. Barrages occurred between colonies of isolates from the same ascocarps. The five ascocarps generating the fewest barrages contained two groups each of interacting isolates. The isolates in the two groups per ascocarp statistically fit a 1:1 ratio. The 25 other ascocarps contained four or more groups generating barrages. Eleven ascocarps contained both benomyl-sensitive and -resistant isolates. The isolates statistically fit a 1:1 ratio. Inheritance of factors determining barrage formation and resistance to benomyl appeared to segregate in a Mendelian manner. The occurrence of barrages between sister isolates in all 30 ascocarps and the presence of benomyl-sensitive and -resistant isolates within the same ascocarp indicate that the fungus is heterothallic. The 1:1 ratio of benomyl-resistant and -sensitive isolates in some ascocarps provide evidence that resistance is monogenic.

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ACTIVITY OF CGA-64250 (PROPICONAZOL) AGAINST *PHYMATOTRICHUM OMNIVORUM*. R.B. Hine and R.S. Whitson. Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

Activity of the sterol inhibiting fungicide, CGA-64250 was tested against 6 isolates of *Phymatotrichum omnivorum* in the laboratory by measuring mycelial growth from agar-disc inoculum placed on different media containing various concentrations of the fungicide. Growth from agar-discs was reduced by 34-47, 88-91, and 98-100% at concentrations of 1.0, 10.0, and 100 ng/ml (a.i.). No phytotoxicity occurred when cotton seeds (SJ-2) were planted into non-sterile field soil containing 6.25, 12.5, or 25 µg/ml (a.i.) whereas plant and root stunting was noted at 50 µg/ml (a.i.). In a field test at Marana, AZ, CGA-64250 applied as a side dress to 6-wk-old plants, reduced the incidence of *Phymatotrichum* root rot of cotton from 49% (check) to less than 5% at rates of 0.5, 1.0, and 2.0 lbs (a.i.)/acre.

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INSTABILITY OF INDUCED BENOMYL TOLERANCE IN *MYCOSPHAERELLA MELONIS*. P.T. Rotkis and M.E. Stanghellini, Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

The potential for *Mycosphaerella melonis* to develop tolerance to benomyl (Benlate) was tested by exposing aqueous pycnidiospore suspensions to ultraviolet (UV) light. Exposure times from 0 to 40 min were used. Both exposed and unexposed pycnidiospores failed to germinate and grow on 10 µg/ml benomyl V-8 agar. Regardless of exposure time, cultures could be established on 10 µg/ml benomyl V-8 agar after first germinating pycnidiospores on V-8 agar. UV-induced benomyl tolerance was maintained in isolates continuously subcultured onto 10 µg/ml benomyl V-8 agar, but was lost after two transfers to benomyl-free V-8 agar. Spontaneous tolerance to benomyl developed in some non-UV-treated isolates, but it was lost more rapidly than that observed in UV-treated isolates. The observed tolerance to benomyl in *M. melonis* was not found to be stable.

CELL WALL HYDROXYPROLINE ENHANCEMENT AS AN EARLY EVENT IN THE RESISTANCE OF CUCUMBER TO *CLADOSPORIUM CUCUMERINUM*. R. Hammerschmidt, D.T.A. Lampion* and E.P. Muldoon*, Department of Botany and Plant Pathology and *MSU/DOE Plant Research Laboratory, Michigan State University, East Lansing, MI 48824

A role for hydroxyproline-rich cell wall glycoproteins in disease resistance has been suggested for a few host-pathogen interactions. Early increases in host cell wall hydroxyproline (Hyp) content were observed in association with resistant, but not susceptible, responses of cucumber to *Cladosporium cucumerinum*. Time course studies demonstrated an increase in cell wall bound Hyp between 12-18 hours after inoculation of resistant host tissue. This increase correlates in time with penetration of the host by the fungus and the deposition of lignin in host cell walls near the points of penetration. Hyp content did not significantly increase in compatible interactions until after 48 hours after inoculation. At this time, the fungus had ramified deeply into the tissue. Enhanced Hyp content was also correlated with mature tissue resistance. No Hyp was detected in hydrolyzates of the fungus.

EFFECT OF VESSEL PLUGGING AND LENGTH OF PATHWAY ON HYDRAULIC CONDUCTIVITY OF XYLEM IN CITRUS TREES WITH BLIGHT. Mortimer Cohen, Agricultural Research Center, University of Florida, Fort Pierce, FL 33454.

Hydraulic conductivity of citrus xylem with no vessel plugging tended to be inversely proportional to the length of the xylem segment. Where a number of vessel plugs were found, the decrease in hydraulic conductivity with increase in length of the xylem segment was greater than inversely proportional. This phenomenon was observed with pencil sized secondary roots and with pegs punched out of wood disks cut across trunks and limbs of large trees. The relationship described helps to explain the extremely reduced water movement which characterized the inner xylem of trees with citrus blight disease.

CAMBIAL REACTIONS IN *ULMUS* FOLLOWING INOCULATION WITH *CERATOCYSTIS ULMI*. E.B. Smalley, N.S. Ehlke, E.F. Clark and S.H. Mai, Dept. of Plant Pathology, Univ. of Wisconsin, Madison, WI 53706

The interaction between *Ceratocystis ulmi* and *Ulmus* sp. was studied on inoculated freshly exposed cambium of axenic elm twigs incubated on water agar. Browning was evident within a few hours after inoculation and was most intense on resistant elm species. Response was least intense in young, greenhouse-grown *U. americana* seedlings, but increased with plant age. Increased response with plant age correlated with increasing host resistance in inoculated intact elms. Growth retardation of the fungus on cambium of older plants was often prolonged (>45 days). Intensity of browning was proportional to spore concentration, but washed carbendazim-inactivated or heat-killed spores at 10^8 spores/ml also induced moderate browning. Cell-free controls were inactive. Spores (10^8) in culture filtrate reduced callus healing, but similar diluted spores (10^7) stimulated callus overgrowth and subsequent plantlet development. Washed spores in 0.01 N HCl induced extreme browning, but permitted fungus growth. Manganese E and F were elicited following cambial inoculation of *U. americana* twigs.

AMMONIA ACCUMULATION AND ITS EFFECT ON THE MEMBRANE POTENTIAL IN BACTERIA-INOCULATED COTTON AND TOBACCO. W. R. Ullrich and A. Novacky, Institut für Botanik, TH Darmstadt FRG and Dept. of Plant Pathology, University of Missouri, Columbia, MO 65211

NH_3 accumulated up to 13 mM in detached cotton cotyledons inoculated with *Xanthomonas malvacearum* and to 10 mM with *Pseudomonas pisi*; in tobacco leaves inoculated with *Ps. pisi* the NH_3 conc. was 15 mM. NH_3 accumulation was 3-4 times higher in protein-rich young tobacco leaves than in old ones. If inoculated tissues were maintained in the dark the membrane potential (E_m) was as low as the diffusion potential (-95 mV). Upon illumination it recovered even beyond the control values (-185 mV). Externally applied NH_3 reduced E_m to the diffusion potential in healthy tissues. In bacteria-inoculated tissues NH_3 must originate from increased protein catabolism. It is suggested that in the light NH_3 is partly reassimilated via chloroplastic GS-GOGAT cycle and thus E_m can be maintained at high values. In the dark, in spite of the high ATP levels, E_m is low. NH_3 may alkalize the cytoplasm in the dark and hence inactivate the H^+ /ATPase at the plasmalemma that regulates E_m . (Supported by DFG and NSF).

CHARACTERIZATION OF A CUTINOLYTIC ENZYME FROM *COLLETOTRICHUM GLOEOSPORIOIDES* ON CARICA PAPAYA. Martin B. Dickman¹, Suresh S. Patil¹, & P.E. Kolattukudy², Dept. of Plant Pathology¹, Univ. of Hawaii, Honolulu, HI 96822, Institute of Biological Chemistry², Washington State Univ., Pullman, WA 99164.

A previously reported (Phytopathology 71:870) cutinase of *C. gloeosporioides*, causal agent of papaya anthracnose was characterized. The enzyme hydrolyzed all p-nitrophenyl esters of C_4 - C_{16} acids at comparable rates in contrast to the cutinase of *Fusarium solani* f. sp. *pisi*, the only other well characterized enzyme, which utilizes only p-nitrophenyl butyrate at appreciable rates. The *C. gloeosporioides* enzyme hydrolyzed p-nitrophenyl butyrate 135 fold slower than the *F. solani* f. sp. *pisi* cutinase. Also, with tritiated cutin as substrate the present enzyme had only 1.5% of the specific activity of that of the *Fusarium* enzyme. Rabbit antibodies made to the *C. gloeosporioides* cutinase showed a single precipitin band in Ouchterlony analysis but did not cross react with the *Fusarium* cutinase. The *C. gloeosporioides* cutinase is a glycoprotein. The carbohydrate residues (16%) are O-glycosidically linked to the protein.

COLLETOTRICHUM GLOEOSPORIOIDES SECRETES A CUTINASE TO BREACH THE CUTICULAR BARRIER OF PAPAYA FRUIT. Martin B. Dickman¹, Suresh S. Patil¹, & P.E. Kolattukudy², Dept. of Plant Pathology¹, Univ. of Hawaii, Honolulu, HI 96822, Institute of Biological Chemistry², Washington State Univ., Pullman WA 99164.

Specific antibodies prepared against purified *C. gloeosporioides* cutinase but not preimmune serum (Control), prevented infection of unwounded papaya tissue by *C. gloeosporioides*. Similarly, diisopropylfluorophosphate (DFP), an inhibitor of *C. gloeosporioides* cutinase also suppressed infection of papayas by the pathogen. However, neither anticutinase nor DFP prevented infection when the papaya cuticle was breached by a needle prick prior to inoculation with the pathogen. Also, neither anticutinase nor DFP affected germination of pathogen spores or mycelial growth. These results show that *C. gloeosporioides* penetrates the cuticle of papaya by secreting a cutinase. Additionally, when papayas were treated with purified cutinase of *C. gloeosporioides* prior to inoculation with spores of *Mycosphaerella* sp., a wound pathogen of papaya, infection and lesion formation occurred.

EFFICACY OF ORGANOPHOSPHOROUS COMPOUNDS IN PREVENTING INFECTION OF PAPAYA BY *COLLETOTRICHUM GLOEOSPORIOIDES*. Martin B. Dickman¹, Suresh S. Patil¹, & P.E. Kolattukudy², Dept. of Plant Pathology¹, Univ. of Hawaii, Honolulu, HI 96822, Institute of Biological Chemistry², Washington State Univ., Pullman WA 99164.

Diisopropylfluorophosphate, an organophosphorous serine hydrolyase inhibitor, and a potent inhibitor of *C. gloeosporioides* cutinase, functions as an antipenetrant against *C. gloeosporioides* in papaya infection. Fifteen other organophosphorous compounds were tested against purified *C. gloeosporioides* cutinase *in vitro*. Their I_{50} values (concentration at which enzyme activity is inhibited by 50%) ranged from 10^{-3}M to 10^{-9}M . The five most potent inhibitors were tested in laboratory bioassays for effectiveness in preventing infection of papaya fruit by *C. gloeosporioides*. Chlorpyrifos, Hinosan, phosphorothioic acid, O, O-dimethyl-O-(2,4,5-trichlorophenyl) phosphate, and O, O-diethyl-O-(3,5,6-trichloro-2 pyridyl) phosphate, were all effective in preventing infection at micromolar or lower concentrations. Field efficacy studies with one of these antipenetrants is in progress.

INVOLVEMENT OF FUSARIUM-PRODUCED PLANT CELL WALL DEGRADING ENZYMES IN VASCULAR GEL FORMATION. G. VanderMolen¹, J. M. Labavitch², L. Strand², and J. E. DeVay¹. ¹Department of Plant Pathology and ²Department of Pomology, University of California, Davis CA 95616.

Analyses of vascular gels extruded from banana roots inoculated with *Fusarium oxysporum cubense* (F.o.c.) indicated that gels contain elements of the pectic and hemicellulosic components of the host cell wall. Accordingly, we are investigating the hypothesis that wall degrading enzymes produced by the pathogen decompose host plant cell walls and that wall degradation products contribute to the formation of vascular gels. The complex mixture of enzymes produced by F.o.c. when grown on purified cell walls of banana leaves causes the formation of gels when introduced into the vascular systems of test plants. Isoelec-

tric focusing of this enzyme mixture was used to obtain purified enzyme components. The relative importance of polygalacturonase, B-1,4-xylanase and pectin lyase in cell wall maceration and vascular gelation will be discussed.

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THE EFFECT OF CUTICLE THICKNESS ON INFECTION OF OLDER BEAN HYPOCOTYLS BY *RHIZOCTONIA SOLANI*. Virginia Stockwell and Penelope Hanchey, Dept. of Botany & Plant Pathology, Colorado State University, Fort Collins, CO 80523.

Rhizoctonia solani fails to form infection cushions or lesions on three-week old Red Kidney bean plants. Previous ultra-structural studies indicate that the cuticle of three-week old beans is 3 - 4 times thicker than that of susceptible one-week old plants. The cuticle of older bean plants was removed by rubbing the hypocotyl with carborundum or chloroform. After inoculation, infection cushions formed and lesions developed. These results suggest that one component of age-induced resistance of bean plants to *R. solani* is related to increased cuticle thickness.

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CHLOROTETRACYCLINE PROMOTES OVERSIZE WALL APPPOSITIONS IN BARLEY EPIDERMAL CELLS. M. R. Marshall, J. R. Aist, and H. W. Israel. Department of Plant Pathology, Cornell University, Ithaca, NY 14853-0331.

Chlorotetracycline (CTC) is a selective fluorescent probe for Ca⁺⁺ in living cells. We have found that inner epidermal cells of barley coleoptiles prepared by dissection, when floated on a solution of 10⁻⁴ M CTC and 10⁻² M Ca(NO₃)₂, formed oversize papillae in response to penetration attempts by *Erysiphe graminis hordei*. These papillae were morphologically similar to those induced in the presence of Ca(H₂PO₄)₂ [Aist, et al., 1979, *Phytopathology* 69:1245], but their CTC fluorescence was more intense and uniform. Numerous oversize appositions were also found on the abaxial walls of the inner epidermis, far removed from the fungus. They were presumably induced by wounds that occurred when the outer epidermis was removed during tissue preparation. Therefore, CTC may sensitize cells to subsequent perturbation and should be used with caution, especially for long-term *in vivo* studies.

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PHYSIOLOGIC RACES OF *PUCCINIA HORDEI* ISOLATED IN THE UNITED STATES FROM 1979 THROUGH 1981. M.W. Andres, R.D. Wilcoxson and A.P. Roelfs, Department of Plant Pathology and Cereal Rust Laboratory, USDA, University of Minnesota, St. Paul, MN 55108.

Isolates of *Puccinia hordei* from 81 field collections made in 16 states were studied from 1979 through 1981. Most of the collections came from the Mississippi Valley, but others were from California, Delaware, Michigan, Pennsylvania, Virginia, Washington, and West Virginia.

Nine standard and some supplemental differential barley cultivars were used for race identification. Six physiologic races were found. Race 8 (virulent on Pa, Pa4, Pa8, avirulent on Pa2, Pa3, Pa7) was most common annually (83% of isolates). Race 4 was next most common overall, but it was not found in 1981. A trace amount of race 40 was present in 1980 and 1981. Traces of races 13, 19, and 42 were found only in 1981. All six physiologic races were avirulent on Aim (CI 3737, Pa3) and Cebada Capa (CI 6193, Pa7).

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Virulence patterns of *Puccinia hordei* in Texas and sources of resistance in barley. M. Reinhold, E. L. Sharp and H. E. Bockelman. Department of Plant Pathology, Montana State University, Bozeman, MT. 59717.

Virulence types of isolates of *Puccinia hordei* from different locations in Texas were determined. The standard differential set representing the resistance genes Pa-Pa9 was used. There was little variation in virulence of the isolates from widely separated locations. Among 176 samples of barley cultivars, numerous usable sources of resistance were detected. The genes Pa3, Pa7, and Pa9 imparted a wide spectrum of resistance. The Pa2 gene gave, in combination with at least one additional gene, sufficient protection in the cultivars 'Peruvian' and 'Ricardo'. Other resistant cultivars represented previously undescribed sources of resistance. The identified resistance sources are being used in a male sterile facilitated recurrent selection population for leaf rust resistance. Barley

populations from several cycles of recurrent selection were monitored at different locations in Texas. A considerable increase of resistance was observed in the later cycles.

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A FIELD COMPARISON OF SEXUAL AND ASEXUAL POPULATIONS OF *PUCCINIA CORONATA* FOR TRAITS OF AGGRESSIVENESS. J.H. Oard and M.D. Simons. ARS, USDA, and Dept. of Plant Pathology, I.S.U., Ames, IA 50011

In 1979 asexual cultures of *Puccinia coronata* were collected from buckthorn (*Rhamnus cathartica*) in a nursery in Minnesota to generate a sample population that had undergone sexual reproduction. A second population distant from buckthorn consisted of uredial cultures from southern Texas. Isolates derived from these cultures were used individually to inoculate 10 susceptible oat (*Avena sativa*) cultivars in irrigated and non-irrigated plots. None of the traits, including latent period, infectious period, or yield reduction indices, exhibited consistently significant differences between the two fungal populations. Highly significant differences were found, however, for all traits within each population. Narrow sense heritability estimates were generally high for each trait indicating that the response to selection is potentially large for both populations.

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EVOLUTION OF VIRULENCE OF *PUCCINIA CORONATA* ON THE OAT CULTIVAR TAM 0-301. M. D. Simons, USDA, ARS, Dept. of Plant Pathology, ISU, Ames, IA 50011; and M. E. McDaniel, Dept. of Soil & Crop Sciences, Texas A&M University, College Station, TX 77843.

The oat cultivar TAM 0-301, which carried resistance to *P. coronata* from *A. sterilis*, was released in Texas in 1973. It was widely grown in south Texas, and was highly resistant for several years. Seedlings of TAM 0-301 in the greenhouse at a mean temperature of about 22 C were highly resistant to all Texas isolates of *P. coronata* in 1974. In 1975-76, a few isolates induced moderately resistant reactions; in 1977-79 the proportion of isolates inducing moderate resistance increased, and some isolates induced susceptibility. In 1980, TAM 0-301 was susceptible to 19% of all isolates. In growth chambers, susceptibility and high resistance, as seen in the greenhouse, were not temperature sensitive, but isolates inducing moderate resistance generally induced high resistance at 20 C and susceptibility at 26 C. The same single gene in TAM 0-301 conditioned both high resistance and moderate, temperature-sensitive resistance.

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A METHOD OF CLONING THE BLISTER RUST FUNGUS IN AXENIC CULTURE. A. M. Diner and R. L. Mott, Department of Botany, North Carolina State University, Raleigh, N. C. 27650

Axenic cultures of *Cronartium ribicola* were grown from single, short vegetative hyphae isolated from a small population of germinating basidiospores. The white mycelial cultures consisted of mononucleate, straight, and regularly branched hyphae. The mycelium was viable in subculture and virulent on white pine callus; intercellular hyphae and intracellular haustoria were abundant in the inoculated host tissue. This method should prove useful in isolating individual races of the blister rust fungus for studies of fungal physiology and host-pathogen interrelationships.

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GENETIC DIVERSITY FOR VIRULENCE IN BEAN RUST COLLECTIONS. J.V. Groth and A.P. Roelfs, University of Minnesota, St. Paul, MN 55108.

Bean rust (*Uromyces phaseoli* var. *typica*) was sampled in fields in the midwestern U.S.A. and Mexico. Uredospores were removed from about 30 leaves from an area 5 M or less in diameter. Single-uredia were isolated and increased on the susceptible bean cultivar Pinto 111. Twelve to 20 selected bean lines were inoculated with each isolate and the reactions scored according to degree of necrosis and size of uredia. Five isolates from central Minnesota were of two phenotypes, three isolates from near Mexico City were of two phenotypes, and ten isolates from east-central North Dakota were of six phenotypes. Sexually reproducing populations of bean rust are highly polymorphic for virulence. This is true even though population density is high

and several uredial generations have preceded sampling. Small collections and small samples within collections were sufficient to reveal variation in all cases. Much of this variation cannot be explained by variation in corresponding host resistance.

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GENETICS OF A MINUTE UREDIUM REACTION TO BEAN RUST IN BEAN LINE 814. J.A. Kolmer and J.V. Groth, University of Minnesota, St. Paul, MN 55108.

Rust isolate S1-5 produces an infection type on bean line 814 characterized by a minute uredium. Cultivars Early Gallatin (EG) and Pinto 111 are moderately, and fully susceptible, respectively, to S1-5. EG has a single dominant gene conditioning hypersensitive resistance to P10-1. S1-5 produced the minute uredium on the F₁ of 814 by 111. The F₂ segregated in a 3:1, minute to large uredium ratio. The F₁ and F₂ were susceptible to P10-1. The F₁ from 814 by EG were resistant to both S1-5 and P10-1. The F₂ segregated in a 9:3:3:1 ratio. Cultivar U.S. #3 has a single dominant gene conditioning a large, sometimes sporulating fleck, when inoculated with S1-5. F₁ progeny of 814 by U.S. #3 produced small necrotic flecks when inoculated with S1-5. The F₂ segregated in a 12:3:1 ratio: progeny with minute, non-necrotic uredia grading into minute necrotic flecks; small sporulating uredia surrounded by areas of necrosis; and large uredia, respectively. The gene conditioning the minute uredium reaction in 814 appears to be epistatic to the gene conditioning the necrotic fleck in U.S. #3.

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IDENTIFICATION OF FIVE RACES OF *SPHAEROTHECA PANNOSA* VAR. *ROSAE*. C. L. Bender and D. L. Coyier, Dept. of Botany and Plant Pathology, Oregon State University and USDA-ARS Ornamental Plants Research Laboratory, Corvallis, Oregon 97330

Methods were developed for isolating and identifying races of *Sphaerotheca pannosa* var. *rosae* on rose. Monoconidial isolates from 9 different Rosa hosts were studied extensively. Isolates were initiated on detached leaves, increased on host plants, and then inoculated to rose cultivars to show differential reactions. Host compatibility was evaluated by assessing leaf necrosis, colony growth and sporulation of the fungus. The reactions of test roses were divided into several susceptibility categories. There was no macroscopic evidence of fungus colonization on roses rated very resistant. Roses rated moderately resistant developed necrosis which prevented or limited sporulation. Sporulation was macroscopically visible without cell death on roses rated susceptible. The 9 monoconidial isolates were grouped into 5 distinct races on the basis of their ability to parasitize the test roses. Races were described using a nomenclature system designed for identification of cereal rusts.

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NUCLEAR BEHAVIOR OF *PHELLINUS ARCTOSTAPHYLI*, *P. IGNIARIUS*, AND *P. TREMULAE*. Paul E. Hennon and Everett M. Hansen, Department of Botany and Plant Pathology, Oregon State University, Corvallis, Oregon 97331.

The number and distribution of nuclei in basidiospores, germinating basidiospores, homokaryons, heterokaryons, and basidiocarp tissues were examined microscopically. Three nuclear stains were used: giemsa, hematoxylin, and DAPI (4',6-diamidino-2-phenylindole). The nuclear behavior of these three species was similar. Basidiospores were produced with one nucleus, but often became binucleate prior to germination. One nucleus usually remained in the basidiospore following germination and germ tube elongation. Both homokaryons and heterokaryons had a variable number of unpaired nuclei, often equidistantly spaced between unclamped septa. The dikaryophase was delayed until basidiocarp formation, when paired nuclei were found in young basidia and mycelium underlying basidia. Basidia with one, two, or four nuclei were observed prior to spore formation. Four basidiospores formed on each basidium. Homokaryotic and heterokaryotic isolates could not be distinguished by nuclear condition or colony morphology. Heterokaryons grew significantly ($p = .05$) faster than homokaryons.

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VALIDITY OF SPECIES OF *THANATEPHORUS* AND *CERATOBASIDIUM*. Gerard C. Adams, Jr., Department of Plant Pathology, Oregon State University, Corvallis, OR 97331.

Taxonomists have distributed isolates of *Rhizoctonia solani* among three species (*Thanatephorus cucumeris*, *T. praticola*, and *T. sasakii*) and have described three perfect states for binucleate *Rhizoctonia* (*Ceratobasidium cornigerum*, *C. anceps*, and *C. ramicola*). The validity of each species is re-examined. *T. sasakii* was described for an ecotype of anastomo-

sis group 1 (AG-1) not for the entire group. However, no genetic data has demonstrated that it is isolated. The validity of *T. praticola* as a morphological species is also challenged because published descriptions of morphological differences between it and *T. cucumeris* included AG-4 isolates in both species. *C. ramicola* was described as the teleomorph of *Rhizoctonia ramicola*, however, the *R. ramicola* was misidentified. In addition, a single isolate expressed the range of differences said to separate *C. ramicola*, *C. cornigerum*, and *C. anceps*.

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A TREE FRUIT CROP MANAGEMENT MONITORING NETWORK. M. R. Schwarz, IPM Program, N. Y. S. Agric. Exp. Sta., Cornell University, Geneva, New York 14456.

A tree fruit monitoring system provides timely, uniform data to research and extension personnel having access to SCAMP, a computer-based information delivery system available throughout New York and adjacent states. Field personnel residing near abandoned and commercial orchards in Western New York State are supplied with identical environmental and biological monitoring equipment. A written protocol is followed by each cooperator to standardize operations and data collection from each site. Current information on tree phenology, apple scab infection periods, insect emergence, weather, and other observations are gathered at least three times per week and reported to the IPM Program central office via a toll-free telephone line. Data are then archived directly into SCAMP where they are available for comparison and interpretation by researchers, agents, and specialists.

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INTEGRATED PEST MANAGEMENT OF CABBAGE AND LETTUCE IN HAWAII. J. J. Cho, P. Jackson, and W. C. Mitchell, Departments of Plant Pathology & Entomology, University of Hawaii, P.O. Box 269, Kula, HI 96790.

Weekly surveys of 8 research-cooperator farm sites showed that 6 diseases of lettuce and two of cabbage to be most important in Hawaii. Tomato spotted wilt (TSWV) and lettuce mosaic (LMV) caused epidemic losses in lettuce on 2 farms and were the most economically damaging of the diseases. From 5 surveys TSWV losses recorded from Farm#1 were 67%, 31%, 18%, 32% and 14% and from Farm#6 were 0.1%, 0.1%, 1%, 5% and 8%. LMV losses recorded from Farm#1 were 18%, 48%, 80%, 11% and 1% and from Farm#6 0.4%, 0.1%, 0.7%, 0.2% and 6%. Three thrips, *Frankliniella occidentalis*, *F. schultzei*, & *Thrips tabaci*, capable of transmitting TSWV were found. The prevalent aphid species found capable of transmitting LMV were *Macrosiphum euphorbiae*, and *Myzus persicae*. Onset of TSWV incidence in each survey resulted after high thrips counts 2 wks prior to disease observation. Applications of methomyl significantly reduced TSWV incidence. Weed samples from IPM farm sites showed 2 of 11 *Malva parviflora* samples to be positive for LMV; 1 of 11 *M. parviflora*, 4 of 10 *Bidens pilosa*, 1 of 6 *Sonchus oleraceus* were positive for TSWV

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ON-FARM PEANUT LEAFSPOT FORECASTING. Jack E. Bailey, Lee Sawyer and Doug Thomson, Dept. Plant Pathology, NC State University, Raleigh, NC 27650

Jensen and Boyle's (1965 and 1966) peanut leafspot forecasting model was programmed into a portable, battery-operated microprocessor in a 22x25x11 cm box. The system is used to determine need for spraying by identifying weather conditions conducive to disease. The system has three main components: (i) relative humidity and temperature sensors, (ii) a microprocessor controlled interface circuitry with which the model is programmed and (iii) an information display panel. The panel displays three types of information: (i) spray or no-spray advisories, (ii) explanations for the advisory (based on work by Parvin, Smith and Crosby, 1974) and (iii) record of the spray advisories issued for the past 14 days. This system is inexpensive, easy to use and maximizes accuracy by using local field weather data to generate advisories.

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A CAPSTONE COURSE IN INTEGRATED PEST MANAGEMENT. Blanche Cournoyer Haning, North Carolina State University, Raleigh, 27650.

This course, Pest Management Seminar at North Carolina State University, is specifically designed to test individual students knowledge of IPM information and the ability to communicate it effectively and efficiently. Students are given relevant cultural and pest information from a typical regional multicrop farm for which they must develop a total management plan. They

work in small groups during the term, meeting weekly to share the assignment, integrate ideas, and assess their progress. Students present and defend their final management plans to a group of interdisciplinary faculty in addition to submitting a written report. This course follows a lecture-laboratory course on the principles and protocols of IPM systems and philosophy. It culminates many topics studied by students and lets them preview the decision-making process they may encounter in many professional settings.

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YIELD LOSS ASSOCIATED WITH EYESPOT ON IRRIGATED DENT CORN IN MINNESOTA. E. L. Stromberg, Department of Plant Pathology & Physiology, VPI & SU, Blacksburg, VA 24061, and M. Wiens, Irrigation Center Research Farm, University of Minnesota, Staples, MN 56479.

Yield losses associated with eyespot disease caused by *Kabatiella zeae* in two dent corn hybrids (Minhybrid 7301 and W64A x W117) under irrigation in Minnesota were determined in a two year study. Eyespot severity levels ranging from 1.4 to 73.0% on leaves spotted and/or blighted on the top half of plants were obtained by means of artificial inoculation at 8 lvs, natural infection, and with or without weekly mancozeb applications. Each hybrid and treatment was rated three times during the season for disease severity and harvested for grain yield. In 1980 losses for Minhybrid 7301 and W64A x W117 were 4.1, 12.8, 27.2% and 5.6, 13.4, 21.8%, respectively, for three disease levels while in 1981 they were 1.3, 19.3, 34.3% and 15.0, 27.4, 42.6%, respectively, over their corresponding naturally infected-mancozeb treatment. Irrigation is conducive to eyespot development in corn in Minnesota and significant yield losses may occur.

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EYESPOT, *Kabatiella zeae* PREVALENCE AND INTENSITY IN FIELD CORN AND ESTIMATED 1981 LOSSES IN MINNESOTA. P.S. Teng, W.C. Stienstra, R.L. Bowden and G.W. Randall, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

Losses caused by eyespot of corn were estimated by (1) a field test and (2) a survey of 43 important corn growing counties. Eyespot severity was assessed at tasseling, dough and full maturity on susceptible hybrid Mn 5201 when (1) inoculated, (2) not inoculated and (3) in plots adjacent to those inoculated. Three critical point models were developed for estimating yield loss (y) from eyespot severity per plant (x). At tasseling, $y = 1.23x$ ($R^2 = 0.90$). At dough, $y = 0.76x$ ($R^2 = 0.85$). At maturity, $y = 0.31x$ ($R^2 = 0.85$). Eyespot intensity data were gathered in 129 corn fields as part of a statewide pest and disease surveillance system. In August, eyespot was found in 12 counties at average severities from 0.1% to 13.5%. Corresponding estimated average losses per county were 0.07% to 10.3%. Average county loss due to eyespot was 0.38% (0.5% average severity). Average severities were greater in corn fields following corn than in fields following soybeans, alfalfa or wheat.

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AN ANALYSIS OF AREA WHEAT YIELDS AS INFLUENCED BY MULTIPLE DISEASES. M. V. Wiese, T. Herrman and M. Grube. Department of Plant and Soil Sciences, University of Idaho, Moscow, ID 83843.

Winter wheat at six sites within the Kootenai Valley in northern Idaho was protected against one or more diseases during 1981 by applying selective fungicides or timing fungicide sprays. Foot rot, powdery mildew, leaf rust, tan spot, sharp eye spot and stem rust were components of the resultant endemic disease syndrome in control plots. Selective control of foot rot, leaf rust and powdery mildew increased yields from 6115 to 7056, 6518 and 6182 kg/ha, respectively. A composite disease control treatment increased yield to 8064 kg/ha. Further estimates of the relative yield loss attributable to each disease were obtained from multiple regression equations built from crop yield and disease severity data. Specific manipulation and measurement of crop diseases coupled with yield measurements at multiple sample sites within a defined crop area permits identification of endemic diseases, description of their distribution and intensity quantitative approximation of their individual and collective impact on crop yield.

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ESTIMATE OF YIELD LOSS FROM CITRUS NEMATODE IN TEXAS GRAPEFRUIT. L. W. Timmer and R. M. Davis, University of Florida, IFAS, Agricultural Research and Education Center, Lake Alfred, FL 33850 and Texas A&I Univ. Citrus Center, Weslaco, TX 78596.

Control of the citrus nematode, *Tylenchulus semipenetrans* Cobb, has consistently increased grapefruit yields in Texas. In this

study, data from chemical control tests conducted from 1973 to 1980 were analyzed to determine the relationship between nematode counts, grapefruit yield and fruit size. The correlation between yield and nematode counts was negative ($r = -0.47$) and highly significant ($P = 0.01$). The data best fit the exponential decay curve: $y = 160.3e^{-0.0000429x}$ where y = yield in kg/tree and x = nematodes/100 cm of soil. There was no correlation between fruit size and nematode counts because yield and fruit size were inversely related. Yield loss in an average untreated orchard was estimated to be 12.4 metric tons/ha. Economic loss to citrus nematode in Texas grapefruit, assuming no treatment and an average on-tree price of \$60/metric ton, was estimated to be \$13.2 million annually.

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EFFECTS OF LEAF BLIGHT CAUSED BY *ALTERNARIA HELIANTHI* ON SUNFLOWER SEED YIELDS AND OTHER AGRONOMIC TRAITS. Carson, Martin L. Department of Plant Science, South Dakota State University, Brookings, SD 57007-1096

The effects of *A. helianthi* leaf blight epidemics initiated at different plant growth stages were studied in a field trial in 1981. Seed yield losses of 0-60% and 10-51% were observed in inoculated plots of the sunflower inbred lines cmsHA89 and Hybrid 894, respectively. Oil percentages and 100-achene weights were also significantly reduced in some inoculated plots, the reductions being less for Hybrid 894. Apparent infection rates were higher in plots inoculated at later growth stages than in those inoculated at earlier stages. The high correlation obtained between yield losses and disease severity ratings indicates development of disease loss assessment models is possible. The widespread distribution of *A. helianthi* in the major sunflower producing area of the United States and its demonstrated potential to reduce yields make it a concern for sunflower pathologists and breeders alike.

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DISEASE LOSSES OF YAM (*Dioscorea* spp.) IN PUERTO RICO. Mignucci, J. S.*, J. Green**, M. Cordero** and P. R. Hepperly*, *Depts. of Crop Protection and **Horticulture, Univ. of Puerto Rico, RUM, Mayaguez, 00708

Two major diseases were found attacking foliage of commercial yams in Puerto Rico. On 90% of farms growing *Dioscorea alata* (cv. Florida), foliage necrosis and defoliation from anthracnose ranged from 30 to 100%. Fungicide control experiments showed yield loss up to 79% with 91% loss in marketable yield. Anthracnose significantly decreased number of surviving plants (58%), number of tubers (61%), tuber weight per plant (58%) and number of marketable tubers (61%). On 35% of farms growing *D. rotundata* (cv. Habanero), virus diseases appeared limiting while anthracnose was not a problem. Virus symptoms included chlorosis, dwarfing of plants and shoestring leaves. By the time of planting, nearly half of all yams stored for seed were rotten. Storage losses were associated with *Penicillium* spp. and *Fusarium* spp. Habanero appeared more susceptible to tuber rotting than Florida.

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FEASIBILITY OF SOIL SOLARIZATION FOR PATHOGEN AND PEST CONTROL. G. S. Pullman, J. E. DeVay, C. L. Elmore, and W. H. Hart, Departments of Plant Pathology, Botany and Nematology, University of California, Davis, CA 95616.

Soil solarization tests were made at six CA field sites ranging from hot inland valleys to cool coastal areas during the summer of 1981. Plots were covered with 25 um clear polyethylene tarps, irrigated under the tarps, and solarized for 4 wk periods from May-Sept. or left fallow. Maximum soil temps of 42-51 C and 32-40 C occurred at depths of 15 and 46 cm, respectively, in plots solarized during June or July at all but the coolest site. Maximum air temps ranged from 26-45 C. Buried populations of plant pathogens (*Verticillium dahliae*, *Fusarium oxysporum* f. sp. *lycopersici*), weed seeds (*Convolvulus arvensis*, *Digitaria sanguinalis*, *Poa annua*, *Portulaca oleracea*), and nematodes (*Meloidogyne* spp.) were greatly reduced in most plots solarized in June or July; populations also decreased, although less, in May, Aug. & Sept. Cotton, sorghum and tomato seeds planted in a greenhouse in solarized soils (0-20 cm) germinated more rapidly and showed an increased growth response which was correlated to soil temps.

CHANGES IN MICROBIAL POPULATIONS IN SOLARIZED SOILS AS RELATED TO INCREASED PLANT GROWTH. J. J. Stapleton and J. E. DeVay, Dept. of Plant Pathology, Univ. of California, Davis, CA 95616.

Solarization of moist soils usually results in increased plant growth response (IGR), even when no pathogens are detected. Qualitative and quantitative changes in soil microflora have been observed following solarization. Isolations from roots of greenhouse-grown plants, however, revealed no significant quantitative differences of bacteria and fungi between solarized and nontarped treatments when plated on selective media. Qualitative differences of bacteria and fungi, based on colony morphology, were observed. Sugarbeet seed coated with plant growth promoting rhizobacteria (PGPR) showed no additional growth response when planted into a solarized soil in the greenhouse. However, population densities of the PGPR on roots in solarized soil were 46% higher than on roots in the nontarped soil ($P=0.05$). Covering tarped moist soil with sheetrock to prevent solar heating resulted in reduced population densities of some microorganisms ($P=0.01$), but no IGR occurred.

SOLAR HEATING FAILS TO CONTROL MACROPHOMINA PHASEOLINA. A. H. McCain, R. V. Bega, and J. L. Jenkinson. Department of Plant Pathology, University of California, and Pacific Southwest Forest and Range Experiment Station, Berkeley 94720.

Solar heating of moist soil beneath clear polyethylene covers is an effective method of controlling a number of important soilborne pathogens. The routine procedure in many California conifer nurseries is to fumigate the soil in the summer with methylbromide-chloropicrin mixture. We evaluated solar heating as an alternative to fumigation. Moist loam soil infested with *Fusarium oxysporum* f. *pini* and *Macrophomina phaseolina* was covered with 0.1 mm clear polyethylene from July 15 to August 9, 1981. Temperatures were monitored at 6 depths. *F. oxysporum* was eliminated from the solar heated soil down to the 10 cm depth, reduced at the 20 cm depth and survived at the 30 and 40 cm depths. *M. phaseolina* survived at all depths. The maximum temperatures recorded were: 0 cm - 63.6C, 2.5 cm - 58.8C, 10 cm - 56.2C, 20 cm - 39.6C, 30 cm - 37.2C, 40 cm - 36.4C.

ROOT INFECTION PATTERN, INFECTION EFFICIENCY AND MICROSCLEROTIAL DENSITY-DISEASE INCIDENCE RELATIONSHIPS OF CYLINDROCLADIUM CROTALARIAE IN PEANUT FIELD SOIL. G. J. Griffin and G. S. Tomimatsu, Dept. of Plant Path. & Phys., VPI & SU, Blacksburg, VA 24061.

The pattern of observed *C. crotalariae* infections on peanut roots gave a good fit to either the Neyman Type A or negative binomial distributions in 8 of 8 plots in a field for 1981. This indicated a clumped infection pattern on roots. Lloyd's index of patchiness (LIP) values for root infections ranged from 1.33 to 4.67. Efficiency estimates indicated that from 0.02 to 0.78% of the observed root infections resulted in shoot symptoms. Regression analyses (not forced through the origin) showed that there was a direct relationship between microsclerotial density and percent diseased plants ($R^2 = 0.91$) for the 8 plots. Correcting for multiple root infections [$\log_e (1/1-Y)$] increased the R^2 value to 0.95 ($ED_{50} = 0.7$ microsclerotia/g soil). R^2 values for the number of observed root infections per m root versus $\log_e (1/1-Y)$ were 0.88 and 0.57 for 7 of 8 and 8 of 8 plots, respectively. LIP was negatively correlated with percent diseased plants.

EFFECTS OF CROP ROTATION ON INOCULUM EFFICIENCY OF CYLINDROCLADIUM CROTALARIAE ON PEANUT. M. C. Black and M. K. Beute, Dept. of Plant Pathology, North Carolina State Univ., Raleigh 27650.

Field microplots were infested with microsclerotia (ms) of *Cylindrocladium crotalariae* at 35 ms/g of soil in the spring of 1979. All possible rotations of 'Pioneer 3368A' corn, 'Ransom' soybean, 'Floriant' peanut and 'NC3033' peanut were grown in 1979 and 1980. In January 1981 inoculum densities were 8.5, 14.5, 15.7 and 8.9 ms/g following corn, soybean, 'Floriant' and 'NC3033', respectively; and water soluble nitrogen was 5, 34, 15 and 15 mg/dm³, respectively, following the same crops. *Cylindrocladium* black rot susceptible 'Floriant' and resistant 'NC3033' were planted in 1981. Severity was lower ($P = 0.05$) on either peanut cultivar following corn or soybean than following 'Floriant' or 'NC3033'. Inoculum efficiency was inversely related to residual nitrogen following corn, soybean and 'Floriant'. Increased inoculum efficiency following 'NC3033' was apparently due to increased virulence since nitrogen residues were similar following 'NC3033' and 'Floriant'.

REDUCED ROOT GROWTH IN POTATOES WITH 'EARLY DYING' DISEASE. J. B. Kotcon, D. I. Rouse, and J. E. Mitchell. Dept. of Plant Pathology, University of Wisconsin-Madison, Madison, WI 53706. To assess the impact of *Verticillium dahliae* (Vd) on potato root growth, quantitative estimates of the size of root systems were obtained from cv. Russet Burbank in a field with 'Early Dying'. Roots were wet sieved from soil cores (10 cm diam. by 15 cm deep) and root lengths were estimated by the Newman Line Intercept Method. Five cores per plant sample were obtained from plots inoculated with Vd, fumigated with methyl-bromide, fumigated and inoculated, and untreated controls. Total root length peaked at 1480 m per plant in fumigated plots 105 days after planting (DAP). Root lengths in Vd inoculated plots averaged 21% less than in uninoculated plots 105 DAP ($P=.10$). Significant differences were not observed between treatments 56, 77, and 119 DAP when mean root lengths were 361, 895 and 864 m per plant, respectively. Root infection with Vd, *Chaetomium* spp., *Colletotrichum coccodes* and *Fusarium* spp., were correlated with root length only at 119 DAP ($r = -.60, .73, -.50, -.57$, respectively).

EFFECT OF HERBICIDES ON HATCHING AND EMERGENCE OF LARVAE FROM HETERODERA SCHACHTII CYSTS. Jack Altman and Arnold Steele. Colorado State University, Ft. Collins, CO 80523. USDA-ARS-WR, P.O. Box 5098, Salinas, CA 93915.

Certain herbicides applied at field rates to nematode infested soil resulted in greater numbers of *H. schachtii* larvae on sugar beets in greenhouse tests. This increased activity was accompanied by greater nematode damage to sugar beets. Beets grown in herbicide-treated, noninfested soil sustained less damage. Tests were established to evaluate hatching of *H. schachtii* larvae from cysts exposed to various concentrations of herbicides. Threefold increases in hatching occurred after exposure to 1.25, 2.50 and 5.00 ppm of Cycloate (S-ethylcyclohexylethylthiocarbamate) and 1.25, 2.50 and 5.00 ppm of Eptam (S-Ethyl dipropylthiocarbamate) compared to tap water controls. Zinc chloride (ZnCl₂) at 4 mmol/L used as a standard hatching stimulus increased larval emergence 25X compared to tap water. Work is continuing to determine if increased larval emergence causes greater host damage from feeding punctures and a subsequent increase in surviving cyst populations.

A HAWAIIAN SOIL SUPPRESSIVE TO PYTHIUM SPLENDENS. C. W. Kao and W. H. Ko, Department of Plant Pathology, University of Hawaii, Beaumont Agricultural Research Center, Hilo, HI 96720.

Only about 15% of *Pythium splendens* sporangia coated with cucumber root extract germinated on suppressive soil collected from South Kohala, whereas more than 90% germinated on conducive soil. When the suppressive soil was infested with *P. splendens* at concentrations of 1, 3, or 9 sporangia/ml soil, and planted with cucumber seeds, percentage seedlings killed ranged from 0 to 30%. However, more than 90% of the seedlings were killed in conducive soil inoculated with the same amounts of the pathogen. Suppressiveness decreased with increasing depth of soil. Germination of *P. splendens* sporangia increased from 6% on the topsoil to 97% on subsoil collected at a depth of 120 cm. Suppressiveness was reduced by amending soil with 500 ppm rose bengal, 10,000 ppm streptomycin-sulfate or 5,000 ppm benlate, and was overridden completely by steam treatment or autoclaving of soil for 15 min. Suppressiveness was restored to autoclaved soil by inoculation with microorganisms isolated from suppressive as well as conducive soils.

NATURE OF HYPOXYLOID STROMATA OBSERVED ON MACADAMIA ROOTS AND TRUNKS INFECTED BY KRETZSCHMARIA CLAVUS. W. H. Ko, W. C. Ho, and R. K. Kunimoto, Department of Plant Pathology, University of Hawaii, Beaumont Agricultural Research Center, Hilo, HI 96720.

Ascomyces from hypoxylid stromata observed frequently on macadamia roots and trunks infected by *Kretzschmaria clavus*, were similar in shape and size to those from typical *K. clavus* stromata. Colonies derived from ascomyces of hypoxylid stromata and kretzschmaroid stromata were morphologically indistinguishable. Conidia produced by isolates from typical kretzschmaroid stromata and from hypoxylid stromata were similar in shape and size on sterilized stem tissues. Anastomoses occurred between hyphae derived from propagules from these

two types of stromata. Isolates from kretzschmaroid stromata produced both kretzschmaroid and hypoxylid stromata on litchi stem tissues. Results strongly suggest that the hypoxylid stromata observed on diseased macadamia tissues are morphological variants of the more familiar *K. clavus* stromata.

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FACTORS AFFECTING SUDDEN WILT OF MELON IN CALIFORNIA. W. D. Gubler and R. G. Grogan, Department of Plant Pathology, University of California, Davis, CA 95616

Sudden wilt of melon in California is induced by at least two species of *Pythium*; *P. ultimum* and *P. aphanidermatum*. *P. ultimum* and to a lesser extent *P. aphanidermatum* have been consistently isolated from nearly mature melon plants displaying sudden, rapid wilting. Both species have caused damping off of seedlings in greenhouse tests but certain stresses are required for the reproduction of the sudden wilt syndrome of mature plants. Two stress factors appeared to be of primary importance; 1) a fruit load and 2) a water deficit stress followed by flooding. Typical symptoms were reproduced on the cultivars Honeydew and Crenshaw only when plants were subjected to the moisture deficit and excess stresses following fruit set. Non-inoculated control plants and inoculated, fruitless plants showed no symptoms of sudden wilt when subjected to the same predisposing moisture stresses.

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INFLUENCE OF *PSEUDOMONAS PUTIDA* AND SYNTHETIC IRON CHELATES ON *FUSARIUM* WILT DISEASES. Fran M. Scher and Ralph Baker, Dept. of Botany and Plant Pathology, Colorado State University, Fort Collins, Colorado. 80523

A fluorescent *Pseudomonas putida*, isolated from *Fusarium*-suppressive soil, significantly reduced the incidence of flax, radish, and cucumber wilt when added at 10^7 cells/g of *Fusarium* oxysporum-infested conductive soil in pot experiments. Addition of 100µg FeEDTA/g soil nullified the biological control. Introduction of 50-100µg EDDHA or FeEDDHA/g of *Fusarium*-infested conductive soil decreased wilt of all three hosts. Disease control by FeEDDHA and *P. putida* was additive. Germ tube elongation of *Fusarium oxysporum* f. sp. lini microconidia on water agar was inhibited by EDDHA; the inhibition was reversed by addition of FeCl₃. This indicated that iron was a limiting factor for germ tube elongation. *P. putida* and various fusaria produced siderophores in a low iron medium. It is suggested that the mechanism of disease control by *P. putida* and EDDHA is related to iron competition in soil.

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BACTERIZATION OF POTATO SEED FOR CONTROL OF VERTICILLIUM WILT OF POTATO. Jawad A. Wadi and Gene D. Easton. Washington State University, TAREC, Prosser, WA 99350.

Several strains of bacteria isolated from plant rhizospheres and *Verticillium microsclerotia* inhibited *Verticillium dahliae* in vitro. In greenhouse studies, these strains coated on potato seed (bacterization) multiplies in *Verticillium* infested soil, inhibited *V. dahliae* in the rhizosphere, reduced *Verticillium* wilt, and increased fresh weight of potato roots, plants, and tubers. In the field, bacterization decreased *V. dahliae* in the soil and decreased *Verticillium* wilt equal to soil fumigation with Telone C-170 (257 l/ha). The bacteria did not improve yields but they did increase tuber quality.

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CHARACTERIZATION OF THE COMPONENT PRODUCED BY *BACILLUS SUBTILIS* WHICH INHIBITS BEAN RUST. C. J. Baker, J. S. MacFall, and J. R. Staveland. 1/ USDA, PPI, APPL, Beltsville, MD 20705 and 2/ Univ. of Delaware, Newark, DE 19711.

Culture filtrates of *B. subtilis* sprayed onto bean plants greatly reduced the number of uredia incited by all races of *Uromyces phaseoli* tested. This reduction was nonspecific and was obtained using different bean varieties and pathogenically unique rust collections. Microscopic studies of treated bean leaves indicated uredospore germination was reduced by more than 95%, and germ tubes which did develop were abnormal. The inhibitory component present in the culture filtrate was found to be nondialyzable and heat stable. Dialyzed culture filtrate was subjected to preparative gel filtration.

Fractions with an apparent MW of 3,000 to 5,000 daltons were inhibitory in bioassays that measured rust severity and germination of uredospores. This fraction contained approximately 5% carbohydrate and 95% protein.

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ROLE OF ANTIBIOSIS IN ANTAGONISM AGAINST ICE NUCLEATION ACTIVE BACTERIA BY EPIPHYTIC BACTERIA. S. E. Lindow. Dept. of Plant Pathology, University of California, Berkeley, CA 94720.

Sixty epiphytic bacterial strains have been isolated that reduced subsequent colonization of ice nucleation active (INA) strains of *Pseudomonas syringae* pv. *syringae* and *Erwinia herbicola* on leaf surfaces. Only 35 of these isolates produced compounds inhibitory to these species in vitro. Mutants of 21 of 23 of these strains which no longer produced either fluorescent or non-fluorescent inhibitory compounds in vitro did not differ significantly from wild type strains in prevention of growth of *P. syringae* on corn leaves. Mutants of all 23 strains reduced subsequent colonization of corn leaves by *P. syringae* significantly compared with non-preinoculated plants. All nineteen ice nucleation deficient mutants of three *P. syringae* strains reduced frost injury significantly at -5°C to corn seedlings when applied prior to ice nucleation active wild type strains. Thus, production of inhibitory compounds is not of primary importance in antagonism among epiphytic bacteria.

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PARASITISM OF THE NEMATODE *CRICONEMELLA XENOPLEX* BY THE FUNGUS *HIRSUTELLA RHOSSILIENSIS* AS INFLUENCED BY NEMATODE STAGE, HEAT TREATMENT, AND SOLUTE POTENTIAL. B. A. Jaffee and E. I. Zehr. Department of Plant Pathology and Physiology, Clemson University, Clemson, SC 29631.

In the laboratory, juvenile and adult *Criconemella xenoplax* were touched to the adhesive spores of *Hirsutella rhossiliensis* (20 spores/adult, 10 spores/juvenile) and incubated in sterile, distilled water for 5 or 10 days. Noninoculated nematodes were controls. Although adults were resistant to fungal penetration, 25% of inoculated juveniles were penetrated and killed by the fungus. Fifty percent of inoculated adults incubated in sterile, distilled water were invaded and killed if heated to 40°C for 120 min prior to inoculation. However, nearly 100% of nonheated, inoculated adults were invaded if incubated for 5 days in a dilute KCl solution (6.4×10^{-3} molal = -0.31 bars). Results suggest that nematode condition and solute potential of medium affect parasitism of *C. xenoplax* by *H. rhossiliensis*.

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HYPERPARASITIC FUNGI OF CORNSTALKS, Nader G. Vakili, ARS/SEA/USDA and Department of Plant Pathology, Iowa State University, Ames Iowa 50011

Eight mycoparasitic fungi were recovered from corn pith in association with the complex of fungi causing stalk rot of corn. These were *Sphaeronaemella helvella*, *Exobasidiellum* sp., *Trichoderma viride*, *Gliocladium roseum*, *Gonatobotrys simplex*, *Trichothecium roseum*, *Pythium acanthicum*, and a species of *Mycelium* *sterilium*. *Fusarium moniliforme* and *Helminthosporium carbonum* had the highest frequencies of association with mycoparasitic fungi while *F. graminearum* and *Cladosporium herbarum* were only occasionally infected. *S. helvella* caused bleaching of *Helminthosporium carbonum* but induced pigment production in *Fusarium moniliforme*. *Pythium acanthicum* formed parthenospores within conidia of *H. carbonum* and was a destructive pathogen of this fungus, as were *G. roseum* and *T. roseum*, and *Exobasidiellum* sp. *Gonatobotrys simplex* was associated only with the *Fusaria*. *T. viride* occurred mainly in roots and crown nodes. Bio-control through breeding for mycovirulence is suggested as a complement to breeding for stalk rot resistance and for stiff stalk in reducing losses due to stalk lodging in corn.

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ENDEMIC DISEASES OF AQUATIC WEEDS IN CALIFORNIA. E.A. Bernhardt and J.M. Duniway, Department of Plant Pathology, University of California, Davis, CA 95616

A survey was conducted in Northern California during 1980 and 1981 for endemic diseases of aquatic weeds that might have potential for biological control. A decline of the emerged weed parrotfeather (*Myriophyllum brasiliense*) was characterized by severe rot of underwater roots and stems. A *Pythium* sp. was isolated and was pathogenic on parrotfeather when inoculated with zoospores in solution culture. Dormant overwintering structures of the pondweeds *Potamogeton crispus* and *P. nodosus* were frequently rotted when collected from soil in drained ir-

rigation canals in winter. *Fusarium lateritium*, *Burgoa* sp., *Fusidium* sp., *Pythium* sp., and a binucleate *Rhizoctonia* sp., isolated from rotten overwintering structures, colonized healthy structures when they were inoculated with mycelium and incubated at 6 or 9°C. A significant amount of rot occurred in overwintering structures of *P. crispus* under field conditions following inoculation with debris infested with *Fusarium lateritium* and *Burgoa* sp.

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A NEW TECHNIQUE FOR DELIVERY OF BIOLOGICAL AGENTS WITH GERMINATED VEGETABLE SEED. K. E. Conway², C. G. Fisher¹, J. E. Motes², Depts. of Plant Pathology¹ and Horticulture and Landscape Arch.², Oklahoma State University, Stillwater, OK 74078.

Several techniques have been utilized for delivering biological control agents to soil for control of soil-borne pathogens (Phytopathology 70:1167-1172, 1980). Carriers such as diatomaceous earth granules, rye seed and wheat bran were employed and required applications of large quantities of material into the field. *Trichoderma hamatum* was used successfully on radish and pea seed to control *Pythium* spp. and *Rhizoctonia solani*. However, other biocontrol agents were not able to colonize seed coats. The use of seeding gels for fluid drilling of small seeds is becoming an accepted procedure. We are using a seeding gel, Laponite 508, (Laporte Ind. Ltd., U.K.) to deliver quantities of *T. harzianum* and *T. hamatum* conidia concomitantly with germinated vegetable seeds. Growth chamber and field studies are in progress to evaluate this technique for control of soil-borne diseases of Bahemian hot chili pepper and asparagus.

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SCLEROTINIA SCLEROTIUM: AN ENDEMIC BIOLOGICAL CONTROL AGENT FOR CANADA THISTLE. Brenda Simmonds-Brosten*, P. K. Fay, and D. C. Sands*, *Dept. of Plant Pathology, Montana State Univ., Bozeman, MT 59717 and Dept. of Plant and Soil Science, Montana State University, Bozeman, MT 59717.

A survey of endemic pathogens of Canada thistle was conducted during the summer of 1981. Pathogens from 5 genera, *Alternaria*, *Fusarium*, *Septoria*, *Pseudomonas* and *Sclerotinia* were obtained. Of these pathogens, *Sclerotinia sclerotiorum* (S.s.) best met the criteria of lethality and ease of culture. Strains of this pathogen were cultured on autoclaved oat kernels which were then dried and then used as inoculum loci. In the greenhouse and in the field, strains of S.s. differ greatly in host range and in virulence. Highly virulent strains capable of killing over 90% of inoculated plants under high moisture conditions in the greenhouse, killed only 30% of the plants in the field under low moisture conditions in the fall. Yet to be solved are problems of host range restriction and determining the optimal method of inoculation.

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STUDIES ON OROBANCHE SPP.: FUNGI ASSOCIATED WITH OROBANCHE CRENATA FORSK. Osama A. Al-Menoufi, Plant Pathology Dept., Alexandria University, Egypt.

Broomrape (*Orobancha crenata*) has been recognized as a destructive parasite of broad bean and other cultivated crop plants in Egypt and several other countries. Isolation from rotted fruits of broomrape yielded *Fusarium oxysporum* Schlecht, *F. solani* (Mort) Sacc., *Alternaria* sp. and *Sclerotinia* sp. The rotted fruits did not open and the seeds failed to disseminate. It was found that the punctured fruits were more susceptible to infection by these fungi than the intact ones. Infection of the broomrape fruits by these fungi significantly decreased the germination percentage of their seeds. The thermal requirements of these pathogenic fungi, for growth and sporulation, ranged from 25-35°C. It was also found these fungal isolates were non-pathogenic on winter crops of broad bean, tomato, flax and wheat in Egypt.

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EFFECT OF VANGARD ON THE CONTROL OF POWDERY MILDEW OF APPLE. Robert H. Daines, Eileen Bunderson, and Darrell Weber. Brigham Young University, Provo, Utah 84602.

Vanguard [1(2-(2,4-dichlorophenyl)-4-ethyl-1,3 dioxolan-2-yl)methyl]-1H-1,2,4 triazole] effectively controlled powdery mildew [*Podosphaera leucotricha* (Ell. and Ev.) Salm.] infection of leaves and fruit russetting when applied at 3 1/2 oz per 100 gal with a regular spray schedule. The treatment was effective with Rome and Jonathan apple trees. Scanning and transmission

electron microscope studies indicated desiccation and membrane disruption in the Vanguard treated powdery mildew fungus. Translocation studies with ¹⁴C Vanguard indicated that 70% of the label remained at the site of application with 30% being translocated to other parts of the tree. Vanguard was more effective in controlling leaf infection as compared to spray programs with two other fungicides, benomyl and Karathane. The effectiveness of these three fungicides has been correlated with the effect of temperature and humidity in relationship to powdery mildew infection over a three year period.

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LOCAL ZONE TREATMENT BY VANGARD VAPOR ACTION FOR CONTROL OF POWDERY MILDEW. Michael Szkolnik, Dept. of Plant Pathology, New York State Agric. Expt. Sta., Geneva, NY 14456.

Local zone (LZ) vapor phase action against powdery mildew is here defined as effective control limited to a small area of a room. This differs from the reported room-wide protection by vapor from Vanguard (CGA 64251) impregnated into cheesecloth and other materials. Each LZ treatment was by impregnation of 30-cm Hercules[®] olfin fiber braided cord with 50-mg CGA 64251 99.5% Tech. in 5-ml methanol. After evaporation of the methanol, the cord attached to a rigid wire was set vertically and centrally among a group of potted plants which were inoculated the day of treatment and maintained in a greenhouse at 23 C and above 50 RH. Vanguard vapor provided very good to complete protection for 60 cm and further from the treated cord against powdery mildews of apple, *Podosphaera leucotricha*; bean, *Erysiphe polygoni*; cucumber, *Sphaerotheca fuliginea*; and rose, *S. pannosa*. Because of prevailing air currents in the greenhouse the local zone of control was elliptical. Beyond the zone, powdery mildew flourished.

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INFESTATION AND INFECTION OF APPLE BUDS BY BOTRYOSPHERA OBTUSA Myra B. Beisel and Floyd F. Hendrix, Dept. of Plant Pathology, University of Georgia, Athens, GA 30602

Infection of apple buds by *Botryosphaeria obtusa* in Georgia is usually of conidial origin. Ascospores were rarely collected in aerial spore traps at the Blairsville Experiment Station maintained from 1978-1982. Conidia of *B. obtusa* which are produced on dead wood and extruded from pycnidia, are adapted primarily to dispersal by splashing rain. Thus the primary inoculum source for bud infestation and infection is located in or under each tree. If weather conditions suitable for conidial discharge occur, bud infestation levels as high as 88% may be reached as early as December. However, infection does not occur until bud swell and requires adequate moisture. The apple spray guide for Georgia currently recommends that black rot control begin at the early pre-pink stage of bud development. Monthly bud surveys at 16 orchards throughout North and Middle Georgia during December-March of 1980-1982, suggest that protectant sprays for black rot control in Georgia should be applied 2-3 weeks earlier, at bud swell, to prevent initial infection.

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EFFECT OF RUBIGAN[®] (FENARIMOL) ON POLLEN GRAIN VIABILITY. I. F. Brown and H. R. Hall, Eli Lilly and Co., Greenfield, IN 46140.

Studies were conducted to determine the influence of fenarimol and other fungicides upon the viability of pollen grains of several types of deciduous fruit. The relative toxicity of the fungicides to pollen grains was demonstrated by measuring pollen grain germination and germ tube development when placed on medium amended with different rates of the fungicides. Captan greatly reduced the germination of pollen grains from apple, cherry, peach and pear blossoms. Fenarimol, nuarimol, triadimefon and etconazole had little or no effect upon pollen grain germination. Similarly, germ tube development of pollen grains was drastically reduced by captan but only partially by fenarimol or the other ergosterol biosynthetic inhibitors evaluated. Fenarimol and captan were applied to the open blossoms of apple, cherry and pear in a field experiment. Pollen grains from treated and untreated blossoms were dusted onto nonamended agar medium. Fenarimol had no influence upon the germination or germ tube development of any pollen type. Captan was detrimental to both processes.

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QUANTITATIVE MODIFICATIONS IN BANANA FIELD SURVEYS TO IMPROVE SPRAY TIMING AND CONTROL OF BLACK SIGATOKA, *MYCOSPHAERELLA FIJIENSIS* VAR. *DIFFORMIS*. C. F. Robert, A. Lopez & R. H. Fulton. Rohm & Haas Centro America, S.A. Apartado 3908, San Jose, Costa Rica. During the last decade, the youngest leaf spotted (YLS) and per-

cent of plants spotted on non-flowered banana plants has been the standard field survey system used to measure disease intensity and prevalence of Yellow Sigatoka, *M. musicola*. Whereas, Black Sigatoka, a more virulent strain, evoked studies to sensitize surveys to take into account the rapid disease cycle. Sigatoka spots are noted with ease in the field on plants in fruit. Leaf surveys were made on these flowered plants by age of fruit (emergence to maturity) at weekly intervals employing the parameters of YLS, spotting classified by grade and total number of leaves. Regression analysis verified the distinct separation in intensity-grades of infection on plants 60 to 63 days after flowering. It is evidenced that this plant type and parameters employed be incorporated in Black Sigatoka surveys. This as a tool for spray timing prior to and during climatic disease pressure periods and similarly for the perennial problem zone areas.

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RESURGENCE OF TOLERANCE TO BENOMYL IN *MYCOSPHAERELLA FIJIENSIS* VAR. *DIFFORMIS*. T. L. Woods, L. Jácome, and R. H. Stover
Division of Tropical Research, United Fruit Company, La Lima, Honduras.

Following widespread use of the systemic fungicide benomyl from 1974 to 1977 for control of black Sigatoka disease of bananas (*Mycosphaerella fijiensis* var. *difformis*) in Honduras, tolerance to the fungicide up to 300 ug/ml developed with resultant loss in disease control. Benomyl was not applied during 1978-1980 and tolerance levels dropped to 10 ug/ml. From June, 1981 to January, 1982 benomyl was reintroduced but used only in combination with oil plus mancozeb alternated with sprays of mancozeb alone to suppress development of tolerant strains. By February 1982 tolerance levels had increased, in some cases up to 200 ug/ml. Reintroduction of a systemic fungicide, to which tolerance had declined, in combination with a protectant, was unsuccessful in the suppression of tolerant strains.

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COMPETITION BETWEEN CONIDIAL ISOLATES OF BENOMYL RESISTANT AND BENOMYL SENSITIVE *MONILINIA FRUCTICOLA* ON PEACH FRUIT. Ronald M. Sonoda, University of Florida, IFAS, Agricultural Research Center, Fort Pierce, FL 33450; J. M. Ogawa and P. L. Sholberg, University of California, Davis, CA 95616

When conidial suspensions of distinct biotypes of *Monilinia fructicola* were mixed in equal proportions and inoculated onto injured peach fruit in the laboratory, they were found to co-exist at the margin of some lesions but in others only one biotype was detected. When 9 benomyl-resistant and 12 benomyl-sensitive isolates of *M. fructicola* were paired and inoculated onto injured peach fruit, sensitive isolates predominated in 232 of 278 lesions. Furthermore, if conidia of a resistant and a sensitive biotype with similar growth rates on potato dextrose agar were equally mixed and inoculated on peach agar the sensitive isolate became predominant. When pairs of biotypes were mixed unequally the biotype in greater concentration usually became more dominant in the resulting lesions. However, in one case a resistant biotype failed to predominate even at a 100 fold higher concentration.

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CONTROL OF *PHYTOPHTHORA* HEART ROT OF PINEAPPLE WITH METALAXYL AND PHOSETHYL AL. S. Schenck and K. G. Rohrbach. Dole Pineapple Co., P. O. Box 490, Wahiawa, HI 96786 and Department of Plant Pathology, University of Hawaii, Honolulu, HI 96822.

Heart rot of pineapple caused by *Phytophthora parasitica* or *P. cinnamomi* was almost completely prevented by metalaxyl (Ridomil®) or phosethyl Al (Aliette®). Pineapple propagative materials were dipped in solutions of the fungicides before planting in *Phytophthora*-infested areas. Both fungicides were tested at 600 ppm, 1200 ppm, and 2400 ppm. In the phosethyl Al test, mortality at four months after planting in the untreated plots was 70%; whereas in the treated plots the mortality was 11%, 1.7%, and 0 respectively. Untreated plots in the metalaxyl test had a mortality level of 76%. Mortality with 600 ppm was 24%, 13.9% with 1200 ppm, and 1.9% with 2400 ppm metalaxyl. Several other tests with various mortality levels gave similar results. Although the occurrence of pineapple heart rot is unpredictable, preplant dip treatments of phosethyl Al at 1200 ppm or metalaxyl at 2400 ppm gave excellent control for 4 to 6 months after application even under severe conditions.

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RESEARCH WITH FENARIMOL FOR CONTROL OF GRAPE POWDERY MILDEW IN CALIFORNIA VINEYARDS. Donald H. Ford, Lilly Research Laboratories, 7521 W. California Ave., Fresno, CA 93706

Fenarimol, a member of the pyrimidine carbinol class of sterol-inhibiting fungicides, demonstrates excellent control of grape powdery mildew (*Uncinula necator*) and has found wide commercial acceptance in several important grape growing regions of the world. Under field evaluation in California vineyards, fenarimol IEC at 10 to 25 G/A foliar applied 3 to 4 times at 10- to 21-day intervals to raisin, table and wine grapes (*Vitis vinifera*) gave 95 to 100 percent powdery mildew control under low to moderate disease pressure. Applications were made 10 to 14 days prior to bloom, again during bloom and once or twice following fruit set. Fenarimol was successfully applied with ground operated concentrate sprayers at 20 to 25 gallons per acre (gpa) and high pressure, dilute sprayers at 150 to 300 gpa. The fungicide has also given promising results when applied by air at 15 to 20 G/A in a spray volume of 10 to 15 gpa. When applied at normal use rates alone or in tank-mix combinations with other vineyard chemicals, fenarimol has demonstrated good safety on major California grape varieties with no evidence of reduced yield or quality of table, raisin or wine grapes. Successful use of fenarimol for powdery mildew control in California vineyards could ultimately replace the presently accepted practice of applying dusting sulfur to the vines at frequent intervals during the growing season.

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THE CAUSAL RELATIONSHIP OF CHERRY LEAFROLL VIRUS AND THE BLACKLINE DISEASE OF ENGLISH WALNUT TREES. S. M. Mircetich, and A. Rowhani, USDA, ARS, Department of Plant Pathology, University of California, Davis, CA 95616.

To establish unequivocally by Koch's postulates the causal relationship of cherry leafroll virus (CLRV-W) and the blackline disease we artificially inoculated 1-year-old healthy English walnut trees on Paradox or *Juglans hindsii* rootstock with purified CLRV-W. The CLRV-W in PO₄ buffer was applied with a glass rod to the cambium under a bark flap (1 x 2 cm) of the scion, then wrapped with grafting tape. Controls received PO₄ buffer. Eleven of 13 trees inoculated with purified CLRV-W developed the blackline symptoms within 18 months. No blackline developed in 10 trees that received only PO₄ buffer. CLRV-W serologically identical to the CLRV-W isolate that was used as the inoculum was recovered only from the inoculated walnut trees that had blackline at the graft union. Our results showed that CLRV-W is the causal agent of the blackline disease of English walnut trees.

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SEED AND POLLEN TRANSMISSION OF CHERRY LEAFROLL VIRUS (CLRV-W), THE CAUSAL AGENT OF THE BLACKLINE DISEASE (BL) OF ENGLISH WALNUT TREES. S. Mircetich, A. Rowhani and J. Cucuzza, USDA, ARS, University of California, Davis, CA 95616

We consistently detected CLRV-W in pollen and nuts from BL-affected English walnut orchard trees by ELISA and bio-assays. CLRV-W was detected in 3-5% of 3-month-old English walnut seedlings among 894 seedlings grown from nuts of 6 naturally infected trees. To determine the role of pollen in transmission of CLRV-W, immature pistillate flowers on healthy English walnut trees were covered and then, when in the most receptive stage, hand pollinated with CLRV-W infected or CLRV-W free walnut pollen; 40-50 nuts resulting from controlled pollination on each of 3 different cultivars were assayed by ELISA 12 weeks after pollination. The percentage of CLRV-infected nuts resulting from CLRV-W infected pollen were: Payne cv. 14%, Eureka cv. 8%, and Vina cv. 9%. CLRV-W was not detected in the nuts from the same trees that resulted from CLRV-W free pollen. Apparently CLRV-W is transmitted through seed and pollen from BL-affected English walnut trees.

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IDENTIFICATION AND DISTRIBUTION OF THREE *PRUNUS* NECROTIC RING-SPOT VIRUS SEROTYPES IN WASHINGTON SWEET CHERRY ORCHARDS. G.I. Mink, Anabel Cole, and S. Regev. Washington State University, IAREC, Prosser, WA 99350.

Three serotypes of *Prunus* necrotic ringspot virus (NRSV) identified by spur formation in agar gel plates were isolated from rugose mosaic diseased sweet cherry trees in Washington and designated CH-3, CH-9, and CH-30. CH-3 and CH-9 antigens were detected by ELISA with equal sensitivity using any combination of anti CH-3, CH-9, or Fulton's NRSV-G gamma globulins and con-

jugates. However, none of these combinations detected CH-30 antigens. Anti CH-30 gamma globulins and conjugate did not detect CH-3 or CH-9 antigens. All NRSV isolates obtained from cherry orchards where rigose mosaic disease has appeared within the past 10 years were CH-9 serotype, as were all NRSV isolates obtained from bee hives entering Washington from California. Serotypes CH-3 and CH-30 have been obtained from only a few diseased trees.

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DISTRIBUTION OF TOMATO RINGSPOT VIRUS IN DANDELION IN PENNSYLVANIA. C. A. Powell, W. L. Mountain, L. B. Forer, L. D. Lathrop, and R. F. Stouffer. PA Dept. Agric., Harrisburg, PA 17110. *Xiphinema* spp. density, dandelion density, and relative infection of dandelion with tomato ringspot virus (TmRSV) was determined in 8 peach orchards with TmRSV-induced disease (Prunus stem pitting; PSP), 8 peach orchards without PSP, and 8 non-orchards in 8 geographic regions of PA representing both intensive and non-intensive peach production areas. There was no difference ($p=.05$) in *Xiphinema* or dandelion density among types of orchard site or geographic regions. The percentage of dandelion infected with TmRSV was higher ($p=.05$) in orchards with PSP (29%) than in orchards without PSP (7%) or non-orchards (5%) and in intensive peach production areas (21%) than in non-intensive peach production areas (7%). There were two geographic regions in which no infected dandelion was detected. We conclude that TmRSV is not yet endemic in dandelion throughout Pennsylvania. TmRSV is probably initially introduced into an orchard via infected nursery stock or dandelion seed and subsequently becomes established in dandelion and other weeds over a period of years.

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EVIDENCE FOR AN UNKNOWN LATENT VIRUS IN STRAWBERRY. R. R. Martin and R. H. Converse. Dept. of Bot. & Pl. Path. Oregon State Univ. (USDA-ARS) Corvallis OR 97331 (BARD Grant US 156-79)

While attempting to purify strawberry mild yellow-edge virus (SMYEV), a clone of strawberry cv. Benton that was free of known viruses by standard indexing techniques was used as a control. Clarified extracts from these control plants, when subjected to rate-zonal density gradient centrifugation, yielded a UV-absorbing fraction that contained viruslike particles. In subsequent tests these particles (28 nm) co-sedimented with those of SMYEV. Repeated leaf graft indexing from this Benton clone to strawberry virus indicator clones Alpine, UC-4, 5, 6, 10, 11 and 12 failed to produce any symptoms, although 28 nm particles were found in grafted UC-10 and 12 after subsequent purification. This latent virus may be a mild strain of a described virus from strawberry, like SMYEV, or may be a new virus not detectable with currently used indicators. Five of six strawberry cultivars subsequently tested were found to contain similar virus-like particles, whereas the virus-indicator clones checked were free of such particles.

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ALFALFA MOSAIC VIRUS IN CALIFORNIA FRESH MARKET TOMATOES. D. A. Knorr, F. F. Laemmlen, and W. O. Dawson. Department of Plant Pathology, University of California, Riverside, CA 92521.

Tomato disease in the Imperial Valley of California was shown to be caused by a necrotic strain of alfalfa mosaic virus (AMV). Virus was associated with localized necroses in diseased plants, resulting in bronze coloration, severe stunting, and death due to phloem collapse. Alfalfa was a reservoir of AMV. Surveys of alfalfa fields showed AMV was less than 5% seed transmitted, but spread to 70-90% of the plants in 3-4 years. Disease severity in tomato was associated with proximity to alfalfa 3 years or older with incidence often 30-40% in tomatoes closest to alfalfa (ca. 30 m), but less than 2% at 150 m. Two aphids, alfalfa, *Acyrtosiphon pisum* and *A. kondoi*, were shown to transmit AMV to tomatoes in a stylet-borne manner. Introduction of *A. kondoi* into the area apparently parallels the increase of this disease. Field and greenhouse trials indicated commercial tomato varieties vary considerably in tolerance to the disease. Presently, it is impractical to control the virus reservoir or vectors, therefore, tolerant tomatoes should be planted within 150 m of alfalfa.

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LUCERNE TRANSIENT STREAK-A VIRUS OF ALFALFA NEWLY RECOGNIZED IN NORTH AMERICA. Y. C. PALIWAL, Chem. and Biol. Res. Inst., Agriculture Canada, Ottawa, Canada.

A virus isolated from alfalfa plants showing scattered yellow streaks and some deformation of leaves in Ontario during cooler

seasons was identified as lucerne transient streak virus (LTSV)-first record outside of Australia and New Zealand. The virus seems to be sporadic throughout Ontario and western Quebec (0-27% incidence). The Canadian isolate of LTSV failed to infect several known hosts of the virus but several economic plants were newly recognized as hosts. The virus purified from *Trigonella* by acidification, differential and sucrose density gradient centrifugation sedimented as one peak at 112 S, had an $R_0.1^{18} = 5.4$ and buoyant density in CsCl of 1.365 g cm^{-3} . LTSV particles (28 nm diameter) contained a protein of mol. wt. c. 32,000 and its infectious RNA contained a covalently bound protein essential for infectivity. The virus was strongly immunogenic giving an antiserum with a titre of 1/2048 (agar diffusion). LTSV did not react with antisera to southern bean mosaic and cocksfoot mottle viruses.

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A MOSAIC DISEASE OF GUINEA GOLD VINE CAUSED BY CUCUMBER MOSAIC VIRUS. A. E.-S. A. Fudl-Allah, L. G. Weathers, and F. C. Greer Jr. Department of Plant Pathology, University of California, Riverside, CA 92521.

A mosaic disease of *Hibbertia volubilis* was found in several nurseries in California. Naturally infected plants showed mosaic, chlorosis, stunting, and flower abortion. Partially purified virus was sap transmitted to leaves of *Amaranthus caudatus*, *Beta vulgaris*, *Cucumis sativus*, *Chenopodium quinoa*, *Nicotiana glutinosa*, *N. benthamiana*, *Vigna sinensis*, and *Saponaria vaccaria*. Virus was not easily transmitted from infected *Hibbertia* to other test plants due to an inhibitor. Transmission was observed only in *N. glutinosa*. Symptoms in *C. quinoa*, *A. caudatus*, and *V. sinensis* consisted of primary local lesions. Plants of *N. glutinosa*, *N. benthamiana*, *S. vaccaria*, *B. vulgaris*, and *C. sativus* showed systemic mosaic with malformation of leaves. Symptoms in *N. glutinosa* and *C. sativus* were typical for cucumber mosaic virus. Electron microscopy of partially purified preparations from infected plants revealed isometric particles of about 30 nm in diameter.

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TABACCO MOSAIC VIRUS STRAINS INFECTING GESNERIADS. F. W. Zettler and J. Nagel. Plant Pathology Department, University of Florida, Gainesville 32611.

Serological and electron microscopic (leaf dips stained in 2% uranyl acetate) techniques detected tobacco mosaic virus (TMV) in 3 collections of cultivated gesneriads (Gesneriaceae). TMV was found in 2/4 *Kohleria*, 2/14 *Nematanthus*, and 24/49 *Columnea* samples assayed. Infected plants had inconspicuous symptoms. The 7 *Saintpaulia* and 10 *Aeschynanthus* plants sampled were not infected. Reciprocal immunodiffusion tests (medium with 0.8% agar, 0.5% sodium dodecyl sulfate, and 1% Na₂S₂O₃) using antisera to the U₂, common, and dahlemense strains of TMV showed the U₂ strain to predominate (66 of 67 samples). One sample ('Rongo' *Kohleria*) was infected with the common strain, and no infections by the dahlemense strain were detected. These results suggest an association of TMV infections in gesneriads with cigarette tobacco (Wetter & Bernard. 1977. *Phytopathol.* Z. 90:257-267). Antisera against a gesneriad isolate of TMV ('Oneidan' *Columnea*) reacted identically to the U₂ strain but heterologically against the common and dahlemense strains.

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PRELIMINARY STUDIES ON A RHABDOVIRUS CAUSING A NEWLY-IDENTIFIED DISEASE OF TOMATOES IN MOROCCO. El Maataoui, Mohamed, Inra, Station de Phytologie, B.P. 415, Rabat, Morocco and B.E.L. Lockhart, Complexe Horticole, B.P. 438, Agadir, Morocco.

A new virus found on tomatoes in three areas in Morocco causes vein-clearing, top necrosis, malformation of leaves and fruit spotting. It was transmissible to several solanaceous species of test plants and *Gomphrena globosa*. Bacilliform particles 360/80 nm were observed in leaf-dip and partially purified preparations stained with ammonium molybdate or uranyl formate. It was partially purified from infected *Nicotiana rustica* by extraction in 0.2 M sodium citrate containing 0.01 M magnesium chloride, clarified by celite filtration, and concentrated by discontinuous sucrose gradient centrifugation. The tomato rhabdovirus resembled potato yellow dwarf virus (PYDV) in particle morphology and in symptoms produced on several test plants, but not in symptoms on potato cultivar Bintje. Preliminary serological tests with antisera against two American strains of PYDV (ATCC PVAS 233 and ATCC PVAS 234) proved negative.

IDENTIFICATION AND CHARACTERIZATION OF A POTEXVIRUS ISOLATED FROM NIGHT-BLOOMING CACTUS. A. E.-S. A. Fudl-Allah, L. G. Weathers, and F. C. Greer Jr. Department of Plant Pathology, University of California, Riverside, CA 92521.

Plants of night-blooming cactus, *Hylocereus undatus*, as well as scions of several species of cactus grafted to it, were stunted, malformed, and systemically mottled. A virus was sap-transmitted from affected plants to *Chenopodium quinoa*, *Gomphrena globosa*, *Amaranthus caudatus*, and *Saponaria vaccaria*. Symptoms in *C. quinoa* consisted of chlorotic primary lesions and systemic mottle and necrosis. *Gomphrena globosa* and *A. caudatus* showed primary local lesions. Plants of *S. vaccaria* showed only systemic mottling. Crystal and spindle-like inclusions were found by light microscopy in epidermal cells of infected *H. undatus*. The virus has a T₁₀ above 92°C and DEP of 10⁻⁷ in sap from infected *C. quinoa*. Electron microscopy of preparations from infected plants revealed flexuous rod-shaped particles with a normal length of about 515 nm. Partially purified preparations of the virus from *C. quinoa* reacted with antisera prepared against California barrel cactus and Chessin's isolate of cactus virus X.

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Graft and Seed Transmission of the Causal Agent of Top Spotting in *Kalanchoe blossfeldiana*. S. S. Hearon and J. C. Locke. F&NCL, USDA, Beltsville, MD 20705

A ubiquitous disease in *Kalanchoes*, characterized by the transient appearance of coalescing chlorotic rings and spots on new leaves emerging in the spring and fall, is herein called top spotting (TS). Seeds were collected from a self-pollinated plant of 3 *Kalanchoe* varieties with TS. Fifty and 100 seedlings of each variety were selected randomly from seeds germinated after 6 and 15 months storage, respectively, at 4°C and maintained in a greenhouse. Approximately 65% of the Cactus Candy, 50% of the Osage Orange and 30% of the Louise seedlings showed TS. Shoots (c. 5 cm) from 1 of 4 Cactus Candy seedlings with TS were top grafted onto 3 rooted cuttings from each of 11 symptomless Cactus Candy seedlings. The 33 tops removed from the healthy stocks were rooted as controls. Disease symptoms developed within 9 months in >90% of the 29 successfully grafted plants, but in no controls. Seedlings were negative when indexed on *Chenopodium quinoa* for two known flexuous rod viruses of *Kalanchoe*.

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VARIATIONS IN MORPHOLOGY AND MOSAIC VIRUS RESISTANCE IN PLANTLETS OF TANIERS (*XANTHOSOMAS* SP.) VIA TISSUE CULTURE. Lii-Jang Liu, Margarita Licha, Delia Baella and Evelyn Rosa, Department of Crop Protection, College of Agricultural Sciences, University of Puerto Rico, Mayaguez, Puerto Rico 00708

More than 3000 plantlets of taniers were obtained via meristem tip and callus cultures. They were inoculated with mosaic virus to determine their degree of resistance. Variations in symptom expressions and in size and shape of leaves of plantlets of taniers were observed. The percent of infection by virus varied from 2-21. According to their size and vigor, plantlets were classified into the following 3 groups: (1) large and vigorous plantlets without virus symptoms, (2) medium size plantlets with virus symptoms, and (3) small and dwarf plantlets with severe virus symptoms. This apparently indicated that the progeny of taniers via tissue culture was heterogeneous thus raising the possibility that virus resistant and vigorous plantlets may be obtained via callus cultures.

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A NEW BACTERIAL DISEASE OF SOYBEAN IN FLORIDA. R. E. Stall, and T.A. Kucharek, Dept. of Plant Pathology, University of Florida, Gainesville, FL 32611.

A bacterial leaf spot disease was prevalent on 'Centennial' cultivar of soybean during 1980 and 1981 in Madison County, Florida. 'Centennial' and 'Lee' were the only ones among 13 cultivars of maturity groups 5-9 that were susceptible in pathogenicity tests. The causal bacterium was identified as *Pseudomonas andropogonis*. Key characteristics of the bacterium were non-fluorescence on KMB medium, accumulation of poly-β-hydroxybutyrate, oxidase negative, arginine dihydrolase negative, non-utilization of arginine, and no growth at 41°C. The bacterium was also pathogenic on 'Bountiful' common bean, but not on corn, white clover, or velvet bean which were hosts of *P. andropogonis* isolated from corn and sorghum in Florida. The corn and sorghum isolates were not pathogenic to soybean or 'Bountiful' bean. Since the soybean pathogen is physiologically indistinguishable from *P. andropogonis*, but pathologically distinct, it is designated as *P. andropogonis* pv. *sojae*.

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ERWINIA CHRYSANTHEMI AS A PATHOGEN OF GRAIN SORGHUM. S. G. Jensen, W. R. Mayberry and J. A. Orligawitch, Dept. of Plant Pathology, Univ. of Nebraska, Lincoln, NE 68583-0722.

A bacterial pathogen of grain sorghum caused a soft rot of the stalk of plants in the preboot stage. The organism is an effective pathogen only at high temperatures (>30°C) but it can attack several commercial genotypes. Three methods were used to compare this bacteria with other authentic cultures: 1) standard morphologic-metabolic criteria, 2) analysis of soluble proteins by polyacrylamide gel electrophoresis, and 3) gas chromatography of cellular fatty acids. The three methods gave good agreement and identified the pathogen as being similar to a strain of *Erwinia chrysanthemi*. A discussion of the diagnostic methods and the significance of the pathogen will be given.

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PSEUDOMONAS SYRINGAE pv. *TAGETIS*: A NEW PATHOGEN ON JERUSALEM ARTICHOKE. W.W. Shane and J.S. Baumer, Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108.

More than 15% of the Jerusalem artichoke (JA), *Helianthus tuberosus* L., plants in several Minnesota fields had apical chlorosis, large chlorotic leaf spots, and small necrotic leaf spots with small chlorotic halos. Isolations consistently yielded an oxidase negative, gram negative, fluorescent bacterium that was identified as *Pseudomonas syringae* pv. *tagetis*. Isolates from JA (15) were compared with known isolates from marigold (3), ragweed (1), and sunflower (3). Tests for carbon source utilization agreed with published accounts for marigold isolates (Trimboli, D., et al., 1978. Aust. J. Agric. Res. 29:831-839) except none grew on DL-β-hydroxybutyrate, and all used L-erythritol. Isolates from JA induced either necrotic spots (N) or necrotic spots and chlorosis (C) upon spray-inoculation or infiltration of JA with bacterial suspensions. Chlorosis was also induced by C isolates infiltrated into marigold, zinnia and sunflower, while N types only induced a slight necrosis in sunflower.

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IDENTIFICATION OF A UNIQUE BACTERIUM ISOLATED FROM DATE PALM TISSUE CULTURE. Joe W. Willis, Elizabeth A. Allingham, and J. V. Leary. Department of Plant Pathology, University of California, Riverside, CA 92521.

A previously undescribed bacterium has been isolated from tissue culture of palm embryos and callus. Repeated attempts to free the tissue from contamination by surface sterilizing with 0.2% to 2.6% NaOCl indicate that the bacterium may be found within the palm tissue or may survive as spore-like structures. The bacterium is a gram-negative, facultatively anaerobic, oxidase- and catalase-variable rod with lophotrichous flagella. The organism is slow-growing, forming small, white, flat, round colonies on complete agar, and requiring added glucose or sucrose for growth in broth. The bacterium has natural resistance to nalidixic acid (30 µg/ml), carbenicillin (50 µg/ml) and penicillin (10 µg/ml) but not tetracycline (30 µg/ml) or streptomycin (10 µg/ml). The bacterium grows in nitrogen-free medium, and, considering the presence of spore-like, electron-dense structures, may be a unique organism.

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EFFECT OF TREATMENT WITH SYMBIOTIC BACTERIA FROM MAR COTTONS ON RESISTANCE TO *XANTHOMONAS MALVACEARUM*. K. M. El-Zik and L. S. Bird, Department of Plant Sciences, Texas A&M University, College Station, Texas 77843

Symbiotic bacteria, *Bacillus* spp., isolated from the cotton cultivar Tamcot CAMD-E were used in greenhouse and field experiments to evaluate their effect on resistance to the bacterial blight pathogen and agronomic characteristics of cotton cultivars. In the greenhouse, the symbiotic bacteria caused the susceptible cultivars Acala SJ-5 and Stoneville 213 to become less susceptible and partially resistant, respectively. The partially resistant Blightmaster became more resistant. In the field, disease grades were significantly reduced 31.7% for Acala SJ-5 and 37.5% for Stoneville 825. Symbionts had no effect on the immune MAR cottons Tamcot SP21S and CAMD-E. Treatments with symbiotic bacteria did not influence lint yield, earliness, gin turnout, lint %, fiber length, uniformity, or strength. A significant increase in micronaire value was obtained with the symbiont treatment. The symbiotic bacteria can cause host resistance to *X. malvacearum*.

EVALUATION OF CLONES OF *Solanum tuberosum* SUBSP. *ANDIGENA* FOR RESISTANCE TO TUBER AND STEM ROT CAUSED BY *ERWINIA CHRYSANTHEMI*. Oscar A. Hidalgo and Eddie Echandi. Dept. of Plant Pathology, N. C. State University, Raleigh 27650.

Tubers of 149 clones of *Solanum tuberosum* subsp. *andigena* from self-pollinated families (Jak 072, 702440, 700718 and Och 5331) were evaluated for resistance to *Erwinia chrysanthemi* and classified into 4 categories: resistant (r), intermediate (i), susceptible (s), and very susceptible (vs). Family Jak 072 had 23% of the clones r and 61% i. Family Och 5331 had 22% of the clones s and 67% vs; and families 702440 and 700718 had 38% and 29% of the clones i and 45% and 40% s, respectively. Clones (11), representing the 4 categories were inoculated with *E. carotovora* subsp. *carotovora* (Ecc), *E. carotovora* subsp. *atroseptica* (Eca) and *E. chrysanthemi* (Ech). Clones responded similarly to Ech and Eca; however, significantly more rot was caused by Ecc. Clones Jak 072-17 and Jak 072-18 were resistant to Ech, Ecc and Eca. When above ground stems of the 11 clones were inoculated with Ech, no correlation was found between the response of tubers and above ground stems to tuber and stem rot.

CALCIUM PROPIONATE AND TARTARIC ACID AT 50C FOR THE CONTROL OF *PSEUDOMONAS SYRINGAE* PV. *LACHRYMAN* IN CUCUMBER SEED. Curt Leben, Ohio Agricultural Research and Development Center and The Ohio State University, Wooster, OH 44691.

Cells of the pathogen in water were vacuum-inoculated into lots of 500 seeds, which were dried at room temperature. Seeds were planted in moist vermiculite in closed, lighted (16 h/day) containers. After 6-8 days at 24C, numbers of plants with lesioned cotyledons were recorded. Seeds, vessels, and chemical preparations were preheated to 50C before mixing preparations and seeds for the 20 minute treating period, which was followed by rapid drying. In three tests with calcium propionate (33 g/liter of water), tartaric acid (33 g/liter), or acidic cupric acetate (Appl. Environ. Microbiol. 39:803-807, 1980) there were less than 1% diseased plants, as compared with more than 60% disease in inoculated control plants. In a field test with two cucumber cultivars, the three treatments also gave good seedling disease control. Germination was not reduced by treatments.

EPIPHYTIC BEHAVIOR OF *PSEUDOMONAS SYRINGAE* PV. *TABACI* ON ROOTS OF CABBAGE SEEDLINGS. Stephen Diachun and G. C. Bergstrom. Departments of Plant Pathology, University of Kentucky, Lexington, KY 40546 and Cornell University, Ithaca, NY 14853, respectively.

When roots of cabbage (cv. 278 Market Topper) seedlings were dipped in suspensions of *Pseudomonas syringae* pv. *tabaci* (PST), the tobacco wildfire bacterium, microscopically visible colonies formed on the root surfaces within 48 hours but no symptoms were observed in the seedlings. When cotyledons and young leaves of cabbage were infiltrated with PST suspensions in excess of 10^8 cells/ml, symptoms also failed to develop. This contrasts with previous work on cucumber seedlings in which infiltrated PST induced local and systemic wildfire symptoms. These results provide further evidence that PST has the general capacity for epiphytic growth on plant roots, as has been previously reported on certain species in the Cucurbitaceae, Gramineae, and Solanaceae families. However, it appears that the epiphytic survival of PST on plant roots is unrelated to its ability to induce symptoms in infiltrated tissues of the same plants.

SYNERGISTIC RELATIONSHIPS OF *VERTICILLIUM DAHLIAE* AND *ERWINIA CAROTOVORA* PV. *CAROTOVORA* IN EARLY-DYING DISEASE OF POTATO PLANTS. M. K. Rahimian and J. E. Mitchell, Dept. of Plant Pathology, Univ. of Wisconsin-Madison, Madison, WI 53706

The effect of simultaneous infections of potato plants with *Verticillium dahliae* (Vd) and *Erwinia carotovora* pv. *carotovora* (Ecc) was studied under growth room conditions. Vd was inoculated by root dip and Ecc was inoculated in a manner that simulated natural foliar infection and natural basal stem infection that occurs through seed pieces. When plants were inoculated with Ecc via the cut stem base severe symptoms of early-dying disease appeared in Russet Burbank and Norgold Russet plants. Only Norgold Russet plants were affected adversely following the additional effect of Ecc inoculations with a micropipette at a leaf axil of the stem. Disease symptoms developed faster in plants inoculated with both pathogens than in plants inoculated with either pathogen alone. The combination of Vd and Ecc was synergistic; plant growth was reduced, severity of chlorosis and wilting was increased, and development of stem soft rot was enhanced.

AN OUTBREAK OF A STEM NECROSIS OF CHRYSANTHEMUM INCITED BY *PSEUDOMONAS CICHORII*. J. B. Jones, A. W. Engelhard, and B. C. Raju. The Agricultural Research and Education Center, 5007-60th Street East, Bradenton, FL 33508-9324 and Yoder Brothers, Ft. Myers, FL 33902.

An unusual stem necrosis of chrysanthemum occurred in Florida on two chrysanthemum farms in January and February, 1982. The symptoms occurred close to harvest and were typified by long dark-colored streaks on the stems. Isolations yielded bacteria that were fluorescent on medium B of King et al., were oxidase positive, arginine dihydrolase negative and induced leaf spots and dark stem streaks on chrysanthemum plants and was identified as *Pseudomonas cichorii*. *P. cichorii* typically causes a leaf spot and bud blight of chrysanthemum plants but not stem necrosis. In this outbreak symptoms did not occur on the leaves or buds. No explanation is available for this unusual expression of symptoms.

THE EFFECT OF MAGNESIUM AND POTASSIUM NUTRITION OF PEPPER ON BACTERIAL SPOT SUSCEPTIBILITY. J. B. Jones, S. S. Woltz, and J. P. Jones. University of Florida, AREC-Bradenton, Bradenton Florida 33508-9324.

Peppers, cv. 'Early Cal Wonder,' were grown at five rates of K (30, 60, 120, 240 and 480 mg/ml) applied as KCl and K_2SO_4 , and at three rates of Mg (0, 40, and 200 μ g/ml) applied as $MgCl_2$ in a factorial arrangement to determine susceptibility to *Xanthomonas campestris* pv. *vesicatoria*. The nutrient solutions were applied to plants grown in Myakka fine sand amended with $CaCO_3$ and superphosphate. Plants grown at the high potassium rates had lowest disease ratings. K and chloride (Cl) levels in tissues were highest at the highest potassium rate. Tissue Mg decreased with increasing K. Plants grown at the two highest rates had lower disease ratings than plants grown without Mg. Tissue Cl was highest in plants grown at the two higher Mg rates. Cl may be a factor in disease severity.

OVERWINTERING OF *PSEUDOMONAS SYRINGAE* PV. *SYRINGAE* IN PEACH TWIGGS. Elke Endert and D.F. Ritchie, Dept. of Plant Pathology, North Carolina State University, Raleigh, NC 27650.

Peach trees (*Prunus persica* cv. 'Redhaven') were inoculated in Oct. during early leaf abscission and in Feb. by either brushing a bacterial suspension onto the buds and leaf scar (BI) or injecting bacteria into internodal punctures (PI). Internal populations of *P. s.* pv. *syringae* were monitored biweekly until early bloom. Oct. BI resulted in initial populations of ca. 10^2 cfu/BI which rapidly declined below detection, whereas Feb. BI resulted in no detectable internal colonization. Populations from Oct. PI remained constant. Feb. PI produced not only higher populations ($>10^3$ cfu/PI) but also the formation of sunken cankers (99%) and necrosis or inhibition of growth initiation of fruit buds (39%) and shoot buds (20%). *P. syringae* was reisolated from 68 and 75% of these buds, respectively, as well as from apparently healthy buds. These results suggest wounds may be more suitable as infection courts than are natural openings, and that movement within peach tissues is associated with rapid initial colonization of the inoculation site.

FREQUENCY AND PATHOGENICITY OF AFLUIDAL VARIANTS OF *PSEUDOMONAS SOLANACEARUM* FROM MOKO-DISEASED BANANAS. A. C. Woods. Division of Tropical Research, United Fruit Co., La Lima, Honduras.

Afluidal variants of *Pseudomonas solanacearum* race 2 are routinely isolated from approximately 30% of Moko-diseased banana mats in Honduras. Of all variant isolates, 75% are mixed with the normal fluidal 'F' colony type, and 25% are pure. When inoculated into young, potted banana plants at 10^5 CFU per plant, the variant type is always less aggressive than the 'F' type. Symptoms of infection with the afluidal variant in mature field plants are usually restricted to vascular browning, especially in the rhizome. External symptoms are slow to develop and are mild when compared to infections with the 'F' type. Afluidal variants are often isolated from groups of neighboring plants indicating mechanical transmission. This is the first report of an afluidal form of *P. solanacearum* as an important field pathogen in bananas.

ERWINIA CHRYSANTHEMI SOFT-ROT OF ALOE VERA. E. N. MULREAN, COTTON RESEARCH CENTER, PHOENIX, AZ., AND T. V. SUSLOW, ADVANCED GENETIC SCIENCES, BERKELEY, CA.

An undescribed bacterial soft-rot of hydroponically grown Aloe vera, (*Aloe barbadense* var. *vera*) was observed in the Phoenix area in 1981. Symptoms first appeared on the older outer leaves with the rot moving basipetally into the crown and adjacent leaves. The causal bacterium was phosphatase (+), facultatively anaerobic, liquified gelatin, and grew at 39°C. These and other characteristics places the Aloe bacterium in the *Erwinia chrysanthemi* group. In greenhouse inoculations these isolates were also pathogenic on *A. jucunda*, *A. dectoidonta* and *A. chaboudii* but not *A. variegata*. Three cactus species, *Gymnocalycium optima rubra*, *Hylocereus undatus* and *H. triangularis*, were also soft rotted by the Aloe strains.

REDUCED SEVERITY OF ERWINIA SOFT ROT IN POTATO TUBERS WITH HIGH CALCIUM CONTENT. R. G. McGuire and A. Kelman, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Application of increasing amounts of CaSO_4 to Superior potato plants grown in sand culture produced tubers with calcium levels ranging from 0.03 to 0.30% DW in the periderm and from 0.02 to 0.07% in the cortical tissues. Although 90% of the maximum tuber yield was attained with a calcium concentration of approximately 40 $\mu\text{g/ml}$ in the nutrient solution, an increase to 500 $\mu\text{g/ml}$ was required to raise the tuber content above deficiency levels. In an infectivity titration when *Erwinia carotovora* pv. *atroseptica* was injected into whole tubers following harvest, a smaller population of cells was required to initiate decay in tubers with low calcium than in high calcium tubers. The middle lamellae of low calcium tubers showed a higher level of KOH-soluble pectic materials while high calcium tubers had more KOH-insoluble pectates. Treatment with bacterial culture filtrates containing pectic lyase caused an accelerated loss of electrolytes and protoplast viability in slices from low vs. high calcium tubers.

AGGLUTINATION OF SAPROPHYTIC PSEUDOMONADS BY WATER WASHES OF BEAN ROOTS AND SHOOTS. Anne J. Anderson, Department of Biology, Utah State University, Logan, Utah 84322.

Water washes of roots and shoots of bean (*Phaseolus vulgaris*) caused agglutination of cells of saprophytic pseudomonads, *P. putida* and *P. fluorescens*. Cells of plant pathogenic pseudomonads were not agglutinated. Agglutinin activity passed without absorption through DEAE and CM-Sephadex and was precipitated with ethanol. Chromatography on Sepharose Cl-6B-200 resulted in activity in the void and included fractions. The higher molecular weight agglutinin possessed neutral sugar, acidic sugar and protein in the ratios (5.6:1:0.5) for shoot and (2.8:1:0.7) for root. Arabinose and galactose were the major neutral sugars. These preparations can be compared to an agglutinin purified from bean leaf homogenates with ratio (4.5:1:0.8). *Pseudomonas putida* cells although agglutinated can multiply in the sterilized root wash. The ready extraction of the agglutinin from the undamaged plant suggests the agglutinins could be involved in regulating the bacteria that flourish in the rhizosphere or phylloplane.

DIFFERENTIAL BINDING ABILITY OF ERWINIA CAROTOVORA VAR. CAROTOVORA AND ATROSEPTICA TO POTATO. R. D. Davidson, R. W. White and D. C. Sands, Dept. of Plant Pathology, Montana State Univ., Bozeman, MT 59717.

Erwinia varieties (*Erwinia carotovora* var. *carotovora* (Ecc) and *atroseptica* (Eca) were tested for differential binding ability to potato tissue. A potato slice was placed in bacterial suspensions of differing amounts (0.2×10^8 cfu/ml. to 4×10^8 cfu/ml.) for each bacterial variety tested. Binding was allowed to occur and then the slice was suspended in water for 1 hour to allow for equilibrium. Bacterial plate counts were taken immediately on the rinse water and on the potato piece after grinding in a blender. Ecc showed at least a ten-fold higher binding rate than Eca in all replications. In mixed cultures (Ecc and Eca), Ecc showed an even higher preferential binding to the tuber, nearly eliminating the Eca binding. This variation in binding ability to the potato may partially account for the differences in disease symptoms between Ecc and Eca observed under field conditions.

RECOVERY OF CORYNEBACTERIUM AGROPYRI FROM OOZE ON NATURALLY INFECTED AGROPYRON SMITHII AFTER 37 YEARS. T. D. Murray, Dept. of Plant Pathology, Washington State Univ., Pullman, WA 99164-6430.

In 1945, heads of *Agropyron smithii* exhibiting symptoms of bacterial head blight, caused by *C. agropyri* (O'Gara) Burk., were collected by R. Sprague, G.W. Fischer, and J.P. Meiners. Diagnosis was based on symptoms and the collections were stored in a classroom at room temperature and ambient relative humidity. In 1982, a yellow, gram positive coryneform bacterium resembling the description given in Bergey's Manual (6th ed., p.395) was isolated from the dried yellow ooze present on the heads. Viability of the bacterium in the ooze was estimated to be 1.3-1.8%. Recovery of this bacterium is significant in two aspects. First, to my knowledge, this is the longest reported survival of a bacterial plant pathogen in a naturally infected host. Second, *C. agropyri* has been deemed *nomina dubia* due to the loss of known cultures. Although it is unlikely that survival for such an extended time period under these conditions is relevant to epidemiology of the disease, recovery of this bacterium from 37-year-old ooze emphasizes the importance of ooze in the survival of bacterial plant pathogens.

PATHOGENICITY, BACTERIOCINS AND COLONY MORPHOLOGY OF CORYNEBACTERIUM MICHIGANENSE SSP. TESSELLARIUS ISOLATED FROM WHEAT SEED. R.R. Carlson, A.K. Vidaver and J.E. Siebler. Dept. of Plant Pathology, University of Nebraska, Lincoln, NE 68583-0722.

Seed from 10 hard red winter wheat cultivars collected at two sites was screened for *Corynebacterium michiganense* ssp. *tessellarius*, the causal agent of bacterial mosaic of wheat. Contamination levels ranged from <100 to 800 CFU/gram of seed at one site and from 1400 to 59,000 CFU/gram at the second. Sodium hypochlorite treatment (0.5% for 3 min) of seed from the second site reduced contamination (range, <100 to 4500 CFU/gram). Colony morphology, bacteriocin production, and pathogenicity were determined for 68 isolates. The pathogenic isolates of some seed samples had multiple bacteriocin types. Sixty isolates were orange, entire and domed; 51 of these were pathogenic. Forty-seven isolates produced *C. michiganense* ssp. *tessellarius* specific bacteriocins; 41 of these were pathogenic. Thus, colony morphology was at least as good as bacteriocin production as a predictor of pathogenicity.

ULTRASTRUCTURAL DEFINITION OF WILT IN JONATHAN APPLE SHOOTS INDUCED BY AMYLOVORIN, AN AMYLOVORIN FRAGMENT AND AN ARRAY OF DEXTRAN POLYMERS. R. Goodman, E. King and K. Sijam. Department of Plant Pathology, University of Missouri, Columbia, MO 65211.

Amylovorin, an extracellular polysaccharide (EPS) of *Erwinia amylovora* that has a MW of $\approx 10^8$ Daltons has been reduced to $\approx 2-4 \times 10^4$ Daltons by an *E. amylovora* bacteriophage polymerase. We have compared the rate, intensity and ultrastructural quality of wilt in Jonathan shoots by amylovorin and its much smaller fragment with an array of dextrans with molecular weights of $5-40 \times 10^6$, 2×10^6 , 2×10^5 and 4×10^4 Daltons. Wilt was induced by amylovorin, its fragment and by the two larger dextrans. A comparison of the wilt induced by the amylovorins and the dextrans at the ultrastructural level revealed differences in the vessel occluding menstrea. Specifically, amylovorin caused the formation of a fibrillar mesh interspersed with membrane-like globular bodies. The higher molecular weight dextrans formed occluding matrices that were devoid membranous globular bodies. The nature of the two types of occluding matrices and the rationale for their formation and their effect is to be discussed.

SELECTIVE DIFFERENTIATION OF GAEUOMANNOMYCES GRAMINIS VAR. TRITICI ON A MEDIUM UTILIZING L-DOPA. M. Elliott-Juhnke, Dept. of Plant Pathology, Montana State Univ., Bozeman, MT 59717.

A medium, designated SM-GGT, was developed which allows ready differentiation of *Gaeumannomyces graminis* var. *tritici* (Ggt) from other soil fungi based on melanin-like pigment production in the presence of L-8-3,4-dihydroxyphenylalanine (L-DOPA). SM-GGT contains 1 mg dichloran, 10 mg quintozene, 30 mg fenaminosulf, 100 mg streptomycin sulfate and 500 mg L-DOPA added to 1 liter potato dextrose agar after autoclaving and cooling to 50°C. In 1981, 16 samples from 13 irrigated wheat fields, visually diagnosed as infected with Ggt, were tested on SM-GGT after surface sterilizing with a 1% silver nitrate solution. Seventy-two percent of the samples yielded Ggt of which 96% produced a melanin-like pigment. This is a diagnostic medium for the isolation and identification of Ggt rather than being a completely selective medium since it does not selectively inhibit *Fusarium* species. However, Ggt is easily distinguishable from *F. culmorum* on SM-GGT based on gross morphology and pigment production.

A SEMISELECTIVE MEDIUM FOR ISOLATING APHANOMYCES FROM PLANT TISSUE. W. Pfender, P. Delwiche, C. Grau, and D. Hagedorn. Dept. of Plant Pathology, Univ. of Wisconsin, Madison, WI 53706.

Pythium, *Rhizoctonia* and *Fusarium* spp. commonly interfere with successful isolation of *Aphanomyces* from infected plant tissue. An effective medium for isolating *Aphanomyces* from roots of pea, bean and alfalfa contained the following (per liter): 10g Bacto water agar, 10g Difco cornmeal agar, 5mg benomyl, 30mg metalaxyl and 200mg vancomycin. The medium prevents growth of *Fusarium solani*, *F. oxysporum*, *Phytophthora megasperma*, *Thielaviopsis* sp., *Verticillium* sp. and several *Pythium* spp. Growth of other *Pythium* spp. and *Rhizoctonia* is not prevented but is inhibited by at least 75%. Growth of *Aphanomyces* isolates from pea, bean and alfalfa is inhibited less than 25%. If *Alternaria* interferes with isolation, 0.5ppm amphotericin B can be used in the medium, though this inhibits *Aphanomyces* growth slightly more. Roots of pea and bean grown in naturally infested soil were incubated on this medium at ca 22C, and 56% of the fungi recovered were *Aphanomyces*; on nonselective medium the proportion was 7%. With alfalfa the respective proportions were 26% and 3%.

A TECHNIQUE TO INCREASE SPORE PRODUCTION OF FUNGAL PATHOGENS ON AGAR MEDIA. H.L. Warren and S.K. Onken. USDA, SEA, Purdue University, West Lafayette, Indiana 47907.

Spore production of *Exserohilum turcicum*, *Bipolaris maydis*, *B. carbonum*, *Diplodia maydis*, and *Colletotrichum graminicola* increased following a multi-point inoculation technique as compared to normal inoculation techniques. One ml of a 10% skim milk (Difco) spore and/or mycelial suspension was flooded on the surface of an agar plate. There was a significant increase in sporulation of *E. turcicum*, *B. maydis* and *B. carbonum* on lactose casein hydrolysate agar and *D. maydis* on potato dextrose agar (PDA) and basal mineral salts medium with 3% sucrose and 50 ppm biotin using the skim milk suspension instead of single point inoculation. Spore production of *C. graminicola* on PDA using the skim milk suspension was equal to single point inoculation on oatmeal agar. Comparisons were also made between skim milk and distilled water suspensions. Spore production increased 3-to 10-fold and time spent inoculating agar plates decreased using the skim milk suspension, multi-point inoculation technique.

SEPARATION OF TILLETIA CRIES AND T. CONTROVERSA TELIOSPORES BASED UPON SPORE WALL CHARACTERISTICS. W. M. Hess, Department of Botany and Range Science, Brigham Young University, Provo, Utah 84602.

Although they have different germination patterns, the teliospores of *T. caries* and *T. controversa* are impossible to distinguish with standard EM procedures with SEM and TEM, including freeze-fracture. If spores are completely hydrated so sheaths are fully expanded, followed by critical point drying, osmication, critical point drying again, and embedment in resin, it is possible to emphasize differences in the outer two wall layers of various races or collections of both species with thin sections. Samples critical point dried only once are not as well stained. It is important to be able to distinguish between these two species as chemical treatment is easily used for *T. caries*, but not for *T. controversa*.

LIPID VACUOLES IN TILLETIA SPP. TELIOSPORES. Ja Kyoung Oh, and W. M. Hess. Dept. of Botany and Range Science, Brigham Young University, Provo, Utah 84602.

Tilletia spp. teliospores from infected *Glyceria striata* were soaked in dilute detergent solution for 13 min, 30 min, and 16h before freeze-fracturing. Spores contained vacuoles, lipid vacuoles, and unidentified organelles. Vacuoles and lipid vacuoles enlarged and became less regular in shape in spores soaked for 16h, and lipid bodies and lipid vacuoles were more variable in size. Lipid bodies were commonly associated with lipid vacuoles, particularly in spores soaked for longer periods of time. Membranes of vacuoles and lipid vacuoles contained relatively few particles compared to membranes of unidentified organelles, which indicates that lipid vacuoles and unidentified organelles have different functions.

RIPE ROT OF MUSCADINE GRAPE AND ANTHRACNOSE FRUIT ROT OF HIGH-BUSH BLUEBERRY CAUSED BY COLLETOTRICHUM GLOEOSPORIOIDES. M. E. Daykin and R. D. Milholland, Department of Plant Pathology, North Carolina State University, Raleigh 27650.

Colletotrichum gloeosporioides Penz. causes destructive fruit rots of muscadine grape (*Vitis rotundifolia* Michx.) and highbush blueberry (*Vaccinium corymbosum* L.) at ripening. Isolations made from grape mummies, pedicels, and fruit spurs and from blueberry twigs during the months of January and February revealed that these are primary sites of overwintering. Abundant conidia of *C. gloeosporioides* were trapped in rainwater run-off from detached plant material during the early spring, but levels of spore production declined through the remainder of the growing season. However, in vineyards and blueberry fields, conidia were trapped from March through October during rainy periods with no decline in spore production. Field inoculations of developing fruits as well as isolations from naturally infected green fruits indicated that infection occurs at anytime after flowering but remains latent until ripening.

BUD INFECTION AND PEACH TREE GUMMOSIS. Kerry O. Britton and Floyd F. Hendrix, Dept. of Plant Pathology, Univ. of GA, Athens, GA 30602

Botryosphaeria spp. characteristically invade wounds and cause cankers on many deciduous hosts. At least three species, *B. obtusa*, *B. dothidea*, and *B. rhodina*, can cause gummosis cankers on peach trees, invading through wounds or occasionally lenticels. The most prevalent of these, *B. obtusa*, is also present in dormant buds, the frequency of isolation increasing steadily until bud swell. Healthy dormant buds on container-grown trees were inoculated with conidial suspensions of each of the three species. Inoculated buds died as they began to swell, whereas uninoculated control trees had no dead buds. In nature the loss of infected buds is probably insignificant in terms of peach production. However, histological examination indicates that these fungi move systemically in the xylem vessel elements. The gum produced by the host in response disrupts translocation more than does pathogen mycelium. Unless these obstacles are overcome by the vigorous addition of new xylem vessels, dieback of the fruiting branches and further systemic movement of the fungus ensue.

COLONIZATION OF 'DELICIOUS' APPLE FRUITS BY ALTERNARIA SPP. M. A. Ellis and J. G. Barrat, Department of Plant Pathology, The Ohio Agricultural Res. & Devel. Center, Wooster, and The Ohio State University, Columbus 43210 and Div. of Plant and Soil Sciences, West Virginia Univ., Kearneysville, WV 25430, respectively.

Fourteen genera of fungi were isolated from the core region of 'Delicious' apple fruits. *Alternaria* spp. were the most commonly isolated fungi. Moldy-core (visible mycelia in the core region) was caused by *Alternaria* spp. *Alternaria* colonized flower parts during and shortly after bloom and later moved (presumably through the open calyx tube) into the receptacle or core region of the fruit. In 2 years of testing, in Ohio, *Alternaria* was recovered from the core region of almost 100% of all Delicious fruits tested at harvest, and resulted in moldy-core of 38 and 65% of the fruits during 1980 and 1981, respectively. A variety of fungicide spray programs (including bloom sprays) had no effect on the rate of flower and fruit colonization by *Alternaria* spp. or on the incidence of moldy-core at harvest.

SOIL POPULATIONS OF MACROPHOMINA PHASEOLINA IN ARIZONA Deborah J. Young and S.M. Alcorn, Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

Macrophomina phaseolina (Tassi) Goid. was the causal agent of charcoal rot of *Euphorbia lathyris* L. plants, growing in a plot newly cleared of Sonoran desert vegetation (Plant Disease 66: in press, 1982). Native populations of this fungus were isolated from soils of three vegetative communities in southern Arizona. Samples from two subdivisions of the Sonoran Desert (Arizona Upland and Lower Colorado) had populations ranging from 1 to 8 sclerotia per gram soil. Plains and Desert Grasslands had 1 to 52 sclerotia per gram soil. Pathogenicity tests of these isolates are in progress. As few as five sclerotia of *M. phaseolina* per gram soil can cause charcoal rot of *E. lathyris*. Sclerotial populations were also determined in agricultural soils near Tucson; these varied from 1 to 250 sclerotia per gram soil. Populations were influenced by prior cropping history, cultural practices, and season. Sclerotia were unevenly distributed throughout the field.

TEMPERATURE EFFECTS ON SIZE OF *MONILINIA FRUCTICOLA* CONIDIA PRODUCED ON FRESH STONE FRUITS. D. J. Phillips, USDA-ARS, P. O. Box 8143, Fresno, CA 93747.

Conidia from 3 isolates of *Monilinia fructicola* (Wint.) Honey were produced in the laboratory on fruit from 1 cultivar each of cherry, peach, and nectarine. Cherries and peaches, but not nectarines, yielded significantly larger conidia at 15 than at 25 C. In 3 tests in which conidia from peaches and nectarines were compared, conidia from peaches had a volume of 1100 μm^3 when grown at 15 C and 630 μm^3 when grown at 25 C; for nectarines the respective values were 1130 μm^3 and 1090 μm^3 . The mean volume of the spores produced in the orchard on peaches, unlike most nectarines tested, was inversely correlated with the mean minimum temperature for the 3 days prior to sampling. Earlier, we found large conidia to be more aggressive than small conidia when inoculated on peaches. Our results suggest that, during warm weather, some cultivars of nectarines with brown rot may yield larger, more aggressive conidia than peaches; consequently, the rate of secondary infection would be higher for these nectarines than for peaches.

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BLACK LEAF SPOT PHASE OF STRAWBERRY ANTHRACNOSE CAUSED BY *COLLETOTRICHUM FRAGARIAE*. C. M. Howard and E. E. Albregts, University of Florida, Agr. Research Center, Rt. 2, Box 157, Dover, FL 33527.

Anthrachnose and crown rot caused by *Colletotrichum fragariae* Brooks (= *C. gloeosporioides* Penz. according to Arx) have been serious problems of strawberries in Florida for many years. A heretofore unidentified leaf spot often has occurred in Florida, sometimes before anthrachnose symptoms were found on stolons or petioles. The spots are round, black, 0.5-2 mm in diameter and often become numerous on leaflets without killing them. Isolations from the lesions consistently yielded *C. fragariae*. Potted strawberry plants in the stolon producing stage were inoculated with spore suspensions from pure cultures of *C. fragariae* isolated from a leaf spot lesion and from the crown of a wilted plant. The plants were then enclosed in plastic bags for 3 days. Both isolates caused typical anthrachnose lesions on stolons and petioles, crown rot, and leaf spots identical to those which occur in the field. The anthrachnose syndrome should be expanded to include black leaf spot.

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CROWN CANCKER DISEASE OF PIGEON PEA CAUSED BY A STERILE BASIDIOMYCETE IN PUERTO RICO. W. J. Kaiser*, P. L. Meléndez**, and M. Zapata**. *USDA, ARS, Regional Plant Introduction Station, Washington State University, Pullman, WA 99164 and **Department of Crop Protection, University of Puerto Rico, Mayagüez, PR 00708.

A sterile, white basidiomycete with hyphal clamp connections was isolated from crown cankers on naturally infected pigeon peas (*Cajanus cajan*) at Isabela, PR. The sterile fungus was similar morphologically to *Sclerotium rolfsii*, but failed to form sclerotia in culture or on infected plant tissues. In inoculation tests, the pathogen caused pre- and post-emergence damping-off of pigeon peas. Sunken, reddish-brown lesions often developed on the hypocotyls of older plants. The fungus also was pathogenic to bean, cowpea, mungbean, soybean, cucumber, squash, and watermelon. Occasionally, sterile fleshy structures reminiscent of fruiting bodies formed beneath the soil surface on hypocotyls of inoculated plants. The sterile pigeon pea fungus is very similar, if not identical, to a sterile soilborne pathogen of bean in Florida (Phytopathology 67:430-433, 1977).

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A DORMANT RHIZOME ROT OF SCOTCH SPEARMINT, *MENTHA CARDICA*, CAUSED BY *RHIZOCTONIA SOLANI*. C.B. Skotland and James A. Traquair, Irrigated Agriculture Research and Extension Center, Prosser, WA 99350 and Agriculture Canada, Harrow Ontario, NOR 1G0.

In the spring of 1975 a sclerotium producing fungus was found associated with a rot of Scotch spearmint rhizomes. This fungus is morphologically similar to *Rhizoctonia solani* because the hyphal branches are constricted at the base with the formation of septum in the branch near the point of origin. The hyphae are 9-10 microns in diameter and possess moniloid cells, dolipore septa and multinucleate cells in the young hyphae. The mycelium is white turning light brown. The sclerotia however, are 2-4 mm in size and consist of moniloid cells with primitive rind. Young sclerotia are produced externally. Mycelial growth on potato dextrose-yeast extract agar is 7 mm per day at the optimum temperature of 20°C. Clamp connections or a spore state have not been observed.

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PLANT DISEASES IN AL-KHARJ REGION, CENTRAL SAUDI ARABIA. I.Y. Trabulsi, Z.M. Abul Hayja, and A.S. Al-Hazmi. Department of Plant Protection, College of Agriculture, Saud University, Riyadh, Saudi Arabia.

Plant diseases were surveyed in Al-Kharj region in central Saudi Arabia. During a 15 month period, 512 diseased plant samples were collected and causal organisms were identified. Of these, 282 were caused by fungi, 164 by viruses, 28 by phanerogamic plant parasites and 8 by bacteria. Fifty seven species of fungi, 2 species of bacteria, 6 viruses, 3 phanerogams and 2 physiological disorders were identified. The most abundant and commonly destructive diseases were foliar pathogens of alfalfa and WMV on squash. Few other diseases were occasionally severe in localized area.

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PYTHIUM MYRIOTYLUM DAMPING-OFF AND ROOT ROT, A NEW DISEASE OF CROWN VETCH. E. M. Dutky, Dept. of Botany, Univ. of Maryland, College Park, Maryland 20742 and R. D. Lumsden, Soilborne Diseases Lab., USDA SEA-AR, Beltsville, Maryland 20705.

Pythium myriotylum was associated with damping-off and root rot of crown vetch seedlings in fields in Dorchester County on Maryland's Eastern Shore. This fungus was isolated from diseased crown vetch roots in July 1981 and from debris in soil in March 1982. Pathogenicity of *P. myriotylum* was established in greenhouse tests which also included *P. aphanidermatum* and *P. ultimum*. *P. myriotylum* produced severe pre-emergent and post-emergent damping-off at 21 and 32 C. *P. aphanidermatum* produced moderate symptoms at 32 and mild symptoms at 21 C. *P. ultimum* produced moderate symptoms at 21 and mild symptoms at 32 C. This is the first report of *P. myriotylum* as a pathogen of crown vetch.

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PATHOGENICITY OF *DRECHSLERA SOROKINIANA* ON ROOTS OF TALL FESCUE. L. E. Trevathan, Department of Plant Pathology and Weed Science, Mississippi State University, Mississippi State, MS 39762.

Fescue is susceptible to *Drechslera sorokiniana* when conidial suspensions are sprayed onto leaves. The number of leaf lesions does not, however, necessarily indicate the degree of susceptibility of roots to infection. Seedlings of three tall fescue (*Festuca arundinacea*) cultivars were tested in the laboratory and greenhouse for development of root and foot rot. In test tube cultures containing conidial inoculum, brown to black elongate lesions and loose cortices developed. Symptom development in roots growing from seed planted in sterile and non-sterile infested soil was similar to that observed in test tube culture. Seedling emergence and vigor were functions of inoculation method, cultivar, root medium, and duration of exposure to the organism.

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ENVIRONMENTAL CONDITIONS CONDUCIVE TO DEVELOPMENT OF RYEGRASS BLAST. Michael A. Moss and Larry E. Trevathan, Department of Plant Pathology and Weed Science, P. O. Drawer PG, Mississippi State, MS 39762.

In greenhouse tests, 21-day-old ryegrass (*Lolium multiflorum* Lam.) plants were inoculated with eight conidial suspensions of *Pyricularia grisea* (Cooke) Sacc. ranging from 6.25×10^3 to 8.0×10^5 conidia per ml. Leaf position had a significant effect on susceptibility when inoculum concentrations were equal to or greater than 2.0×10^5 conidia per ml. At these concentrations, the third and fourth youngest leaves were more susceptible than the two younger leaves. Field tests were conducted to determine the effects of planting date and shading on disease development. Ryegrass was planted at 2-wk intervals from September 15 to November 1. Plots were inoculated with *P. grisea* 21 days after planting, covered with 30, 47, or 73% shade cloth, and misted continuously for 4 days. Based on lesion incidence and leaf dry weight recorded 3-wk after inoculation, incidence and severity of infection were greatest at the first planting date and shading reduced disease development.

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THE REACTION OF HOP, *HUMULUS LUPULUS*, VARIETIES TO PHYTOPHTHORA CITRICOLA. C.B. Skotland, TAREC-WSU, Prosser, WA 99350.

Rhizomes of 17 hop varieties were planted in a field naturally infested with *Phytophthora citricola*. After two years the

crowns were dug and examined for rot. The varieties Alliance, Brewers Gold, Bullion, Calicross, Cascade, Columbia, Comet, Talisman, Tettananger and Willamette were free of infected crowns. From 20-60% of the crowns were infected in the Early, Late and Yakima Cluster varieties. The infection appeared to start in the crown and spread to the roots. Occasionally isolated areas of roots were infected. In the laboratory tests, hop rhizomes were inoculated by placing a 5 mm diameter plug of mycelium and agar (PDA) in a 5x5 hole and sealed. The rhizomes were incubated for 5 days at 19°C. Ten rhizomes per treatment and 6 replications were used. Field resistant varieties showed less discoloration in the laboratory tests with the exception of Comet, which was the same reaction as the susceptible varieties.

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ALTERNATE CROPPING WITH PEAS PLUS SUDAN GRASS FOR CONTROL OF VERTICILLIUM WILT OF POTATO. Gene D. Easton and Michael F. Nagle. Washington State University, IAREC, Prosser, WA 99350.

Potato production and incidence of *Verticillium dahliae* in a soil previously cropped to peas + sudan grass were compared to that in portions previously cropped two successive years to potatoes. The peas + sudan grass portion has been seeded to sudan grass late in the summer following a spring crop of green peas, rototilled and plowed that winter and planted to potatoes the following year. The previous cropping treatments were replicated six times and the experiment was repeated twice. Previous cropping to peas + sudan grass did not noticeably reduce *Verticillium* wilt symptoms but did reduce by one-half the colonization of stems by *V. dahliae*. This rotation also increased percent U.S. No. 1 tubers 10 to 20%, increased specific gravity .002 to .008, and increased yield over 112 q/ha compared to previous cropping of potatoes. Fumigation of the soil previously cropped to peas + sudan grass did not increase potato quality or production; whereas, it did produce a significant increase in yield, but not quality, in soils previously cropped to potatoes.

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FUNGAL COLONIZATION OF SOYBEAN STEMS. T.S. Abney and T.L. Richards. Dept. of Botany & Plant Pathology, Purdue University, and USDA, ARS, West Lafayette, Indiana 47907/

Fungi colonized 'Wells II' stems at maturity in 1981. Nature and extent of colonization by *Diaporthe* (Phomopsis) spp. and by *Colletotrichum* spp. were determined for soybean plants in the second year of testing in the Integrated-Pest-Management systems at Lafayette, IN. A colonization pattern from the base of the stem towards the apex was apparent with *Colletotrichum* while *Diaporthe* colonization occurred along the stem axis without a definite pattern. Extensive colonization by both organisms was apparent when data were averaged across crop rotation, three tillage practices and three levels of weed management. *Diaporthe* colonization was more extensive with the continuous soybean cropping than in the corn-soybean sequence. *Colletotrichum* colonization was extensive with the corn-soybean sequence. However, when minimum tillage was involved, *Diaporthe* colonization was always more extensive than *Colletotrichum* colonization and especially with minimum weed management.

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REASSESSMENT OF THE ESSENTIALITY OF STEROLS FOR SEXUAL REPRODUCTION IN PHYTOPHTHORA. W. H. Ko and W. C. Ho, Department of Plant Pathology, University of Hawaii, Beaumont Agricultural Research Center, Hilo, HI 96720.

In 1964, scientists from four different institutes independently reported that sterols were required for oospore production by *Phytophthora*. The conclusion was based on observations that addition of sterols to basal agar media induced sexual reproduction. Moderate numbers of oospores were produced by *P. parasitica* in chemically-defined media with agar. However, very few oospores were produced in liquid media even in the presence of sterols or when Bacto agar was replaced by highly-purified Sea-Kem agarose. Elimination of agar also reduced the number of oospores produced by *P. cactorum* in defined media. Abundant oospores were produced by *P. parasitica* and *P. cactorum* in a basal medium containing lecithin but no sterols. Chromatographically-purified lecithin was more effective than β -sitosterol in promoting oospore formation by both fungi. These results indicate that sterols are stimulatory to, but not essential for, sexual reproduction in *Phytophthora*.

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HISTOPATHOLOGY OF CHRYSANTHEMUM ROOTS EXPOSED TO SALINITY AND PHYTOPHTHORA CRYPTOGEA. T.J. Swiecki and J.D. MacDonald, Dept. of Plant Pathology, Univ. of California, Davis, CA 95616

Rooted cuttings of *Chrysanthemum morifolium* 'Paragon' grown in nutrient solution were given a 24 h pulse exposure to salinity by amending the solution with 200 meq NaCl/l. After cuttings were returned to nonsalinized solution, half were inoculated with motile zoospores of *Phytophthora cryptogea*. Nonsalinated cuttings were also inoculated with *P. cryptogea*, and roots from all treatments were sampled at intervals for light microscopy. Penetration of nonstressed roots by *P. cryptogea* was frequently limited to 3-4 cell layers 6-12 h after inoculation. Formation of appositions adjacent to hyphae, increased wall staining density, and accumulation of osmophilic material in cell vacuoles were associated with sites of limited penetration. These changes were rarely observed in salinity-stressed roots, through which hyphae of *P. cryptogea* ramified rapidly causing extensive necrosis 12-24 h after inoculation. These results indicate that exposure of roots to salinity stress may interfere with their normal defense reactions.

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FORMATION AND INDIRECT GERMINATION OF SPORANGIA OF PHYTOPHTHORA PARASITICA AND P. CRYPTOGEA IN SALINE SOILS. N.S. Blaker and J.D. MacDonald, Dept. of Plant Pathology, Univ. of California, Davis, CA 95616.

Isolates of *P. parasitica* and *P. cryptogea* from a nonsaline, agricultural soil formed sporangia most abundantly in naturally saline or artificially salinized soils having salt concentrations less than 100 meq/l in the soil solution. In contrast, an isolate of *P. parasitica* from saline soil formed the maximum number of sporangia in soil at salt concentrations of 100-600 meq/l. Indirect germination by *P. cryptogea* was reduced if sporangia were formed in soil with salt concentrations greater than 50 meq/l, or if the solution into which sporangia were placed to germinate had a salt concentration equal to or greater than the soil in which the sporangia had formed. Effects of salinity on indirect germination by the isolate of *P. parasitica* from saline soils were generally similar, except that maximum indirect germination occurred when sporangia were formed in soil with 100-300 meq/l salt in solution.

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INFLUENCE OF SOIL MATRIC POTENTIAL ON INFECTION OF TOBACCO BY PHYTOPHTHORA PARASITICA VAR. NICOTIANAE. H. D. Shew. Department of Plant Pathology, N. C. State University, Raleigh 27650.

Soil matric potentials of -10 millibars (mb), -20 mb, and -50 mb were controlled by buchner funnel (6 cm diameter) tension plates. A one-month old 'Hicks' tobacco seedling was transplanted into a noninfested soil layer (65 g) overlying a layer of soil (65 g) naturally infested with *P. parasitica* var. *nicotianae* with an inoculum density of approximately 5 propagules/gram of soil. The soil was mixed with coarse sand (.5 to 2 mm) to give a 2:1 (v/v) soil:sand mix. Infection and severity decreased as water potential decreased. Percent infection after 3 wk averaged 60, 27, and 7% at constant matric potentials of -10, -20, and -50 mb, respectively. If soil at -50 mb was saturated (0 mb) for 24 hours after one wk of the test, and then returned to -50 mb for 2 wk, 100% infection resulted. Subsequent tests indicated that soil saturation periods as brief as one half hour completely overcame the inhibitory effect of low matric potentials on infection.

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HOST NON-SPECIFICITY OF PHYTOPHTHORA MEGASPERMA ISOLATES RECOVERED FROM 14 DIFFERENT PLANT SPECIES. W. F. Wilcox and S. M. Mircetich, USDA, ARS, Department of Plant Pathology, University of California, Davis, CA 95616

Mahaleb cherry seedlings grown in soils infested with *P. megasperma* isolates originally recovered from grape, pear, Douglas fir, kiwi, almond, cherry, juniper, walnut, apple, apricot, raspberry, or lilac had 95-100% root rot regardless of the isolate's origin; alfalfa (cv. 'Moapa 69') transplants had 50-90% less shoot and root fresh weights after 4 months in soils infested with these same isolates than did controls. Two soybean and four alfalfa isolates of *P. megasperma* caused 30% and 35% root rot of cherry and 35% and 99% alfalfa shoot and root weight loss, respectively. *P. megasperma* was reisolated from diseased roots of all host-pathogen combinations but one (cherry-soybean). Isolates varied in their virulence on the test hosts, but all were pathogenic. A lack of host specificity among various isolates of *P. megasperma* in these studies suggests that the use of "formae speciales" designations to separate specific subgroups within this taxon may be inappropriate.

MORPHOLOGICAL DIFFERENTIATION OF HOST SPECIALIZED GROUPS OF *PHYTOPHTHORA MEGASPERMA*. Hansen, E. M. and Hamm, P. B., Dept. of Botany & Plant Pathology, Oregon State Univ., Corvallis, OR 97331.

Pathogenicity, morphology, and growth of 54 isolates of *Phytophthora megasperma* from Douglas-fir, soybean, alfalfa, clover, and 10 other hosts were compared. Soybean and clover isolates showed pathogenicity specific to host of origin. Douglas-fir and alfalfa isolates formed three pathogenicity groups: D1, specific to Douglas-fir; A1, specific to alfalfa; and D2-A1, weakly pathogenic on both hosts. D1 isolates caused a severe reaction in soybean after hypocotyl inoculation, but caused only slight root rot after soil infestation. Isolates from the 10 other hosts did not cause disease on alfalfa, Douglas-fir, soybean, or clover. Isolates grouped by pathogenicity had similar morphology, growth at several temperatures, and sensitivity to the fungicide metalaxyl. No single morphological character, of the 10 compared, differentiated all host groups, but clover, soybean, D1, A1, and D2-A1 isolates were clearly separated from each other by the combination of oogonium diameter, sporangium length, and temperature response. Most isolates from hosts other than Douglas-fir, soybean, clover, and alfalfa were similar in morphology and growth to D2-A1 isolates. The host groups could not be assigned to var. *megasperma* or var. *sojae* because many isolates had oogonia of intermediate diameters.

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QUANTIFICATION OF TREATMENT EFFECTS ON NATURAL INOCULUM OF *PHYTOPHTHORA MEGASPERMA* F. SP. *GLYCINEA*. C. H. Canaday and A. F. Schmitthenner, Dept. of Plant Pathology, Ohio Agricultural Research and Development Center, Wooster, OH 44691.

A standardized baiting procedure has been used to quantify the effects of fungicides and fertilizers on natural inoculum of *Phytophthora megasperma* f. sp. *glycinea*. Naturally infested soil is mixed with water in a Waring blender to make a 1:1 (w:w) soil slurry. The slurry is poured into cylinders on stacks of paper towels, and excess water is absorbed. Uniform cubes of soil are cut from the resultant soil cakes. Cubes are incubated under conditions suitable for oospore germination and sporangium production but not zoospore release. Cubes are then flooded with water, baited with soybean leaf discs, and the leaf discs plated on a selective medium. Leaf disc infection is proportional to the amount of inoculum present. The effects of chemicals applied to the soil on inoculum levels can be quantified. Modifications of this procedure have been used to study the effects of physical treatments on natural inoculum.

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POPULATIONS OF *PYTHIUM* SPP. AND ASSOCIATED MICROORGANISMS IN A MINIMUM TILLAGE-MULTICROPPING SYSTEM. P. A. Rayside, D. J. Mitchell, & R. N. Gallaher, Univ. of Florida, Gainesville 32611

Populations of soil microorganisms were monitored every 4 weeks during the 5th year of a soybean (*Glycine max* 'Bragg')-oat (*Avena sativa* 'Florida 501') rotation under conventional and minimum tillage. Populations of total *Pythium* spp. over one season of oats and two seasons of soybeans ranged from 55 to 163 propagules per gram of soil (ppg). Generally, there were no significant differences in total populations among tillage practices (conventional, minimum with subsoiling, or minimum without subsoiling), but populations were influenced by changes in the crop and the environment. *Pythium irregulare* was the predominant species found, with populations gradually increasing during the beginning and dropping at the end of each soybean and oat season. Populations of *P. myriotylum* were found at levels greater than 5 ppg of soil only during the first month of the soybean season. Populations of *Rhizoctonia* spp., total fungi, bacteria and actinomycetes were influenced with changes in the crop and soil moisture, but generally not with tillage practice.

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DISTRIBUTION OF *PYTHIUM APHANIDERMATUM* IN THE RHIZOPLANE OF FIELD-GROWN SUGARBEETS. Stanghellini, M.E., Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

Soil sections bearing the impression (rhizoplanes) of mature sugarbeet tap roots were collected from a field with a *Pythium aphanidermatum* (PA) population of 9 oospores/g of soil. The distribution of PA in 38 rhizoplanes was determined by two methods. Depending on the surface area of each rhizoplane, 4 to 20 consecutive soil samples, each measuring 1 cm² and 0.1 cm deep, were individually scraped from 12 rhizoplanes and assayed for PA on a selective medium (SM) (Phytopath. 63: 1499-1500). Additionally, 4 to 13 slices of potato tuber tissue, measuring 1 cm² and 0.3 cm thick, were randomly placed on 26 rhizoplanes. Percent colonization of slices by PA was determined after 48 hr incubation at 27 C. Using the SM, PA was isolated from 83% of the rhizoplanes and a mean of 32% (range: 11-75%) of the total surface area (125 cm²) of infested rhizoplanes. Using the potato method, PA was isolated from 73% of the rhizoplanes and a mean of 36% (range: 10-90%) of the total surface area (166 cm²) of infested rhizoplanes.

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ROOT ROT OF HYDROPONIC-GROWN SPINACH CAUSED BY *PYTHIUM APHANIDERMATUM* AND *PYTHIUM DISSOTOCUM*. M.L. Bates and M.E. Stanghellini, Environmental Research Laboratory and Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

Root rot is currently the limiting factor to commercial production of spinach, *Spinacia oleracea*, in greenhouse recirculating hydroponic systems in Arizona. Infected plants either die or are severely stunted. Two species of *Pythium*, *P. aphanidermatum* and *P. dissotocum*, are associated with the root rot. *Pythium aphanidermatum* predominated as the primary causal agent of root rot during the warm summer production months when water temperatures are greater than 23 C, whereas *P. dissotocum* predominated as the primary or sole causal agent of root rot during the cool winter production months when water temperatures are between 18 and 22 C.

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THE EFFECTS OF Cl⁻ AND *PYTHIUM OLIGANDRUM* ON THE ECOLOGY OF *PYTHIUM ULTIMUM*. F. N. Martin and J. G. Hancock, Dept. Plant Pathology, University of California, Berkeley, CA 94720.

Suppressiveness to *Pythium ultimum* is frequently associated with finely textured basin soils in the Oxalis and Lethem soil series in California's San Joaquin Valley. Saprophytic colonization of virgin organic substrates (e.g. cotton leaves) by *P. ultimum* is reduced in these Low-Pyrium (LP) soils. Quantitative comparisons of saprophytic colonization in High-Pyrium (HP) and LP soils indicate that soil Cl⁻ concentrations and inoculum densities of *Pythium oligandrum* are correlated with soil suppressiveness to *P. ultimum*. However, results show that Cl⁻ alone cannot entirely account for the differences in behavior of *P. ultimum* in LP and HP soils. *Pythium oligandrum* can effectively compete with *P. ultimum* for organic substrates when its inoculum densities are greater than *P. ultimum*. The addition of Cl⁻ or *P. oligandrum* to HP soils at levels encountered in LP soils significantly reduces the saprophytic activities of *P. ultimum*. *Pythium oligandrum* is more tolerant of Cl⁻ than *P. ultimum*, a characteristic that may allow it to dominate *P. ultimum* in LP soils.

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EFFECT OF SOYBEAN SEEDLOT QUALITY ON THE RELATIONSHIP OF DAMPING-OFF TO INOCULUM DENSITY OF *PYTHIUM ULTIMUM*. R. S. Ferriss, Department of Plant Pathology, University of Kentucky, Lexington, Ky 40546.

Six soybean (cultivar Williams) seedlots, all with less than 5% infection by pod and stem blight fungi, were evaluated using standardized tests of seed quality. Seeds from each seedlot were planted in pasteurized soil infested with *P. ultimum* at 1 to 50 sporangia per gram (spg) soil. For all seedlots, incidence of damping-off 2 weeks after planting (DI) increased with increasing inoculum density below 10 spg. For 5 of the 6 seedlots, DI was less than 100% of all IDs. For 3 seedlots DI reached plateaus of 39, 65, and 69% damping-off above 10 spg. Relative susceptibility of the seedlots to damping-off correlated better with the results of an accelerated aging test than a seed conductivity test or a standard germination test.

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THE PASSIVE SURVIVAL OF *PYTHIUM ULTIMUM* AT CONSTANT SOIL TEMPERATURES AND WATER MATRIC POTENTIALS. R. Lifshitz and J. G. Hancock, Dept. Plant Pathology, University of California, Berkeley, CA 94720.

Changes in the germinable propagule (GP) densities of *Pythium ultimum* were followed at constant soil water potentials (ψ_m) and temperatures. Two distinct phases of declines in GP densities were noted. The initial decline was more rapid at the higher temperatures when they ranged between 9 and 27 C and more rapid at -0.4 ψ_m than at -3 bars. After 30 to 90 days of incubation a distinct slower rate of decline of GP densities was noted in moist soil. The initiation of the second phase was associated with the time of ripening of dormant oospores. Indirect propagule assays and direct microscopic observations showed significant oospore ripening at 15 to 27 C in moist soils (ψ_m of -3 and -0.4 bars) after 6 weeks, but not in air dried soil. The delayed ripening of oospores apparently accounts for the survival of *P. ultimum* during periods of drought and low temperatures. On the other hand, the most rapid ripening of oospores appears to coincide with soil environmental conditions most favorable for disease.

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REDUCTION OF *PYTHIUM ULTIMUM*, *THIELAVIOPSIS BASICOLA* AND *MACROPHOMINA PHASEOLINA* POPULATIONS IN SOIL FROM NH₃ GENERATED FROM UREA. D. Chun and J. L. Lockwood, Department of Botany and Plant Pathology, Michigan State Univ., East Lansing, MI 48824

Sandy loam and loam soils were infested with *Thielaviopsis basicola* chlamydospores, *Macrophomina phaseolina* sclerotia, and

Pythium ultimum sporangia and placed in cylindrical clay tiles [20.3 (diam.) x 30.5 cm] sunk 28 cm into a field. Urea was added to the soils to establish concentrations of 0.1% and 1% (w/w). *Pythium* population was 2/3 of the control 12 days after addition of 0.1% urea, and 1/20 of the control after 31 days. *Pythium* was undetectable 4 days after addition of 1% urea. *Thielaviopsis* population was 1/3 of the control with 0.1% urea after 31 days. Population with 1% urea after 4 days was 2/5 of the control and was undetectable after 12 days. *Macrophomina* population was 1/2 of the control after 31 days, with 0.1% urea. With 1% urea, the population was about 1/5 of the control after 4 days, and after 12 and 31 days was near undetectable levels. Increased soil pH and ammonia generated from urea was associated with decreases in pathogen densities.

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IMPROVEMENTS IN ASSAYS FOR SOIL POPULATIONS OF *PYTHIUM ULTIMUM* AND *MACROPHOMINA PHASEOLINA*. D. Chun and J. L. Lockwood, Dept. of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824.

Field assays of populations of *Pythium ultimum* and *Macrophomina phaseolina* were facilitated by improvements to previous methods. *Pythium* was enumerated by suspending soil in 0.2% water agar and dispersing 1 ml aliquots into 2.5 mm diam. x 2 mm wells made in 2% water agar plates at 0.01 ml per well. Hyphal growth from *Pythium* propagules was easily observed growing from the walls of the agar wells. The advantages are: more rapid reading of samples, economy of materials, and higher population counts than obtained from dispensing as small drops on the agar surface. *Macrophomina phaseolina* sclerotia in soil were efficiently recovered by 60% sucrose buoyancy floatation centrifugation. The sclerotia were collected from the surface by suction onto 20 micron or smaller nylon mesh screens. The sclerotia were rinsed, surface-sterilized, suspended in 0.2% water agar, and plated on a selective medium. The procedure expedites processing of large numbers of field samples.

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SOIL-BORNE BACTERIOPHAGE AS A LIMITING FACTOR IN ROOT COLONIZATION BY BENEFICIAL RHIZOBACTERIA. T. V. Suslow, Advanced Genetic Sciences, Inc., Berkeley, CA.

A soil-borne bacteriophage has been shown to be potentially limiting to the colonization and growth-promotion of a beneficial rhizobacteria. Phage was recovered from rhizosphere soil of sugar beets, treated with strain SH5, and grown in *Hesperia* fine sandy loam (HSL). Population densities of SH5, 3.0×10^2 cfu/cm root, were far below those in previous trials. Four phage-resistant, spontaneous mutants of SH5 reached population densities averaging 2.5×10^4 cfu/cm root in HSL, in the presence of 1.0×10^7 plaque-forming units (pfu)/g soil, as compared to 3.7×10^2 cfu/cm root for the phage-sensitive strain. Naturally occurring SH5-infecting phage in HSL were detected at 1.7×10^3 pfu/g air-dried soil. Phage infection prevented plant growth-promotion by phage-sensitive rhizobacteria strain SH5 (2.9 g/plant) as compared to phage-resistant SH5 (5.3 g/plant). Two of 12 other beneficial rhizobacteria were sensitive to the phage. Phage-limiting effects on SH5 colonization were only significant in soil maintained near field capacity.

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SIDEROPHORE PRODUCTION BY *PSEUDOMONAS FLUORESCENS* IN SOIL EXTRACTS. M.W. Olsen and I.J. Misaghi. Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

The effect of soil pH on siderophore production by fluorescent pseudomonads was studied using extracts of nontreated field soil (pH 7.9) and of soils adjusted in the field with sulfur to pH 2.9, 4.1, or 5.8. The soil extracts were filtered and adjusted to pH 7.0. Extracts were amended with different levels of succinate broth (1.0% sodium succinate, 0.6% K_2HPO_4 , 0.3% KH_2PO_4 , 0.02% $MgSO_4$ and 0.1% NH_4SO_4), filter sterilized, and inoculated with *Pseudomonas fluorescens* at 25°C. All the amended extracts supported bacterial growth, but siderophore production was substantially reduced with decreasing soil pH. The results show that siderophores may be produced by fluorescent pseudomonads in certain soils in the presence of appropriate nutrients. However, siderophore production may be reduced or inhibited around the rhizosphere of some plants where the level of available iron (an inhibitor of siderophore production) may be high due to the low pH.

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INFLUENCE OF BACTERIAL SOURCES OF INDOLE-3-ACETIC ACID (IAA) ON ROOT ELONGATION OF SUGAR BEET. J. E. Loper, M. N. Schroth and N. J. Panopoulos. Dept. Plant Pathology, University of California, Berkeley, CA 94720.

IAA production by rhizosphere-colonizing bacteria has been associated with symptoms of root deformation and stunting. The influence of this exogenous source of IAA on root elongation was elucidated by applying the IAA-producing *Pseudomonas savastanoi* strain 2009-6 to sugar beet seed. Washed cells of strain 2009-6 caused a 55% decrease in root elongation of sugar beet. Two derivative *Ps. savastanoi* strains, 2009-3 and 2009-561, deficient in IAA production caused no such effects. Results indicate that IAA produced by strain 2006-9 in the presence of the developing root can influence root elongation of sugar beet. Plant growth-promoting rhizobacteria and deleterious rhizobacteria were screened for *in vitro* production of IAA. Fifteen percent of beneficial bacteria and 53% of deleterious bacterial strains produced IAA *in vitro* as indicated by a positive colorimetric reaction upon the addition of Salkowski reagent to culture filtrates. These IAA producing strains cause root deformity and decreased root elongation when applied to sugar beet.

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CHEMOTAXIS OF BACTERIA TO FUNGAL PROPAGULES. D.K. Arora, A.B. Filonow and J.L. Lockwood. Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824.

Chemotaxis of *Erwinia herbicola*, *Pseudomonas fluorescens* and *P. putida* to conidia of *Cochliobolus victoriae* and sclerotia of *Macrophomina phaseolina* was studied. Open-ended capillaries (1 µl, 3 cm long) were filled with exudate or buffer; one end was inserted into a chamber containing a propagule suspension or buffer, and the other end into a bacterial suspension. Bacteria accumulated in capillaries containing exudate and in chambers containing fungal propagules. For example, number of bacteria accumulating in capillaries inserted into a *C. victoriae* conidial suspension, after 90 min, was 23-48-fold greater than when the chamber contained buffer. Accumulation was greater in response to *C. victoriae* than to *M. phaseolina*. In general, *P. fluorescens* exhibited greater accumulation than *E. herbicola* and *P. putida*. Movement of bacteria towards the same fungal propagules and their exudates in a sandy loam soil also was shown.

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EXUDATION OF ^{14}C FROM FUNGAL PROPAGULES IN THE PRESENCE OF SPECIFIC MICROORGANISMS. D.K. Arora, A.B. Filonow and J.L. Lockwood. Dept. of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824.

Exudation of ^{14}C compounds from labelled conidia of *Cochliobolus victoriae* and sclerotia of *Macrophomina phaseolina* in the presence of bacterial cells or fungal propagules was examined in infested sterilized sandy loam soil and in phosphate buffer (0.05 M, pH 6.8). ^{14}C exudation was taken as the sum of ^{14}C respired and that fixed by microbial cells or free in the soil or solution. Bacteria caused greater exudation than fungi. ^{14}C exudation in the presence of microorganisms was 1.5-9.9% and 1.0-3.8% for *C. victoriae* *in vitro* and on soil, respectively, and was 1.7-3.2% and 1.3-3.3% for *M. phaseolina* *in vitro* and on soil, respectively. Exudation of ^{14}C by *C. victoriae* conidia was correlated ($r = -0.72$) with inhibition of germination. The results indicate that specific microorganisms can enhance exudation from fungal propagules, and that enhanced exudation may be related to the induction of fungistasis.

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POSSIBLE BIOTIC CAUSE OF CORKY ROOT OF LETTUCE. A. A. Cook, Department of Plant Pathology, University of Florida, Gainesville, FL 32611

The cause of corky root, an important field problem in some lettuce growing areas, has been a subject of controversy. Plants of various lettuce types are susceptible in all stages of development. Early plant infection is followed by progressive degeneration of the fibrous root system and subsequent death of the plant. Late infection results in severe reduction of the fibrous root system and extensive, external corking of the tap root with notable yield reduction but plants may live. Factors found by previous investigators to influence occurrence of the disease have led to the assumption that a pathogen is involved rather than the disease being the result of abiotic conditions. There has been general agreement among previous investigators that the causative agent is soil-borne but no pathogen has been isolated or identified. Pieces of diseased lettuce roots collected from plants grown near Belle Glade, FL were examined histologically and found to include stages of an organism belonging to the Plasmodiophorales.

EFFECT OF NITROGEN SOURCE AND RATE ON BACTERIAL SPOT OF PEPPER. John Paul Jones, Jeff B. Jones, and S. S. Woltz, University of Florida, AREC-Bradenton, Fla. 33508.

Nutrient culture techniques were used to determine the effect of nitrogen (N) source (80:20, 50:50, 20:80 $\text{NH}_4:\text{NO}_3$) and rate (60, 120, 240, 360 mg/l) on the development of bacterial spot of 'Early Cal Wonder' pepper incited by *Xanthomonas campestris* pv. *vesicatoria*. Plants were inoculated by misting with a 10^7 CFU/ml suspension, incubated 24 hrs in a dew chamber, and placed in a greenhouse. Tissue N decreased with decreasing N rate. Nitrogen sources equally affected tissue N whereas tissue Mg was decreased by an increase in $\text{NH}_4\text{-N}$. Disease development was significantly affected by N source, the least disease being associated with 80% $\text{NH}_4\text{-N}$. There was a significant rate X source interaction so that disease severity decreased with decreasing rate where 80% $\text{NH}_4\text{-N}$ was used.

THE EFFECTS OF BOTRYTIS ALLI INFECTION OF THE SEED STOCK (SCAPE) OF YELLOW SWEET SPANISH ONION ON SEED YIELD AND WEIGHT, GERMINATION, AND SEEDLING VIGOR. G. F. Stallknecht and J. Garrison, Montana State Univ., Huntley, MT 59037 and Res. Assoc., Desert Seed Co., Brooks, OR 97305.

The effect of *Botrytis alli* was evaluated on three yellow sweet Spanish onion cultivars grown under field conditions in south-west Idaho. Browning and desiccation of the lower portion of umbel and scape were observed during the later flowering and early seed development. Seed yield and weight were significantly reduced. Average seed yield from healthy and diseased plants was 5.17g and 1.92g, respectively, while seed weight per 300 seeds was 1.36g and 0.95g, respectively. Seed germination averaged 79% from healthy umbels and 62% from the diseased umbels. Seedlings grown under greenhouse conditions averaged 240 mg from seed produced on healthy umbels as compared to 114 mg from seed produced on diseased umbels.

EVALUATING SWEET POTATO FOR REACTION TO SCLEROTIAL BLIGHT (SCLEROTIUM ROLFII Sacc.) IN FIELD PLANT BEDS. P. D. Dukes, Alfred Jones and J. M. Schalk; USDA, ARS, U. S. Vegetable Laboratory, Charleston, SC 29407.

An efficient method has been developed for evaluating breeding lines and cultivars of sweet potato for reaction to sclerotial blight (SB) in field plant beds. Disease evaluations were made during the normal course of plant production, and highly susceptibles were readily identified and removed from the breeding program. Beds were prepared in the normal manner in a well-drained sandy soil infested with the pathogen. After bedding, roots were covered with soil containing sclerotia of the fungus, then a black plastic mulch was applied directly on the soil surface and sealed at the edges. The mulch was left on so that heat damage was sustained by the emerging shoots. Upon removal of the mulch, the plants recovered quickly and disease evaluations for SB were made. Plots of 'Jewel', a resistant cultivar, and W-13, a USDA line with high susceptibility, were randomly placed throughout the beds as disease standards.

CLUB ROOT DISEASE CONTROLLED WITH SPENT SUGARBEET LIME. Great-head, A.S., Cooperative Extension Service, 118 Wilgart Way, Salinas, CA 93901; R.N. Campbell, D.F. Myers, Dept. of Plant Pathology, U.C. Davis, Davis, CA 95616.

Two year's of field trial work including three small hand-applied trials and two commercially-applied trials have been conducted on the control of club root disease in the Salinas Valley of California. Substantial reduction of disease to commercially-acceptable levels has been consistently obtained by incorporating between 4484 and 33627 kg/ha of spent sugarbeet lime (60% CaCO_3) into the soil prior to direct-seeding of broccoli and cauliflower. Applications below 4484 kg/ha resulted in inconsistent levels of control. Trials established in 1980 were replanted in 1981 with no further additions of lime resulting in the same level of control. pH levels of the soil after lime incorporation were as low as 6.7 in plots where good control was obtained.

CLUBROOT OF CRUCIFERS: INTERACTION OF PH AND NUTRITION IN DISEASE CONTROL. Donald F. Myers and Robert N. Campbell, Dept. of Plant Pathology, Univ. of California, Davis, CA 95616

Infection of broccoli roots by *Plasmodiophora brassicae* Wor. is

influenced by the pH and the nutrient status of the soil. We used a nutrient-sand culture system with Good buffers to study the relative importance of these factors. While numerous root hair infections and clubroot development were observed at pH < 7.1, root hair infections were reduced significantly, but not eliminated, at pH > 7.3, and neither secondary zoospores nor clubroot were observed. If Ca or Mg were reduced to low or deficient amounts, limited disease development could occur at pH 7.2-7.6. Increasing the concentrations of Ca or Mg in the system reduced infections. These reductions were mediated by pH; eg., 3-7 meq Ca/l significantly reduced infections at pH 7.2, while 15-50 meq Ca/l were necessary for the same effect at pH 6.8. Disease control by soil liming may be due to a soil pH > 7.2 and/or to an increase in the concentration of Ca or Mg to the equivalent of 15 meq/l or greater at pH 6.8-7.2.

EFFECTS OF METALAXYL-TREATED TOMATO TRANSPLANTS ON PLANT SURVIVAL AND FRUIT YIELD. S. H. Kim and C. A. Jaworski, BPI, PA Dept. of Agr., Harrisburg, PA 17110 and ARS, USDA, Coastal Plain Station, Tifton, GA 31793.

Tomato transplants, cv. Peto 98, were sprayed with metalaxyl at 0, 0.28 and 0.56 kg a.i./ha 2 hr. prior to pulling from a Georgia field. The transplants were stored for 4, 6, 9 or 11 days before transplanting to a Pennsylvania field fumigated with methyl bromide-chloropicrin. Plot size was 1.5 x 30.5 m and each treatment was replicated 4 times in a randomized complete block design. Fruit harvests were made 8 times from 2-m segments in a row by pulling the entire plant at weekly intervals, 70 to 119 days after each planting date. Transplant mortality rate was reduced by metalaxyl at both 0.28 and 0.56 kg a.i./ha. Initial growth rate, height and width were more vigorous with metalaxyl than nontreated. Yield of disease-free fruit per ha increased with metalaxyl but not per plant yield. Individual fruit weight, number of diseased fruit per plant and percent diseased fruit by number were not affected by metalaxyl. Yield of disease-free red fruit per ha increased with metalaxyl.

COMBINING RESISTANCE TO ROOT ROT AND BACTERIAL BROWN SPOT IN PHASEOLUS VULGARIS. D. J. Hagedorn and R. E. Rand, Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706.

The two major diseases of processing beans in Wisconsin are root rot, incited primarily by a complex of *Pythium* sp. and *Aphanomyces euteiches*, and bacterial brown spot (BBS), incited by *Pseudomonas syringae*. We developed beans resistant to root rot by hybridization and recurrent selection under field conditions beginning with resistant selections from Oregon and New York breeding lines, and then adding resistance genes from other sources. High level resistance to BBS was discovered in 1972 in PI 313537. Selected plants were hybridized with commercial cultivars and the resistant Wis. (BBSR) 130 breeding line was released in 1977. Recently crosses have been made between our best root rot resistant lines and those resistant to BBS. In 1980-81, BBS greenhouse tests were made on 930 plants, with 122 resistant plants selected. Field root rot determinations were made on 46 of the best lines containing ca 4,000 plants; 52 doubly resistant plants were selected. These studies indicate resistance to root rot and BBS can be successfully combined.

EXPRESSION AND GENETIC CONTROL OF RESISTANCE TO PSEUDOMONAS SYRINGAE AT DIFFERENT STAGES OF DEVELOPMENT OF BEAN, PHASEOLUS VULGARIS. S. H. Antonius and D. J. Hagedorn, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Resistance to bean bacterial brown spot (bbs) was laboratory tested at 3 different stages of plant development, i.e. seedling, foliage, pod and in the field. Tests involving 16 *P. vulgaris* lines indicated that expression of resistance is positively and significantly correlated among testing methods. No evidence was found to suggest separate genetic control for the developmental stages. Correlations among testing methods in segregating populations from R x S crosses were also positive although often lower than those obtained with pure lines. No recombinant lines were recovered following 4 generations of selection for foliage resistance and pod susceptibility. Either a single genetic mechanism is responsible for the expression of resistance to bbs in foliage and pods, or the genes controlling expression of resistance are tightly linked, in contrast to common blight and halo blight where pod and foliage responses are reportedly under separate genetic control.

RESISTANCE IN COWPEA TO BACTERIAL BLIGHT AND CANKER. R. D. Gitaitis. Department of Plant Pathology, University of GA, Coastal Plain Exp. Stn., Tifton, GA 31793.

Primary leaves of 50 collections of cowpea (*Vigna unguiculata* (L.) Walp were infiltrated with a concentrated suspension (10^8 cfu/ml) of *Xanthomonas campestris* pv. *vignicola* (Burkholder) Dye. Confluent necrosis visible within 24 hr occurred in some or all of the replications in 'Acre', 'Brabham', 'Brabham K892', 'Chinese Red' x 'Iron', 'Iron', 'Iron K329', 'Jackson Bunch', 'Jackson Bunch Purplehull', 'New Era', 'Sekgald', 'Suwanee', and 'Victor'. Multiplication of bacteria in vivo confirmed that the observed reaction of these cowpea lines was a hypersensitive response (HR). Mean concentration of bacteria after 5 days in a disc (1 cm dia) of necrotic leaf tissue from cowpeas exhibiting HR was 5.2×10^6 cfu/ml. Bacterial concentration in susceptible lines was 1.2×10^9 cfu/ml/disc of necrotic tissue. Reactions on primary leaves of Brabham K892 with 10 isolates of *X. campestris* pv. *vignicola* from diverse origins have been negative for the presence of a second race.

THE INHERITANCE OF THREE SOURCES OF RESISTANCE TO BACTERIAL SPOT OF PEPPER. W. C. Adamson and G. Sowell, Jr., USDA, ARS Regional P.I. Station, Experiment, GA 30212

Resistant pepper PI's 163189, 163192 and 322719 and the susceptible 'Yolo Wonder' were crossed and backcrossed in all possible combinations. The F_3 progenies, backcross (BC) F_2 progenies and the parental lines were sprayed with a suspension of *Xanthomonas vesicatoria*. Coefficients of variation were calculated with the numbers of plants classed as resistant in plots of the susceptible cultivar and with the numbers of susceptible plants in plots of resistant lines. Populations falling between these percentages were considered as segregating. Based upon the number of F_3 and BC F_2 populations that were resistant, segregating or susceptible the genetic compositions of the F_2 and BC plants were determined. PI 163192 carries a single dominant gene as previously reported. PI 322719 carries a different single dominant gene. PI 163189 carries 2 or more additive genes, at least one of which is linked with the locus of the dominant gene of PI 163192.

EVALUATION OF POPCORN VARIETIES FOR REACTION TO GOSS'S BACTERIAL WILT AND BLIGHT DISEASE. David S. Wyson, Benjamin Doupnik, Jr., and John Linscott. University of Nebraska, Lincoln, NE 68583-0722.

Sixteen inbred lines and 38 hybrid popcorn varieties were evaluated for Goss's bacterial wilt reaction over a two-year period. A randomized complete block design with four replications was used for each of two field locations in each year. The fields had a previous history of severe infection, and no artificial inoculation was done during this study. By late August, the disease reached epidemic levels in one of the two fields in both years. Inbred lines averaged 2.9 in disease intensity, and hybrids 3.4 (0 = no disease; 6 = dead plants). Disease intensity ranged between 1.5 (Purdue Expt. 42235) and 5.0 (A328 and Robust 49-49) among hybrids. Inbreds yielded 40% less grain, and hybrids 33% less, in the two severely infected fields than in the remaining fields having lower infection. There was no significant influence of the disease on popcorn volume expansion.

GREENHOUSE AND LABORATORY METHODS FOR IDENTIFYING RESISTANCE IN SWEET POTATO TO *STREPTOMYCES IPOMOEA*. J. W. Moyer¹, C. L. Campbell¹, E. Echandi¹, and W. W. Collins², Depts. of Plant Pathology¹ and Horticulture Science², North Carolina State University, Raleigh, NC 27650.

A system for evaluation of sweet potato resistance to *Streptomyces ipomoea* was developed. *S. ipomoea* was grown on sterile sand, amended with 12g horse manure and 2g $CaCO_3$ per Kg sand, for at least 4 wk at 30C. Greenhouse trials were conducted in 15 cm clay pots (2 plants/pot) containing infested medium diluted with sand to 10^6 colony forming units/g sand. In laboratory tests tuberous root slices (2 cm thick) of each cultivar were incubated 2 wk at 30C on infested media (10^6 and 10^5 colony forming units/g media). There was a significant correlation ($P=0.05$) between disease severity reactions of resistant and susceptible cultivars and breeding lines observed under greenhouse and under laboratory conditions and performance of the lines in field plots artificially infested with *S. ipomoea*.

BREEDING FOR TOLERANCE TO PEA APHANOMYCES ROOT ROT. D. J. Hagedorn and E. T. Gritton, Depts. of Plant Pathology and Agronomy, respectively, Univ. of Wisconsin, Madison, WI 53706.

Mutation and conventional breeding was used to develop improved tolerance to root rot of *Pisum sativum* incited by *Aphanomyces euteiches*. Mutations were induced by soaking seeds of peas with reputed tolerance in ethyl methanesulfonate. After inoculation tests in the greenhouse and field testing in highly infested soil, progenies of the most tolerant plants were intercrossed to accumulate and concentrate genes for tolerance. In 1980 ca 400 entries were tested in plots at two locations, and 122 were chosen for further tests. In 1981, 101 of these lines were again evaluated at the two locations (Arlington and Hancock). The 10 best entries at Arlington had a mean yield of 5.89 g per plant in contrast to 3.83 for all entries and 3.08 for the susceptible control. The comparative data for Hancock were 2.72, 1.12, and 0.82, respectively. Five of the best entries at Arlington were among the 10 best at Hancock. These experiments indicate measurable progress toward development of *Aphanomyces* root rot tolerance in peas.

EVALUATING FOR RESISTANCE TO WHITE LEAF SPOT INCITED BY *ARISTASTOMA OECONOMICUM* (ELLIS & TRACY) TEHON IN SOUTHERN PEA (*VIGNA UNGUICULATA* (L.) WALP.). L. G. Brown and P. D. Dukes, Clemson University, Summerville, SC 29483; USDA, Vegetable Breeding Laboratory, Charleston, SC 29407.

A quick and reproducible greenhouse test for resistance was needed since field testing has proven too variable. Pea leaf agar (PLA) made from the supernate of 200 g/liter of washed and boiled leaves of *V. unguiculata* proved superior overall to carrot potato dextrose agar (C-PDA), potato dextrose agar (PDA) and V-8 juice agar (V8-A). Optimum temperature for pycnidial production and radial growth of the fungus on PLA was 25°C while 30°C and 10°C were the maximum and minimum temperatures respectively. On water agar, under continuous dark, spore germination averaged 93% between 20-30°C. The percent germination dropped to 68% at 10°C and 2.3% at 35°C. Plants at the first trifoliate stage were sprayed with $> 5 \times 10^5$ pycnidiospores/ml in H_2O plus 0.1% Tween 20 from 2-3 week-old cultures. Plants were maintained under plastic for 24 hours. Evaluations for resistance were made at 14 days.

A TECHNIQUE FOR SCREENING ONION SEEDLINGS FOR PURPLE BLOTCH RESISTANCE. Marvin E. Miller, Texas Agricultural Experiment Station, Weslaco, Texas 78596.

Onion seedlings at the 2-3 leaf stage were inoculated with 850 *Alternaria porri* spores/ml, incubated for 24 hours in a dew chamber at 23.9°C and then placed under light banks for 72 hours at 23.9°C. Spores were suspended in a hydrolyzed starch-polyacrylonitrile graft copolymer to improve adhesion to the small, waxy onion leaves. Purple blotch ratings were taken after 72 hours under lights. The seedlings were then replanted in 7.62 cm pots, grown to the 5-6 leaf stage, reinoculated as described above and purple blotch ratings taken again. Results from both inoculations were similar to the reactions obtained on mature plants in field plots. Ratings for purple blotch resistance were taken on 117 breeding lines. Progeny from crosses of Beth Alpha and New Mexico Yellow Grano resulted in 1 line with a high level of resistance and 3 lines with medium levels of resistance.

NEW RACE OF *PERONOSPORA EFFUSA* VIRULENT ON RESISTANT SPINACH IN VIRULENT ON RESISTANT SPINACH IN TEXAS. R. K. Jones, Plant Pathologist, Texas Agricultural Extension Service, and F. J. Dainello, Research Horticulturist, Texas Agricultural Experiment Station, Uvalde, Texas 78801.

A new race, designated race 3, of *Peronospora effusa* capable of attacking spinach (*Spinacia oleracea*) resistant to races 1 and 2 has been found in Texas. Race 3 currently causes severe damage on fresh market savoy spinach. A source of resistance, based on the gene designated M_3 , has been identified in some breeding lines and commercially available hybrids. A technique for resistance screening is described.

BREEDING FOR CLUBROOT, DOWNY MILDEW AND TURNIP MOSAIC RESISTANCE IN CYTOPLASMIC MALE STERILE BRASSICA CAPESTRIS L. H. LEUNG, XIN-KE NIU, AND P. H. WILLIAMS. Dept. Plant Pathology, Univ. of Wisconsin, Madison, WI 53706.

Cytoplasmic male sterility (cms) in *B. campestris* induced by *Raphanus* cytoplasm (R1) was used to facilitate the incorporation of multiple disease resistance (mdr) in cms *B. campestris* through cyclic backcross and screening. Resistance to *Plasmiodiophora brassicae* race 6, *Peronospora parasitica* and turnip mosaic virus (TuMV) was incorporated into cms *B. campestris* ssp. *pekinensis* and *B. campestris* ssp. *chinensis* lines. Clubroot resistance is conditioned by independent dominant *Pb1* and *Pb3* genes. Hypersensitive resistance to TuMV C1 strain is dominantly inherited. Downy mildew resistance, expressed in cotyledons as a reduction in the sporulation capacity of *P. parasitica*, is conditioned by a dominant gene *Pp*. A mdr screening method applicable to other *Brassica* species was developed. The efficiency of disease resistance incorporation was improved by using stored pollen of recurrent parents. *B. campestris* pollen stored dry at -22 C remained highly viable for over one year.

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SEPTORIA LEAF SPOT RESISTANCE IN LYCOPERSICON SPECIES. R. L. Clark and C. C. Block, Regional Plant Introduction Station, USDA, S&E-ARS and Iowa State University, Ames, Iowa 50011

The PI tomato collection (4700 lines) was screened in the greenhouse, 5 plants/12.7 cm pot, 2 pots/entry. Plants were inoculated at 6 weeks by immersing the tops in a spore suspension (1.5×10^6 /ml) prepared by soaking infected leaves for 20 min. in water. Incubation was 48 hr at 100% r.h., 22 C. Disease ratings (1 to 5 scale) were made 2 weeks after inoculation. One = no infection; 2 = only slight lesion development (<0.5 mm), no sporulation; 3 = small lesions (1-1.5 mm), little or no sporulation; 4 = slightly reduced lesions (2-2.5 mm), reduced sporulation; 5 = large (3-5 mm), abundant lesions, abundant sporulation. *L. esculentum* lines ranged from 3 to 5, most being 5; *L. cheesmanii* from 3 to 5; *L. pimpinellifolium* from 2 to 5; *L. hirsutum* from 3 to 4; *L. parviflorum* and *L. peruvianum* from 2 to 3. A complete list of PI lines tested is available.

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THE EFFECT OF INOCULUM CONCENTRATION ON THE APPARENT RESISTANCE OF WATERMELONS TO FUSARIUM OXYSPORUM F. SP. NIVEUM. R.D. Martyn and R.J. McLaughlin, Department of Plant Sciences, Texas A&M University, College Station, TX. 77843

Seventeen watermelon cvs. were rated for resistance to *F. oxysporum* f. sp. *niveum* by seedling root-dips at four concentration levels: 10^3 , 10^4 , 10^5 , and 10^6 spores/ml. Plants were rated for wilt 2 and 3 wks post-inoculation. Four categories were established: susceptible (>80% wilt), slightly resistant (50-80% wilt), moderately resistant (21-50% wilt), and highly resistant ($\leq 20\%$ wilt). At low inoculum levels (10^3), eight cvs. were highly resistant, four moderately resistant, four slightly resistant, and only one was susceptible. As the inoculum level increased, significant shifts occurred in the ratings. At 10^4 spores/ml there was a 50% reduction in the number of highly resistant cvs. (8-4), and a large increase in the number of susceptibles (1-7). At 10^6 spores/ml only two cvs. (Smokelee and Dixielee) were rated as highly resistant while 12 were susceptible. The trend of decreasing resistance with increasing inoculum pressure was the same for both 2 wk and 3 wk results.

562. Withdrawn.

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THREE CONJUGATIVE PLASMIDS ISOLATED FROM PSEUDOMONAS TABACI AND P. ANGULATA DO NOT ENCODE TABTOXIN PRODUCTION OR PATHOGENICITY. Mark Obukowicz and Paul D. Shaw, Dept. of Plant Pathology, University of Illinois, Urbana, IL 61801

Three cryptic plasmids isolated from strains of *Pseudomonas tabaci* 11528 (pJP1), *P. tabaci* BR2 (pBPW1), or *P. angulata* 45 (pJP30) were labeled with TnA using RSF1010::TnA as a vector. pBPW1 was labeled in four different sites, pJP1 in two sites, and pJP30 in one site. No significant symptom differences were observed in tobacco or green bean plant leaves injected with suspensions of wild-type strains or strains cured of resident plasmids. In addition, *P. tabaci* strains 11528 and BR2 still produced approximately the same amount of tabtoxin after loss of the indigenous plasmid. Thus, the three plasmids do not apparently encode genes for tabtoxin production or pathogenicity. All three plasmids, however, are conjugative.

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MECHANISM OF COMOBILIZATION OF RSF1010 BY A CONJUGATIVE PLASMID FOUND IN THE PLANT PATHOGENIC BACTERIUM PSEUDOMONAS TABACI. Mark Obukowicz and Paul D. Shaw, Dept. of Plant Pathology, University of Illinois, Urbana, IL 61801

pBPW1, a conjugative plasmid isolated from the plant pathogenic bacterium *Pseudomonas tabaci* BR2, mobilizes itself and RSF1010 at high frequencies into *Pseudomonas mellea* recipients. Mobilization of both plasmids are separate events in that pBPW1::TnA and RSF1010 are found singly in nearly half of the transconjugants. No detectable homology by Southern blotting was found between pBPW1 and RSF1010 in donors prior to conjugation or in recipients containing both plasmids after conjugation. These data indicate that pBPW1 and RSF1010 do not recombine during mobilization. Rather, pBPW1 donates RSF1010 by a yet unknown mechanism.

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GENETIC AND PHYSICAL CHARACTERIZATION OF TRANSPOSON Tn5 INDUCED MUTANTS IN PSEUDOMONAS SOLANACEARUM. B. Staskiewicz, D. Dahlbeck, J. Miller and D. Damm. IPRI, San Carlos, California 94070.

We have used the wide host range suicide plasmid pJB4JI to introduce the transposon Tn5 into the genome of *P. solanacearum*. The *E. coli* strain HB101(pJB4JI) was mated overnight with a race 3 strain of *P. solanacearum* Ps82 Rif^R, and exconjugants were selected on a complex medium containing Km (25 ug/ml) and Rif (100 ug/ml). The frequency of mutations to auxotrophy was .5%. In addition, non-fluidal avirulent mutants were detected by streaking the Km^R exconjugants on a complex medium containing .005% TZC. Southern blotting of total *P. solanacearum* DNA cleaved with the restriction enzyme EcoRI revealed that the majority of the exconjugants examined contained single Tn5 insertions at unique sites in the genome. Physical analysis of some of the non-fluidal mutants has been correlated with a 3.1 kb hybridizable sequence to a Tn5 probe. We are currently attempting to clone this sequence to determine its possible role in the virulence of *P. solanacearum*.

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CLONING AND EXPRESSION OF ICE NUCLEATION GENES FROM PSEUDOMONAS SYRINGAE AND ERWINIA HERBICOLA IN ESCHERICHIA COLI. C. S. Orser, J. B. Staskiewicz*, N. J. Panopoulos, D. Dahlbeck*, and S. E. Lindow. Dept. Plant Pathology, University of California, Berkeley, CA 94720. *International Plant Research Institute, San Carlos, CA 94070.

Genomic libraries of two *Pseudomonas syringae* and one *Erwinia herbicola* ice nucleation active (INA⁺) strains were constructed by *in vitro* packaging of partially digested DNA fragments ligated to the high capacity vector pLAFR and transducing *E. coli* HB101. The libraries were subsequently screened for expression of ice nucleation activity. Several clones from each library that expressed INA⁺ phenotype in *E. coli* and complemented INA⁻ mutants of the source strains were identified and further characterized. In addition to exhibiting the same threshold temperature of ice nucleation as the DNA source strains when grown at 22C, the INA⁺ *E. coli* expressed ice nucleation activity at temperatures up to 42C. Subcloning of the active DNA fragment into the multicopy vector pBR325 led to an increase in ice nucleation activity in *E. coli* HB101.

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PSEUDOMONAS PLASMID HOMOLOGY: A POSSIBLE CLUE TO PLASMID EVOLUTION. R. Quant and D. Mills, Genetics Program and Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, Oregon 97331.

Plasmid DNA from 5 strains of *Pseudomonas syringae* pv. *phaseolicola* (PSP) and one strain of *P. syringae* pv. *glycinicola* (PSG) were analyzed for homology with respect to pMC7105, a 98 megadalton (Mdal) integrative plasmid of PSP. Plasmid homology was considered basic to discerning evolutionary relationships among plasmids. The strains were obtained from diverse geographic areas and each contained 1 or 2 plasmids which ranged in molecular weight from 4.5 to 109 Mdal. BamHI and EcoRI fingerprints of total plasmid DNA from the PSP strains revealed a high level of sequence homology with pMC7105. Southern blot analyses of the BamHI digests identified only 4 fragments which failed to hybridize with pMC7105. The 4 fragments (25 Mdal total) originated from 1 PSP strain containing 2 plasmids. These fragments did not hybridize to PSP chromosomal DNA, but did hybridize with plasmid DNA from the PSG strain. A genetic model explaining the evolution of these plasmids is being developed.

CHARACTERIZATION OF AN INTEGRATIVE PLASMID, pMC7105, FROM *PSUEDOMONAS SYRINGAE* PV. *PHASEOLICOLA*. L. J. Szabo & D. Mills. Dept. of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331

The phytopathogenic bacterium *Pseudomonas syringae* pv. *phaseolicola* strain LR700 has been shown to contain a 147 kbp plasmid, pMC7105, which can replicate autonomously or integrate into the bacterial chromosome. Approximately 10 percent of randomly screened colonies derived from the integrated form harbor excision plasmids. These plasmids can be grouped into 5 classes on the basis of the sites at which excision recombination occurred. Three of these classes contain F'-like plasmids composed of pMC7105 and chromosomal DNA. The integration site on pMC7105 has been identified on a 3.1 kbp fragment and cloned. This fragment was subcloned from a 7.5 kbp fragment which shows homology with regions of pMC7105 and the plasmid-chromosomal junction fragments. A partial restriction map of pMC7105 has been constructed. The integrative property of this plasmid is being used to develop an Hfr system similar to that in *Escherichia coli*.

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LOCALIZATION OF *prd1* IN *PSEUDOMONAS SYRINGAE*. J. R. Vincent and D. W. Fulbright, Department of Botany and Plant Pathology, Michigan State University, East Lansing, MI 48824

Plasmid *prd1* contains the genes for histidine and the nitrogen fixation complex from *Klebsiella pneumoniae*, and genes for resistance to kanamycin, tetracycline, and ampicillin. When *prd1* was transferred to *P. syringae* PSSD2 *nalA-1 met-1 his-1*, the intact plasmid was maintained by selection for histidine prototrophy and kanamycin resistance. After four successive transfers on a medium that selects for these markers, *prd1* was no longer detectable. To determine whether or not *prd1* integrated into the host chromosome, DNA-DNA hybridizations were performed by the method of Southern using ³²P labeled cloned *nifKDH* genes as the probe. *prd1* may be used to identify *recA* mutants, due to its spontaneous degradation and loss of the *his* - *nif* portion of the plasmid in *recA* organisms. Mutants of *P. syringae* that may be recombination deficient were isolated by nitrosoguanidine mutagenesis and selection for increased sensitivity to methylmethanesulfonate and UV light.

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CHROMOSOME MOBILIZATION IN *ERWINIA STEWARTII* MEDIATED BY PLASMID pDC252.1 AND BACTERIOPHAGE *Mu*cts62. S. L. McCammon and D. L. Coplin, Dept. of Plant Pathology, Ohio Agricultural Res. & Devel. Center, and The Ohio State University, Wooster 44691.

Gene transfer in *Erwinia stewartii* strain SS104 was achieved using donor strains containing plasmid pDC252.1 and lysogenic for bacteriophage *Mu*cts62. pDC252.1 was derived from an indigenous plasmid in *E. stewartii* which was derepressed for transfer and has been labeled with transposons *Tn1* and *Tn10*. This plasmid transferred chromosomal markers poorly (2×10^{-9} to 4×10^{-7} recombinants/donor cell). However, when *Mu*cts62 replication was heat-induced in donor strains, all selected recipient markers were transferred between 1×10^{-6} and 5×10^{-5} /donor cell. These frequencies indicate multiple origins of transfer. Recombinants also acquired unselected donor markers. The following linkages were observed: *his-1* with *trp-2* and *ser-1*; *leu-1* with *lys-2* and *pro-5*; *arg-3* with *pro-5* and *ilv-1*; *arg-4* with *rif*; and *cys-2* with *ilv-1* and *ser-1*.

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A MATHEMATICAL FORMULA FOR PREDICTING PLASMID LOSS, R. V. Miller W. W. Esty*, and D. C. Sands, Dept. of Plant Pathology and *Dept. of Mathematics, Montana State Univ., Bozeman, MT 59717.

A bacterial cell can lose its plasmid components either as a spontaneous event or as a result of chemotherapy. If a plasmid is required for virulence as in the case of *Agrobacterium tumefaciens*, or *Pseudomonas savastanoi*, then plasmidless strains are of special value for biochemical analysis of virulence traits. We present a mathematical formula derived from the theory of stochastic processes for prediction of plasmid loss rates, given cell growth rates with and without the plasmid and the probability of plasmid loss in a single generation. In an *E. coli* *prk-2* plasmid system, plasmidless cells increased from 0% to 97% of the population in 700 generations. The curve that best fits this phenomenon involved a 2% greater growth rate of plasmidless cells, and a probability of loss at 0.0005 per cell generation. Expansion of this formula may be useful in predicting loss of multicopy extra chromosomal genetic elements such as virus infected cells.

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CROWN GALL SUPPRESSIVE GENES ARE LOCATED IN THE TRANSFER REGION OF INC-W PLASMID pSa. R. C. Lundquist, R. C. Tait and C. I. Kado. Department of Plant Pathology, University of California, Davis, California 95616

Our laboratory has shown previously that the IncW plasmid pSa completely suppresses virulence of *Agrobacterium tumefaciens* (J. Bact. 139:591, 1979; Mol. Gen. Genetics 181:44, 1981). We have now constructed a genetic map of pSa by using deletion derivatives and cloned fragments of pSa DNA. The genes that encode resistance to chloramphenicol, sulfonamides, spectinomycin, streptomycin, kanamycin, gentamycin and tobramycin are clustered in two adjacent plasmid regions. A 4 kilobase pair region necessary for replication in *A. tumefaciens* and *E. coli* has been mapped and two additional regions involved in plasmid transfer have been identified. One of the transfer regions is involved in the inhibition of oncogenesis by pSa when it is present in *A. tumefaciens*. (Supported by NIH Grant CA-11526)

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GENES ESSENTIAL FOR CROWN-GALL ONCOGENESIS ARE LOCATED OUTSIDE THE T-REGION OF THE *AGROBACTERIUM TUMEFACIENS* TI PLASMID. J. C. Kao and C. I. Kado. Department of Plant Pathology, University of California, Davis, California 95616

Virulent *A. tumefaciens* strains harbor large Ti plasmids that carry the genes responsible for crown gall oncogenesis. Our laboratory has shown previously that Ti plasmid genes encode host-range specificity, a function that is important in virulence maintenance (J. Bact. 139:591, 1979). We have now located the genes that are absolutely essential for crown gall oncogenesis. These *Onc* genes were located by insertional mutagenesis with transposon *Tn5*. Ti plasmid mutants were obtained by filter matings between *E. coli* 1830 (pPH1::Mu_{cts}::Tn5) and *A. tumefaciens* C58 (pTiTra^C) followed by double patch matings between the resulting transconjugants and plasmid-free *A. tumefaciens* recipients carrying selectable antibiotic resistance markers. Avirulent mutants screened on broad-strain-recognition host plants were collected. The site of insertion were found by restriction fragment displacement analysis of pTi::Tn5 DNA. *Onc* genes were found clustered ca. 17 kbp left of the T-region. (Supported by NIH Grant CA-11526)

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VARIABILITY IN VIRULENCE OF THE BACTERIUM CAUSING PIERCE'S DISEASE OF GRAPEVINE. D. L. Hopkins, Agricultural Research Center, University of Florida, Leesburg, FL 32748.

Isolates of the Pierce's disease (PD) bacterium obtained from bunch grape (*Vitis vinifera*), muscadine grape (*V. rotundifolia*), wild grapes (*V. munsoniana*), citrus (*Citrus jambhiri* and *C. sinensis*), American elder (*Sambucus canadensis*), and pepper vine (*Ampelopsis arborea*) were compared for virulence on various bunch and muscadine grape cultivars. When grouped by original source, the groups of isolates were very similar in average virulence. Members of all groups varied in virulence from avirulent to highly virulent. Twenty PD isolates, obtained from a single bunch grape plant and single-celled twice, had the same range in virulence as that of the groups above. There was no indication of host specificity, but the ability of an isolate to produce PD symptoms in a moderately resistant muscadine cultivar depended on the level of virulence of the individual isolate. The variability in virulence appears to result from either natural variation in the bacterial populations or from a loss of virulence in culturing.

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VARIATION AMONG STRAINS OF *CORYNEBACTERIUM SEPEDONICUM*. S.H. De Boer, Agriculture Canada Research Station, 6660 N.W. Marine Drive, Vancouver, B.C. V6T 1X2.

Nineteen strains of *Corynebacterium sepedonicum*, originating from several different locations in Canada and the United States, were compared. All except two of the strains produced symptoms in root-inoculated rooted potato (cv. Red Pontiac) stem cuttings and in stem-inoculated eggplant (cv. Black Beauty) seedlings. Nine of the strains produced mucoid and ten non-mucoid colonies on a yeast extract-glucose-mineral salts medium. Only some of the strains produced acid from arabinose, gluconate, glucose, mannitol, salicin, and xylose but all strains produced acid from dextrin, fructose, and raffinose, and no acid from twelve other carbohydrates. Most strains were sensitive to bacteriocins produced by seven different *C. michiganense* strains; three strains showed either no or only very small zones of inhibition on bacteriocin test plates. All strains were positive in immunofluorescence with antisera produced in rabbits against glutaraldehyde-fixed cells of one of the mucoid strains.

VARIABILITY IN THE VIRULENCE OF *ERWINIA AMYLOVORA* TO APPLE CULTIVARS. J. L. Norelli and H. S. Aldwinckle, Cornell University, N.Y.S. Agricultural Experiment Station, Geneva, N.Y. 14456

A single virulent strain of *Erwinia amylovora*, EA 273, has been used to screen apple seedlings and cultivars for resistance to fire blight. EA 266, which is pathogenically specialized for the apple cv Quinte, was more virulent than EA 273 on some New York apple breeding selections and some *Malus* species but not on several other apple cultivars. Following shoot tip inoculation with 10^{10} CFU/ml of EA 266 and EA 273, the % shoot lengths infected were McIntosh 72% and 82%, Golden Delicious 83% and 53%, Delicious 32% and 44%, *Malus atrosanguinea* 38% and 1%, Novole 24% and 0%, Robusta-5 9% and 0%, and NY 72700-141 4% and 0%, respectively. In 1981 when apple seedlings from controlled crosses were inoculated with a mixture of cells of EA 273, EA 266 and three other EA strains, the pattern of segregation for fire blight susceptibility changed markedly from that observed in previous years when the inoculum consisted of EA 273 alone.

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PROPERTIES OF AVIRULENT *ERWINIA STEWARTII* MUTANTS LACKING UDP-GALACTOSE-4-EPIMERASE. D. L. Coplin, C. Meaney, J. J. Bradshaw-Rouse and S. L. McCammon. Departments of Plant Pathology, Ohio Agricultural Res. and Devel. Center, The Ohio State University, Wooster 44691 and Univ. of Wisconsin, Madison 53706.

Mutants of *Erwinia stewartii* which cannot produce capsular polysaccharide are generally able to cause water-soaked lesions in corn seedlings but not systemic wilting. Two nonencapsulated mutants were isolated which produced only small lesions without water-soaking. These mutants could not utilize galactose and their growth was inhibited by galactose. Enzyme assays revealed that they lack UDP-Gal-4-epimerase (*galE*) a key enzyme in the production of nucleotide sugar precursors for cell wall biosynthesis. *Gal*⁺ recombinants, obtained by mating *galE* mutants with *Gal*⁺ donors, regained virulence. Experiments with *galK* (galactokinase) and *galE galK* mutants indicate that sensitivity to or inability to degrade galactose *in planta* is not responsible for the avirulence of *galE* mutants. Instead, the avirulence of these mutants may be due to production of galactose-deficient cell wall lipopolysaccharides.

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FIELD REACTION OF SELECTED BARLEYS TO *PUCCINIA GRAMINIS*. B. J. Steffenson, R. D. Wilcoxson and A. P. Roelfs. Department of Plant Pathology, University of Minnesota and Cereal Rust Laboratory, ARS, USDA, St. Paul, MN 55108.

Barley cultivars were evaluated for stem rust resistance in separate nurseries of *Puccinia graminis* f. sp. *tritici* (Pgt-a composite of races 113-RTQ, 151-QFB, 151-QSH, 29-HJC and 15-TNM) and *P. graminis* f. sp. *secalis* (Pgs-one isolate) by infection response and terminal rust severity (TS). In the Pgt nursery, cultivars with the T-gene differed from those without it. Chevron was R-MR with TS of 1%; other T-gene cultivars were MR-MS with TS up to 18% suggesting that other genes influence resistance conferred by the T-gene. All cultivars without the T-gene were S-MS with TS of 35-80%. In the Pgs nursery, T-gene barleys were S-MS and TS varied from 35-70%. Among cultivars without the T-gene, Hiproly and Steptoe were S with TS of 60-70% while Valkie, Hispont and Heitpas-5 were S-MR with TS of 18-25%. Black Hulless (R-MS and TS of 3%) was most resistant to Pgs. Thus genetic factors other than the T-gene confer resistance to this isolate of Pgs.

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INHERITANCE OF SLOW LEAF-RUSTING RESISTANCE IN WHEAT. T. S. Lee and G. Shaner, Department of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907.

The inheritance of slow leaf-rusting resistance of the wheat cultivars SW72469-6 and L574-1 was studied in crosses with the susceptible cultivars Morocco and Suwon 92. Parental, F₁, F₂, F₃, and BC generations were grown in the greenhouse and inoculated with *Puccinia recondita* at the boot stage. The latent period was measured. Genotypic hypotheses based on F₁ and F₂ data were tested by evaluation of F₃ families and BC F₁ progeny. Evaluation of the crosses SW72469-6/Morocco and L574-1/Suwon 92 indicated that short latent period was inherited as a partially dominant trait. In each cross the genetic data could be explained by a model that assumes two genes with equal effect control latent period. This study shows that long latent period, which is often regarded as an example of general resistance, can be controlled oligogenically rather than polygenically.

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PROBABLE GENOTYPE OF *TRITICUM AESTIVUM* 'NEWTON' FOR REACTION TO *PUCCINIA RECONDITA*. L. E. Browder and Rashied Modawi. USDA, ARS, Dept. of Plant Pathology and Dept. of Agronomy, Kansas State University, Manhattan, Kansas 66506.

Three kinds of hypothesis about reaction genotype of a wheat cultivar may be developed from infection-type (IT) data by using gene-for-gene theory. These are: 1) that it does not have a gene known to occur in another cultivar, 2) that it has a gene different from a gene in another cultivar, and 3) that it probably does have a gene known to occur in another cultivar. These hypotheses can be tested through conventional genetic studies. The *P. recondita* reaction genotype of Newton wheat was studied using these methods. We hypothesized that Newton has *Lr1*, that some plants of the cultivar have *Lr3*, and that it has two specific genes which condition an X IT. One of these X IT-conditioning genes may be *Lr11*. A cross of Newton with a line having *Lr1* showed no segregation to a *Lr1 P. recondita* culture, indicating the first hypothesis to be correct. Newton does not have *Lr2*, *Lr9*, *Lr10*, *Lr16*, *Lr17*, *Lr19*, *Lr24*, *Lr25*, or *Lr26*.

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THE RESIDUAL AND INTERACTIVE EXPRESSIONS OF "DEFEATED" STEM RUST OF WHEAT RESISTANCE GENES. R. R. Nelson, Uzi Brodny, and L. V. Gregory, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Eight wheat lines developed by Loegering in a Chinese Spring background and representing all combinations of stem rust genes *Sr6*, *Sr8* and *Sr9a* from Red Egyptian were studied for their reaction to an isolate of race RKKQ possessing virulence genes for *Sr6*, *Sr8*, and *Sr9a*. With respect to reduced pustule size and reduced sporulation, the following results were obtained: the seven lines with one or more dominant rust genes were significantly more effective than the line with no dominant rust genes; lines with two dominant genes were more effective than lines with single dominant genes; and, the line with three dominant genes was more effective than lines with two dominant genes. We conclude that the three rust genes each has a residual expression when confronted by matching virulence genes and that the genes interact to provide a greater residual resistance.

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MANAGING GENETIC RESOURCES OF WHEAT. B. S. Gill and W. J. Raupp, Department of Plant Pathology, Kansas State University, Manhattan, Kansas 66506.

We have studied the University of California-Riverside collection of wild relatives of wheat for the past two years, and will discuss our experience with this unique resource. Maintenance and seed increase operations consume the most time, are the most labor-intensive, and require extensive greenhouse space. Evaluation of disease and insect resistance and other desirable traits is an interdisciplinary effort in which seedling screening is emphasized. New sources of genetic resistance to leaf rust, wheat streak mosaic virus, powdery mildew, greenbug and Hessian fly have been identified. Chromosomal location of new genes is being determined by cytogenetic analyses. Genetic stocks are being developed for rapid and efficient transfer of the new disease and insect resistance genes into winter wheat. Improved germplasm will be released for wheat breeding.

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ASSOCIATION BETWEEN CRUDE FIBER, CELLULOSE, AND LIGNIN IN STEMS OF WINTER WHEAT AND RESISTANCE TO *PUCCINIA HERPOTRICHOIDES*. T. D. Murray and G. W. Bruehl, Dept. of Plant Pathology, Washington State University, Pullman, WA 99164-6430.

In a previous study, resistance to *P. herpotrichoides* in winter wheat was correlated with a wide, heavily lignified hypodermal layer. Four winter wheats, varying from resistant to susceptible to *P. herpotrichoides*, were sampled at 2-week intervals from jointing to maturity. Length, diam., and weight of 200 first internodes of each wheat were measured and then analyzed for fiber, cellulose, and lignin content. When compared on a dry weight basis there was no correlation between fiber, cellulose, and lignin content and width of hypodermis. However, when values of fiber, cellulose and lignin were adjusted for differences between wheats in internode diameter and weight per unit length of internode, there was a significant correlation between lignin content and hypodermis width ($r = 0.85$). There was no significant correlation between fiber and cellulose content and hypodermis width ($r = 0.72$ and $r = 0.69$, respectively). These results agree with previous observations and indicate that lignification is involved in resistance to *P. herpotrichoides*.

RESISTANCE TO POWDERY MILDEW CONDITIONED BY THE ml-o ALLELE IN BARLEY: POSSIBLE ROLE OF PAPILLAE. M.C. Stolzenburg, J.R. Aist, and H.W. Israel, Department of Plant Pathology, Cornell University, Ithaca, NY 14853

Powdery mildew resistance in barley conditioned by the ml-o allele is expressed during primary penetration and is associated with large papillae. Low speed centrifugation, which stratifies contents of living epidermal cells of detached coleoptiles, was used to test the disease resistance role of papillae in near-isogenic barley lines differing at the ml-o locus. Inoculated coleoptiles were centrifuged during attempted fungal penetration such that most penetration attempts occurred on cytoplasm-poor (CP) zones of cells, where papilla formation is inhibited. Penetration efficiency (PE) on CP zones of cells of the susceptible isolate (ml-o) did not differ significantly from that of uncentrifuged cells. In contrast, PE on CP zones of the resistant isolate (ml-o) showed a four-fold increase from that on uncentrifuged controls. These preliminary results suggest that cytoplasmic response, such as papilla formation, may be implicated in ml-o-mediated resistance.

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CHARACTERIZATION OF RESISTANCE IN PEANUT TO CYLINDROCLADIUM BLACK ROT. J. K. Pataky, M. C. Black and M. K. Beute. Dept. Plant Pathology, NC State University, Raleigh, NC 27650

Resistance to *Cylindrocladium* black rot (CBR) was characterized by inoculum density (ID)-root rot index (RRI) relationships, disease development and *C. rotalariae* microsclerotia (ms) production. In greenhouse tests, positions of log₁₀ID-RRI regression lines were significantly different for peanut genotypes. Predicted RRI (range 0-5) at 1 ms/g soil was 1.4, 1.6, 2.0, 2.7 and 3.0 for NC 18016, NC 3033, NC 18229, NC 8C and Florigiant, respectively. In field evaluations, disease development in plots of Florigiant and NC 8C was more rapid than in plots of NC 18229 and NC 18016. Production of ms in roots of field grown plants was analyzed by regression of log₁₀ms (per g root) on RRI. Generally, more ms were produced on resistant than on susceptible lines at similar RRI. Therefore, at high ID when all lines are severely diseased, ms production may be greater on resistant lines; however, at low ID where resistant lines sustain less disease than susceptible lines, ms may be more abundant on susceptible lines.

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EFFECTS OF ULTRAVIOLET LIGHT (UV) ON ETHYLENE MALEIC ANHYDRIDE (EMA) INDUCED RESISTANCE TO TOBACCO MOSAIC VIRUS (TMV). M. Chessin and A. Mitra, Department of Botany, University of Montana, Missoula, MT 59812.

Attached leaves of *Nicotiana tabacum* Var. Samsun NN and Burley 21 were injected with EMA solution (500 mg ml⁻¹) and then irradiated with UV (85% 253.7nm) 24 hours after injection. Irradiated plants were kept in dark overnight to prevent photo-reactivation before inoculating with TMV (18.64 mg ml⁻¹). All UV doses enhanced EMA induced resistance with respect to lesion number. Effects on lesion size were dose dependent. From 125 ergs mm⁻² to 1000 ergs mm⁻² UV also enhanced EMA induced resistance with respect to lesion size but partially reversed resistance at doses from 1500 ergs mm⁻² to 5250 ergs mm⁻². Results at higher UV dose levels agree with hypothesis that EMA induced resistance resembles that produced by previous virus infection in that cellular transcription is involved. Enhanced localization at lower UV doses may be related to that reported for UV-visible light interaction by Wu and Dimitman (Virology 40:820). (Supported by BARD grant no. I-133-79).

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CULTIVAR BY ISOLATE INTERACTION BETWEEN SUGARBEET AND ERWINIA CAROTOVORA BETAVASCULORUM. E. D. Whitney, Agricultural Research Station, USDA, ARS-WR, P. O. Box 5098, Salinas, CA 93915.

Both quantitative and qualitative host resistance to *E. carotovora betavascularum* the incitant of bacterial vascular necrosis and rot of sugarbeet has been shown. Early studies of isolates inoculated onto United States cultivars suggested that the cultivars tested reacted similarly to the pathogen but that aggressiveness differed among the isolates. However, when European cultivars were included in tests with nine isolates of the pathogen, a cultivar by isolate interaction was observed. Thus, biotypes or races of the pathogen were differentiated by the host reaction. This differential reaction between United States and European cultivars suggests additional factors for resistance and divergence between European and United States cultivars.

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GREENHOUSE AND FIELD COMPARISON OF RESISTANCE IN ALFALFAS TO COLLETOTRICHUM TRIFOLII. S. J. Allen, J. L. Caddel, and G. L. Barnes. Departments of Plant Pathology and Agronomy, Oklahoma State University, Stillwater 74078.

The value of resistance to *Colletotrichum trifolii* in alfalfa has been documented (Elgin et al, Crop Sci. 21: 457-460). Cultivars highly resistant to *C. trifolii* are available, but few are well adapted to Oklahoma. Several alfalfas were tested for resistance to *C. trifolii* under greenhouse and field conditions. Resistance in the greenhouse was measured in percent survival after inoculation. Field measurements included stems per unit area, yield, and number of shepherds crooks (caused by *C. trifolii*). Cultivars selected for resistance to *C. trifolii* were the only alfalfas which showed significant resistance in the greenhouse. Resistant cultivars did not show a production advantage in the field. Greenhouse survival data and number of shepherds crooks appeared related. Further analyses of data on a plot-by-plot basis should show if symptoms were concentrated, and if differences occur within these areas.

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IN VITRO SELECTION OF POTENTIAL DISEASE RESISTANT SOMACLONAL VARIANTS OF LETTUCE FROM REGENERATED PROTOPLASTS. Dean E. Engler and R. G. Grogan, Dept. of Plant Pathology, University of California, Davis, CA 95616

We have previously reported the development of an efficient method for regeneration of whole lettuce plants from isolated leaf mesophyll protoplasts. This technique has been modified by the imposition of selection pressures in an attempt to screen large populations of protoplasts for resistance to some important factors in the production of lettuce diseases. Oxalic acid is strongly implicated as a requisite toxin for pathogenesis by *Sclerotinia* sp. We have regenerated lettuce plants from protoplasts cultured on media with sufficient oxalic acid to kill 99.98% of exposed cells. Similar degrees of selection pressure were imposed on other protoplasts for tolerance to very low levels of calcium in the regeneration medium. Regenerated plants will be tested for resistance to tipburn (a disease induced when the calcium in interior head tissues drops below a critical level) and lettuce drop in the hope that the resistance factors expressed by protoplasts also will be expressed in whole plants.

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INFLUENCE OF DISEASE RESISTANCE, FUNGICIDES, AND PATHOGEN INOCULUM DENSITY ON PHYTOPHTHORA CAPSICI CROWN ROT OF PEPPER. S. Tüzün & R. S. Ferriss, Department of Plant Pathology, University of Kentucky, Lexington, 40546.

One-month-old plants of 6 pepper cultivars grown in steamed soil in 165 ml plastic tubes in the greenhouse were drenched with either metalaxyl at 0.1 or 1.0 mg per tube, triphenyl-tin-OH at 0.29 or 2.9 mg per tube, or deionized water. One week later plants were inoculated with *P. capsici* at either 0, 10³, 10⁵, or 10⁷ zoospores per plant. Time until plant death (TPD) was recorded for each of 7 plants per treatment. For all cultivar-fungicide combinations where death occurred, mean TPD decreased with increasing inoculum density. No significant plant death was observed during the course of experiment (35 days after inoculation) in all treatments receiving the high rate of either fungicide, and 10³ or 10⁵ zoospores per plant. In general, the relative ranking of the cultivars was: most resistant = Phyto 636 (selected from a cross of PM217 and Yolo Wonder) > PM217 > PM662 ≈ Maras ≈ California Wonder ≈ Yolo Wonder. Fungicide treatment had little effect on the relative ranking of the cultivars.

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SCREENING PEAS (PISUM SATIVUM) FOR RESISTANCE TO RHIZOCTONIA SOLANI STEM ROT. Randy J. McCoy and John M. Kraft, USDA-ARS-WR P. O. Box 30, Prosser, WA 99350

Two inoculation techniques for screening seedling peas for resistance to stem rot caused by *Rhizoctonia solani* (AG-4) were compared. The first technique consisted of soil infestation with either sclerotia (0.250-0.425 mm) or mycelial (cornmeal-sand) inoculum. Stem inoculation with four mm agar plugs of *R. solani* grown on either PDA, V-8 juice agar, or a liquid dextrose-asparagine medium comprised the second technique. Twenty differential pea lines consisting of 6 cultivars, 4 breeding lines, and 10 plant introduction nos. were screened with each source of inoculum. Growing peas in infested soil resulted in significantly higher disease indices than the stem inoculations with PDA or V-8 cultures. However, inoculum plugs from the dextrose-asparagine medium caused as high disease indices as infested soil. Stem rot resistance was not related to seed coat color or seedling emergence rate. A highly significant correlation was found between thickness of seedling epicotyls and resistance.

GENETICS OF RESISTANCE TO *UROMYCES PHASEOLI* IN *PHASEOLUS VULGARIS* BREEDING LINE B-190. J. R. Stavelly, USDA, ARS, Applied Plant Pathology Laboratory, Beltsville, MD 20705.

A Puerto Rican line, B-190 is resistant (R), with pustules less than 300 μm in diam., to many pathogenically unique pathotypes of rust, but susceptible (S) to two rust cultures, 15B and 15G, that were selected in the greenhouse at Beltsville. Both pinto cv. Olathe and snap cv. GG447 are R or highly resistant (HR), with necrotic spots without sporulation, to 15B and 15G. Each of 160 F_2 plants of GG447 x B-190 were separately and simultaneously inoculated with each of eight pathotypes. They segregated in a ratio of 3R:1S for each pathotype. Reactions of 110 F_2 plants of B-190 x Olathe and 90 F_2 plants of the reciprocal Olathe x B-190 fit a 3R:1S ratio for those pathotypes to which Olathe is susceptible. For pathotypes to which Olathe is HR and B-190 is R, these same plants segregated 12HR:3R:1S, indicating that the Olathe factors for HR are independent from and epistatic to the B-190 factors for R. Our data suggests that B-190 contains a linked set of monogenic factors for resistance.

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CORRELATION OF SPORE PRODUCTION BY *ALBUGO CANDIDA* AND A VISUAL WHITE RUST RATING SCALE. David T. Fox, Department of Plant Pathology, University of Wisconsin, Madison, WI 53703.

Correlations of spore production by *Albugo candida* with a visual white rust rating scale were made, in order to elucidate the epidemiological significance of the scale. Each cotyledon of four-day-old *Brassica campestris* stock PHW-Aaa-1 was inoculated with two 10 μl drops of a 10^5 zoospore/ml suspension of *A. candida* race 2. After 16 hours at 100% R.H. and 20°C, followed by 7 days at 24°C and 250 $\mu\text{E m}^{-2} \text{ s}^{-1}$ continual illumination, plants were visually evaluated as 1, 3, 5, 7, or 8 according to increased pustule size, number, and distribution on the lower cotyledon surface. Twenty cotyledons from each rating class were selected, weighed, and washed in 0.1% Tween 80. White rust pustules were ruptured during washing. The number of sporangia in eight samples of washing solution from each rating class were counted. Spore production was highly correlated with numerical rating based on visual assessment of disease ($r > .95$). Means of spore production on plants rated as 1, 3, 5, and 7 were significantly different from each other.

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EVIDENCE THAT ASTER YELLOWS IS ASSOCIATED WITH NONHELICAL MYCOPLASMA-LIKE ORGANISMS RATHER THAN SPIROPLASMAS. J. W. Kloepper and D. G. Garrott, Department of Plant Pathology, University of California, Berkeley, CA 94720

Spiroplasmas were isolated from 33 of 39 samples (89%) from 9 *Plantago* major plants which lacked typical aster yellows (AY) symptoms and which were infected with a spiroplasma originally derived from *Plantago* with Tulalake AY. All isolations were attempted with a double centrifugation procedure to minimize effects of spiroplasma inhibitors in plant tissues. During a 20 month study, no spiroplasmas were isolated from 103 samples of 33 greenhouse-maintained *Plantago* and lettuce plants with Tulalake, dwarf, and severe AY. Electron microscopy revealed helical organisms in phloem of plants from which spiroplasmas were isolated; however, only nonhelical MLOs were visible in plants with typical AY. These results support the conclusion that the MLO associated with AY is noncultivable, nonhelical and not a spiroplasma.

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INFECTION BY MYCOPLASMA-LIKE ORGANISMS IMPORTANT IN ASH DECLINE IN NEW YORK. J. A. Matteoni and W. A. Sinclair, Department of Plant Pathology, Cornell University, Ithaca, NY 14853-0331.

Decline of white ash (*Fraxinus americana*) in New York State in recent decades was ascribed to multiple causes. Mycoplasma-like organisms (MLO, associated with witches'-broom) were first unknown and later judged unimportant. We used graft inoculations, Dienes' stain, diffusive resistance measurements, and observations in ash stands to learn the association of MLO with decline. Newly recognized symptoms were deliquescent branching, early and prolonged growth but at subnormal rate, subnormal cold hardiness, and high death rate. On 5 field plots where dieback or death averaged 44% incidence, MLO were detected in 56% of living trees including 26% of those appearing healthy, 60% of slow growing ones and 83% of those with dieback. Twig growth of infected seedlings in the field 2 years after inoculation was 25% that of noninfected seedlings ($P < .01$). Dieback occurred in 62% of infected and 17% of noninfected seedlings ($P = .09$).

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ASSOCIATION OF SPIROPLASMAS WITH A NEW DISEASE OF CORN. J. W. Kloepper, D. G. Garrott, and B. C. Kirkpatrick. Plant Pathology Department, University of California, Berkeley, CA 94720.

In 1981 a previously unreported disease was epidemic in late planted corn in Kings County Calif. The most consistent symptom was bright red leaf blades at harvest time. Most plants developed to mature height and did not exhibit tillering, axial branch development, or chlorotic leaf bands. Spiroplasmas were isolated from 80 of 83 (96%) individually sampled plants from diseased fields, but no spiroplasmas were isolated from 40 plants in disease free fields. *Dalbulus maidis* collected from diseased fields also yielded spiroplasmas. Electron microscopy of diseased plants revealed numerous, wall-less helical organisms in phloem cells. Twelve characterized isolates appeared similar to each other but distinct from reference strains of corn stunt spiroplasma, *Spiroplasma citri*, and honeybee spiroplasma based upon spiroplasma deformation tests, electrophoresis of SDS-solubilized proteins, growth temperature maxima, and production of exogenous DNAs. These results indicate that a newly recognized spiroplasma is associated with this corn disease.

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SERUM-FREE MEDIUM FOR IN VITRO CULTIVATION OF PHYTOPATHOGENIC AND EPIPHYTIC SPIROPLASMAS.

L.-M. Lee and R. E. Davis. Department of Botany, University of Maryland, College Park, MD 20742; and Plant Virology Laboratory, PPI, ARS, U.S. Department of Agriculture, Beltsville, MD 20705.

Bovine serum albumin (BSA), palmitic acid, and cholesterol have been used together to substitute for sera in media for determination of cholesterol requirements of animal mycoplasmas and of plant and insect spiroplasmas. Such media permit only relatively poor growth of phytopathogenic spiroplasmas (*Spiroplasma citri* and corn stunt spiroplasma). However, these spiroplasmas, as well as insect pathogenic and epiphytic spiroplasmas (i.e., honey bee spiroplasma AS 576 and flower spiroplasmas SR3, *brevi* and *S. floricola*), grow vigorously in a new serum-free medium which contains medium LD8A base, BSA, fatty acids, phospholipids, and cholesterol. Maximum titers and growth rates of the spiroplasmas in the serum-free medium were comparable to those in medium LD8A containing serum. The new serum-free medium facilitates determination of cholesterol and other lipid requirements of spiroplasmas.

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USE OF CHICKEN EGG YOLK AND RADIOIMMUNOSORBENT (RISA) ASSAY IN THE SEROLOGY OF SPIROPLASMAS. B. C. Raju, E. Khalil, A. Rowhani and G. Nyland, Department of Plant Pathology, University of California, Davis, CA 95616

White chickens were intravenously and intramuscularly injected with antigens prepared from pure cultures of *Spiroplasma citri*, corn stunt spiroplasma, honeybee spiroplasma AS-576, and spiroplasma isolates BEE-8 and SR-3. Eggs were collected one week after the last injection. Yolk was separated and used in a modified ELISA. Purified gamma globulin from rabbit anti-serum was used for coating and chicken yolk was used for conjugation. This system was 10-20 times more sensitive than regular ELISA for the detection of spiroplasma in diseased tissues and in distinguishing various spiroplasmas by serogroups. RISA was highly sensitive and specific in detecting spiroplasmas in host tissue. No cross reactions were observed among the strains representing various serogroups. RISA was especially useful in separating subgroups in a serogroup.

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PATHOGENICITY PROVED FOR ISOLATES OF SPIROPLASMA CITRI FROM SIX HOST SPECIES. R.M. Allen and C.R. Donndelinger, Department of Plant Pathology, University of Arizona, Tucson, AZ 85721

Pathogenicity has been proved for isolates of *Spiroplasma citri* from six host species including navel orange, celery, London rocket, common foxglove, zucchini squash, and watermelon. Each isolate, suspended in 5.0 per cent sucrose, was microinjected into lots of 25-70 healthy adult beet leafhoppers (*Circulifer tenellus*). Each lot was then caged on a 8-12 cm Madagascar periwinkle seedling for inoculation periods ranging from 15-36 days. Typical stunting and foliar discolorations associated with infections by spiroplasmas were first observed 24-159 days later. Controls receiving non-injected leafhoppers remained symptomless. Spiroplasmas were recovered from diseased plants only within 6-17 days after culturing. The orange isolate, AZ-103, deposited as ATCC 33723, retained pathogenicity through 170 sub-transfers in artificial media.

Spiroplasma citri in Maryland: Isolation from field-grown plants of horseradish (*Armoracia rusticana*) Robert E. Davis and Jacqueline Fletcher. Plant Virology Laboratory, PPI, USDA, Beltsville, MD 20705; and Department of Plant Pathology, University of Illinois, Urbana 61801.

Horseradish plants exhibiting a darkened phloem region in roots and other symptoms suggestive of brittle root disease were found growing in commercial fields in Maryland during 1981. Spiroplasmas were isolated in pure culture *in vitro* from surface sterilized samples of roots from such plants. Results from serological tests of growth inhibition, enzyme-linked immunosorbent assay, and polyacrylamide gel electrophoretic analyses of cellular proteins indicated triply cloned isolates of these spiroplasmas to be strains of *Spiroplasma citri*. The findings establish the occurrence of brittle root disease and *S. citri* in plants in the field on the eastern seaboard and raise questions about the potential for spread of *S. citri* to other crops and weed hosts in eastern U.S.

SPIROPLASMA CITRI INDUCES BRITTLE ROOT IN HORSEADISH WITH OR WITHOUT TURNIP MOSAIC VIRUS. Jacqueline Fletcher, Catherine Eastman, Gerald Schultz, and Michael McGuire. Dept. Plant Pathology, Univ. of Ill., and Illinois Natural History Survey, Urbana, IL 61801.

Horseradish (*Armoracia rusticana*), a perennial brassicaceous plant grown in Illinois commercially as an annual through vegetative propagation, is commonly infected with one or more mosaic viruses, predominantly turnip mosaic (TuMV). Brittle root disease, caused by infection with *Spiroplasma citri*, may also affect horseradish already infected with TuMV. Studies were initiated to determine if the presence of TuMV affected the development of brittle root disease. Horseradish seedlings, naturally free of TuMV or inoculated with TuMV, were found to be as susceptible to *S. citri* as were virus-infected horseradish plants grown from secondary roots. Tissue-cultured horseradish plants with and without TuMV were also equally susceptible to infection by *S. citri*. Prior infection with TuMV does not appear to be necessary for or to prevent subsequent establishment of brittle root disease.

PRESENCE IN HORSEADISH OF A VIRESCENCE AGENT DISTINCT FROM THE CAUSAL AGENT OF BRITTLE ROOT. C. Eastman, G. Schultz, J. Fletcher, and M. McGuire. Sect. of Econ. Ent., Ill. Nat. Hist. Surv. and Dept. Plant Path., Univ. of Ill., Champaign, IL 61820.

In tests of transmission of *Spiroplasma citri*, the causal agent of brittle root (BR), by *Circulifer tenellus* from field-collected horseradish, virecence and phyllody developed in periwinkle, indicating the presence of a second agent in horseradish. In contrast, *S. citri*-infected periwinkle produced diminutive flowers of normal color. The virecence agent, which caused no visible symptoms in vegetative horseradish, was transmitted from horseradish both affected with or free of BR, while *S. citri* was obtained only from BR-diseased plants. Examination of a field of horseradish cv. 984, which unlike most Illinois horseradish flowers routinely, indicated that 4.4% of the flowering plants in 1981 had virecence. The virecence agent was transmitted experimentally from plants of this type to periwinkle, radish, and wild mustard. These results show the presence in horseradish of a virecence agent, which has no direct role in BR disease.

THE ASTER LEAFHOPPER, MACROSTELUS FASCIFRONS: A NEWLY DISCOVERED VECTOR OF SPIROPLASMA CITRI. K. O'Hayer, G. Schultz, C. Eastman, J. Fletcher, and R. Goodman. Ill. Nat. Hist. Surv. and Univ. of Ill., Champaign, IL 61820.

The causal role of *Spiroplasma citri* in brittle root disease of horseradish (*Armoracia rusticana*) has recently been discovered. We tested the ability of *Macrosteles fascifrons*, a common insect in Illinois horseradish fields, to transmit the spiroplasma. *M. fascifrons* injected with an isolate of *Spiroplasma citri* obtained from brittle root-diseased horseradish transmitted the spiroplasma to horseradish and aster (*Callistephus chinensis*). *M. fascifrons* fed on *S. citri*-infected plants transmitted the spiroplasma from aster to aster and horseradish, from yellow rocket (*Barbarea vulgaris*) to aster, and from turnip (*Brassica rapa*) to turnip. This is the first report of transmission of *S. citri* from diseased to healthy plants by *M. fascifrons*. This leafhopper may be important in the epidemiology of brittle root disease in Illinois. In addition, the wide host and geographical ranges of *M. fascifrons* enhance the potential significance of this insect as a vector of *S. citri*.

PROPERTIES OF TRANSMISSION OF A BRITTLE ROOT ISOLATE OF SPIROPLASMA CITRI BY THE LEAFHOPPER, CIRCULIFER TENELLUS. G. Schultz, M. McGuire, C. Eastman, K. O'Hayer, and J. Fletcher. Econ. Ent., Ill. Nat. Hist. Surv., and Dept. Plant Path., Univ. of Ill., Champaign, IL 61820.

Laboratory and field research on brittle root disease of horseradish (*Armoracia rusticana*) require efficient manipulation of the causal agent *Spiroplasma citri* and its vector *Circulifer tenellus* for transmission experiments. The following salient features of transmission have thus far been elucidated using *Brassica rapa* as source and test plants: Individuals of *C. tenellus* transmitted *S. citri* with frequencies ranging from 64% to 89%. To date an incubation period of 7-12 days of the pathogen in vectors has been demonstrated. Leafhoppers transmitted spiroplasmas after acquisition access feedings of 1 1/2 hr but not 1/2 hr. In tests of inoculation times with single insects, 22% of the leafhoppers-transmitted *S. citri* after 30 minute feedings on test plants. These results are of value in brittle root management programs on horseradish directed at insecticidal control of vector populations.

EVIDENCE FOR INVALID RESULTS IN THE USE OF ELISA FOR DETECTION OF SPIROPLASMA CITRI IN PLANTS AND INSECTS. Jacqueline Fletcher, Kathleen M. Franklin, and Robert M. Goodman. Dept. Plant Pathology, University of Illinois, Urbana, IL 61801.

ELISA, an accepted assay for the detection of certain plant pathogens, was used to detect *Spiroplasma citri* in tissues. For periwinkle, turnip, radish, and three brassicaceous weeds, results of simultaneous ELISA and isolation attempts always agreed. With horseradish, many samples shown positive by isolation were missed by ELISA. Addition of horseradish extract to cultured spiroplasma samples significantly reduced color development. Samples of field-collected dandelion, aster, goldenrod, bull nettle, vetch, plantain, and plum, tested as possible reservoirs of the agent, gave positive reactions in ELISA which could not be confirmed by isolation. Two weeds gave positive ELISA reactions in the absence of antibody-enzyme conjugate. ELISA results were reliable for tests on beet leafhoppers, but not for aster leafhoppers, used in laboratory transmission tests. Interpretation of ELISA results requires careful controls to ensure validity.

CONTROL OF PEACH YELLOW LEAF ROLL WITH TREE INJECTION OF TERRAMYCIN. George Nyland, B. C. Raju, and S. K. Lowe. Department of Plant Pathology, University of California, Davis, CA 95616

An epidemic of peach yellow leaf roll (PYLR) in northern California continues. Present evidence indicates that sources of inoculum are outside the orchards with little or no tree-to-tree spread making tree removal of little or no value. Terramycin^R solution containing 1.5-2.0 g/tree of Terramycin injected into trees by either pressure or gravity soon or after harvest results in near normal crop production on treated trees the following year. In three orchards of peach trees with a total of 1409 trees aged 5, 12 and 15 years, treatments reduced percent of trees with any symptoms the year following infection by 94, 74 and 46%, respectively. Even in treated trees that showed symptoms only slight reduction in crop was noted. Best results of treatments are with infected trees less than 12 years old. Terramycin was significantly superior to 8 other chemicals tested in remitting symptoms of PYLR. PYLR is a lethal disease. Tree injection with Terramycin is highly cost-effective.

Properties and Relationships of Two Xylem-Limited Bacteria and a Mycoplasma-like Organism Infecting Bermudagrass. M.J. Davis, University of Florida, IFAS, AREC, Ft. Lauderdale, FL 33314; R.H. Lawson, A.G. Gillaspie, Jr. and R.W. Harris, USDA, BARC-West, Beltsville, MD 20705.

A bacterium isolated from Bermudagrass, but not the sugarcane ratoon stunting disease (RSD) bacterium, incited a severe stunting described herein as Bermudagrass stunting disease (BSD). Bermudagrass with white leaf disease (WLD) associated with a mycoplasma-like organism (MLO) exhibited chlorosis, excessive axillary shoot development, and stunting. The combination of BSD and WLD manifested a severe disease often causing early death of the plant. Associations of BSD bacterium with BSD, of MLO with WLD, and of both organisms with early death were confirmed by electron microscopy. Cell walls of the RSD and BSD bacteria contained major amounts of 2, 4-diaminobutyric acid, glycine, glutamine, alanine, fucose, and rhamnose. A 66-68 mole % G+C content of DNA for both bacteria was calculated from T_m determinations. These results are consistent with the hypothesis that the RSD and BSD bacteria are plant pathogenic corynebacteria.

IN VITRO ANTIBIOTIC SUSCEPTIBILITY OF XYLEM-LIMITED PLANT PATHOGENIC RICKETTSIA-LIKE BACTERIA. C. J. Chang and N. W. Schaad, Dept. of Plant Pathology, Univ. of Georgia, Georgia Experiment Station, Experiment, GA 30212.

The minimal inhibitory concentrations of 12 antibiotics to 3 strains of Pierce's disease (PD), 1 strain of plum leaf scald, and 2 strains of each of phony disease of peach, periwinkle wilt, and elm scorch organisms (ES) were determined by agar dilution technique. CS20, an improved agar medium, developed for the susceptibility tests contained soy peptone, tryptone, $(\text{NH}_4)_2\text{HPO}_4$, KH_2PO_4 , $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, Hemin Cl, L-glutamine, L-histidine, starch, dextrose, phenol-red, and agar. All strains were resistant at 40 $\mu\text{g}/\text{ml}$ or greater to bacitracin, gentamycin, kanamycin, neomycin, and streptomycin. Chloramphenicol and tetracycline at 6.25 $\mu\text{g}/\text{ml}$ or less inhibited all strains. Penicillin, cephalothin, and ampicillin at 6.25 to 25.0 $\mu\text{g}/\text{ml}$ were active against all strains. Carbenicillin at 12.5 $\mu\text{g}/\text{ml}$ or less was active against all PD strains, however carbenicillin was much less active against the other organisms. Erythromycin was active at 12.5 $\mu\text{g}/\text{ml}$ or less against all strains except PD and ES organisms.

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THE EFFECT OF PLANT SIZE ON THE EPIDEMIC DEVELOPMENT OF DISEASE IN GENETICALLY DIVERSE HOST POPULATIONS. C. C. Mundt, and J. A. Browning, Dept. of Plant Pathology, North Carolina State Univ., Raleigh, NC 27650 and Dept. of Plant Science, Texas A&M Univ., College Station 77843.

We used oat crown rust as a model to test the effect of plant size on the disease-reducing effectiveness of host-mixtures. Genotypic mixtures of crops with differing plant sizes were simulated by altering the planting arrangement of oat isolines within plots. Increasing simulated plant size up to 288 times that of a single oat plant did not significantly affect the disease-reducing effectiveness of the mixtures. In mixtures of susceptible and resistant oats, decreasing the percentage of susceptible plants from 100 to 25% accounted for more of the disease-reducing effect of the mixture than did dispersing the susceptible plants among the resistant ones. Our results suggest that multilines and cultivar mixtures, which have been used successfully for small grain crops, may also be effective for crops with a larger plant size.

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MULTISPECTRAL SENSING OF LEAF RUST OF WHEAT. D. Marshall and G. Shaner, Department of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907.

Epidemics of leaf rust (*Puccinia recondita*) of wheat that occur before or during flowering can reduce both the amount and size of grain. Information on how fast and how far leaf rust spreads needs to be accurate and timely. Spectroradiometric readings indicated that rusted wheat (cvr. Monon) could be detected best in the 0.55 to 0.70 μm , 1.45 to 1.85 μm , and 2.00 to 2.30 μm wavelength regions. Reflectance of rusted wheat was first significantly different ($\alpha=0.05$) from non-rusted wheat at severities of 0.1 to 0.2%. Reflectance of rusted wheat increased at a rate similar to that of the apparent infection rate of disease progress ($R^2=0.951$). Leaf rust severity could be predicted from reflectance values at 0.65 μm by $\hat{Y} = -9.873 + 1.173X$ and at 2.00 μm by $\hat{Y} = -8.867 + 1.134X$. Both spatial and temporal leaf rust progress could be accurately determined by multispectral sensing.

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SPECTRAL CLASSIFICATION OF WHEAT INFECTED WITH BARLEY YELLOW DWARF AND STRIPE RUST. S. P. Pennypacker, Dept. of Plant Pathology, The Pennsylvania State University, University Park, PA 16802; A. L. Scharen, E. L. Sharp, and D. C. Sands, Dept. of Plant Pathology Montana State University, Bozeman, MT 59717.

Plant canopies of spring wheat and winter wheat grown in Montana were "described" by spectroradiometric data recorded at 10 nm intervals over the 380-800 nm region. Concurrent data included crop height, growth stage, and visual estimates of disease severity. Crop yield estimates and protein content were recorded for each site. The spectral reflectance of the canopy changes as the plants develop and as disease severity increases. A distinct reflectance difference exists between healthy plants and plants possessing barley yellow dwarf (BYD) symptoms. A characteristic signature also distinguishes stripe rust from healthy and BYD infected plants.

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PROBLEMS ENCOUNTERED IN FITTING DISEASE-PROGRESS CURVES TO THE WEIBULL FUNCTION. R.D. Berger, Univ. Florida, Gainesville 32611

The Weibull function [$y=1-\exp(-(t-a)^c)$] has been used to characterize disease type (ideally, shape $(c)=1$ for simple or 3.6 for compound interest). Several problems were encountered when natural or generated disease-progress curves were fitted to the function by ordinary least-squares programs. Poor estimates of a , k , and c were obtained if all three terms were solved simultaneously regardless of starting values. For a logistically generated curve, estimates of a , k , and c varied with the portion of the curve selected for the fit. Also, as y_{max} decreased, the k and c values decreased. The best parameter estimates for unfamiliar curves were obtained if only one or two terms were solved and if there was a y value near the pivotal 0.63. The time (t) when $y_0=0.001$ was used as an estimate of position (a) . If the curve to be fitted was sigmoidal, the rate (k) was approximated as $0.15r$ ($r=\text{logistic rate}$). Several runs with alternating parameter searches and updated estimates were required to obtain the best fit. The problems with curve fitting and the variable c values for epidemics did not allow reliable characterization of disease type by the Weibull function.

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APPLICATIONS FOR A REGRESSION-DERIVED SOYBEAN PLANT GROWTH MODEL. Steven B. Johnson and R. D. Berger, Department of Plant Pathology, University of Florida, Gainesville, FL 32611.

A soybean growth model was derived with regression techniques on whole-plant samples collected in 1981. The model, driven by leaf dry-matter accumulation over time, partitioned the plant dry matter into that of stems, petioles, leaves, pods, and seeds. The model was validated with 200 data sets from 1980 and 80 data sets from 1981. The intended applications of the model are establishment of disease threshold levels and disease intensity / yield loss relationships. Experiments with specific-acting and selective chemicals were performed over two years, but the range of disease intensity was sufficient to calibrate the model only partially. The *ex post facto* nature of the model does limit its value in prediction, but it is useful for the intended application.

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PREDICTION OF STRIPE RUST EPIDEMICS ON WINTER WHEAT USING STATISTICAL MODELS. S.M. Coakley and R.F. Line, NCAR, P.O. Box 3000, Boulder, CO 80307 and ARS-USDA, WSU, Pullman, WA 99164.

Statistical models developed for predicting stripe rust (caused by *Puccinia striiformis*) on winter wheat cultivars Omar, Gaines, and Nugaines at Pullman, Washington can be used at four other locations in the Pacific Northwest, if negative degree days data used to develop the models are standardized (NDD₂) before regression equations are calculated. Using the NDD₂ models for 1970 to 1979 resulted in no significant difference between mean disease intensity and mean predicted disease intensity at any of the sites. The models developed were used in combination with data on vulnerability of cultivars and distribution, prevalence, and pathogenicity of rust in the fall to accurately predict stripe rust epidemics in 1980 and 1981. In 1981, the predicted epidemic was part of the documentation used for obtaining emergency registration of the fungicide Bayleton, for control of rust.

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FREQUENCY OF BACTERIAL ICE NUCLEI ON SNAP BEAN (*PHASEOLUS VULGARIS* L.) LEAFLETS AS A PREDICTOR OF BACTERIAL BROWN SPOT. S. S. Hirano, D. I. Rouse and C. D. Upper*, Dept. of Plant Pathology and *ARS, SE, USDA, Univ. of Wisconsin, Madison, WI 53706

The frequency of high populations of epiphytic *Pseudomonas syringae* pv. *syringae* on individual snap bean leaflets can be used as a predictor of brown spot incidence under field conditions (Lindemann et al., *Phytopathology* 71:890 1981). We estimated the frequency with which high populations of pv. *syringae* occurred on individual bean leaflets by a freezing assay based on the ice nucleating ability of the bacterial pathogen. The frequency with which leaflets froze at -2 to -2.5°C was highly predictive of brown spot incidence. Bean seeds of the cv. 'Eagle' were a) inoculated with the pathogen, b) soaked in streptomycin, or c) left untreated and planted in replicated field plots. The foliage in these plots was either not treated, or sprayed with Kocide according to two application schedules. Significant correlations ($p<0.05$ to 0.001) resulted for each of 8 separate times that plants were assayed by freezing and rated for disease 8 days later.

FREQUENCY WITH WHICH OAT SEEDS ARE INFESTED WITH *PSEUDOMONAS SYRINGAE* PV. *CORONAFACIENS* AS A PREDICTOR OF HALO BLIGHT DISEASE. S. S. Hirano, D. I. Rouse, D. C. Army and C. D. Uppert*. Department of Plant Pathology, *ARS, SE, USDA, University of Wisconsin, Madison, WI 53706.

Since *P. syringae* pv. *coronafaciens*, the casual agent of halo blight of oats, is ice-nucleation-active, the presence of the pathogen on oat seeds can be detected by a freezing assay. Oat seeds were germinated for five days, after which time each seedling was submerged in sterile phosphate buffer in a test tube and subjected to -4 C for 30 min. For a given seed lot, the number of seedlings out of 100 to 200 that froze was used as an estimate of the frequency with which individual seeds harbored the pathogen. These estimates for each of five seed lots representing five oat cvs. were highly predictive of halo blight incidence in field plots at 53 days after planting (DAP) in 1980 ($r=0.993$, $P<0.001$). In 1981, these estimates for each of 10 seed lots representing five cvs. were predictive of halo blight up to 55 DAP. Correlations were significant ($P=0.05$ to 0.001) for disease measurements made every 3 days from 37 to 55 DAP.

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EFFECTS OF RADIATION ON THE GERMINATION OF *ALTERNARIA SOLANI* CONIDIA. R. E. Stevenson and S. P. Pennypacker, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Germination of conidia required relative humidities (RH) > 96% and proceeded at a maximum in darkness with temperature near 25 C only when free moisture was present. Conidia were maintained under controlled temperature and moisture conditions to study the effects of radiation on germination. Conidia were irradiated (I) with five intensities of "sunlight" (> 300 nm) and selected narrow bands of "sunlight" for 11 hr at 20 C. Non-irradiated (N) conidia were maintained in darkness at 20 C for 11 hr. The I and N conidia were allowed to germinate in darkness for 3, 6, 9, and 12 hr at 10, 15, 20, 25, 30, and 32 C with RH > 96%. The ratio of mean germination for I to N conidia indicated an inverse relation between germination and light intensity. The 400-500 nm region of the spectrum was common to all treatments where reduced germination occurred.

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EFFECT OF RELATIVE HUMIDITY ON PRODUCTION OF CONIDIA BY *CERCOSPORA ARACHIDICOLA* ON PEANUT LEAVES. H. A. Melouk, and D. L. Ketring. USDA-ARS, Departments of Plant Pathology and Agronomy, Oklahoma State University, Stillwater 74078.

Leaflets of Tannut 74 with mature lesions of *Cercospora arachidicola* were maintained at various relative humidities (RH) over calcium chloride solutions in Parafilm-sealed petri plates (150 x 20 mm) at 25 ± 1 C under continuous light (800 lux) for 96 hr. Conidial production was expressed as conidial density (conidia/mm² of necrotic area) and determined as described earlier (Peanut Sci. 8:11-12, 1981). Conidial density was highest on leaflets incubated in 100% RH and lowest at 93% RH. Psychrometrically determined water potentials of infected leaflets incubated in 93% and 90% RH were -50 and -55 bars, respectively. Conidial production ceased when leaflets were incubated in 90% RH. Exposure of lesions on leaflets from 1 to 3 hr to 100% RH, followed by incubation in 96% RH for 96 hr, significantly increased conidial production as compared to incubation at a constant RH of 96%.

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INFLUENCE OF DEW PERIOD AND TEMPERATURE ON LESION DEVELOPMENT IN ONION BY *BOTRYTIS SQUAMOSA*. S. C. Alderman and M. L. Lacy, Dept. of Botany and Plant Pathology, Michigan State University, East Lansing, Mich. 48824.

One month old onion plants sprouted from bulbs were inoculated with 2.0 mg (ca. 10^6) dry conidia using a 61 cm diam X 77 cm settling tower. This method resulted in a higher number of lesions than using a water suspension of an equal number of conidia. Lesion production was optimum at 20 C, with lesions forming after 8 hr in the dew chamber (at least 7 hr continuous dew). Lesion numbers increased sigmoidally through 24 hr in the dew chamber. Lesion production was lower at 15 C and greatly reduced at 25 C. On leaf surfaces, 70-80% of the conidia germinated and 60-70% caused lesions after 24 hr in the dew chamber at 20 C. Hyphal development within lesions increased with increasing periods of constant dew at 20 C, but was restricted under conditions of low (50% ± 10%) relative humidity. Sporulation was initiated after 3-4 days of constant dew at 20 C.

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FURTHER STUDIES ON THE EFFECT OF INTERRUPTED WET PERIODS ON INFECTION OF SOUR CHERRY BY *COCOSMYCES HIEMALIS*. Scott P. Eisenmith, Tropeninstitut, Justus Liebig-Universität, D-6300 Giessen, Federal Republic of Germany.

Montmorency sour cherry (*Prunus cerasus*) trees inoculated with conidia of *C. hiemalis* were subjected to interrupted wet periods (IWP) and continuous wet periods (CWP) of various durations. Lesion numbers were independent of the dry interruption length in IWP having 4 hr initial wet periods separated from 8 hr final wet periods by dry interruptions of 1, 2, 3, 4, 5, and 6 hr, and in IWP having 4 hr initial wet periods separated from 24 hr final wet periods by dry interruptions of 4, 8, 12, 16, 20, and 32 hr. IWP with initial 8 hr wet periods, 8 hr dry interruptions, and 4 or 8 hr final wet periods resulted in fewer lesions than IWP with 12, 24, or 48 hr final wet periods. Lesion numbers from an IWP with 8 hr initial and final wet periods separated by an 8 hr dry interruption were not different from 8 or 16 hr CWP; however, infection from an 8 hr CWP was less than from a 16 hr CWP.

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EFFECT OF SEEDBORNE INOCULUM AND SOIL MOISTURE ON DEVELOPMENT OF FUSARIUM STALK ROT OF CORN IN KANSAS. M. A. El-Meleigi and L. E. Clafflin, Department of Plant Pathology, Kansas State University, Manhattan, KS 66506.

Soilborne and airborne inocula of *Fusarium moniliforme* Sheld. and its occurrence in roots and stalks of corn (*Zea mays* L.) were monitored throughout the growing season at two Kansas locations in 1980. Four soil moisture levels and seeds of two corn hybrids with *F. moniliforme* infestation levels of 2, 12, 25, and 50% were utilized. Airborne inocula levels reached a maximum of 0.02 propagules/l of air during August. Levels of soilborne inoculum increased with time, with the highest level (300 prop./g soil) found under the dryland conditions. An overall average of 30% of the corn seedlings were colonized regardless of soil moisture, seedborne inoculum level, or hybrid. Incidence of stalk rot was highest (22%) under dryland conditions and lowest (6%) with the soil moisture level of 50% of field capacity. A higher percentage of roots (90%) than stalks (first internode above brace roots; 45%) were colonized at maturity.

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Evaluation of Some Factors Affecting *Fusicladium effusum* Conidia Dispersal and a Pecan Scab Epidemic. A. J. Latham, Ala. Agric. Exp. Stn., Auburn University, AL 36849.

Investigations were conducted in unsprayed pecan orchards to relate some meteorological factors to numbers of airborne conidia of *Fusicladium effusum* dispersed and to the development of pecan scab. Dispersal of conidia was diurnal with peak numbers recorded by spore traps at 1200 hr. Few conidia were dispersed at night when leaves were wet, the relative humidity (RH) was 100%, or wind was absent. Highest daily numbers of conidia were recorded during the last week of May and the first 2 weeks of June. Numbers of airborne conidia were positively correlated with wind velocity, temperature, and rain, and negatively correlated with RH during May, June and part of July 1974, 1975 and 1980. Contributing to the scab epidemic of 1975 were the following factors: lesion numbers for the fourth week of April were nearly equivalent to the fourth week of May of non-epidemic years, numbers of airborne *F. effusum* conidia were 63% higher than in non-epidemic years, and rainfall occurred weekly during May and June.

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INFLUENCE OF ENVIRONMENT ON INFECTION BY *PHOMOPSIS* SP. OF SOYBEAN SEEDLINGS PLACED IN THE FIELD. J. C. Rupe and R. S. Ferriss, Department of Plant Pathology, University of Kentucky, Lexington, Ky. 40546.

The influence of rain and in-canopy environment on infection by *Phomopsis* sp. was studied with potted soybean seedlings placed in the field at various times during the season. Weekly from 7/6/81 to 10/29/81, 2-week-old greenhouse-grown seedlings were placed in a soybean field for 3 days before being returned to the greenhouse. Half of the seedlings were then misted with water and placed in a moist chamber for 2 days. Control plants remained in the greenhouse but otherwise were treated the same. After 2 days, the petioles and stems were cut into 1 cm pieces, surface sterilized, and plated on amended PDA for determination of the proportion of infected pieces. No *Phomopsis* was found on control plants. Infection of the field seedlings was higher on the moist chamber treated plants than the nontreated. However, both were significantly correlated ($P<0.001$) in a multiple regression model to temperature, leaf wetness, rain, and date, during the field incubation period.

ETHYLENE EFFECTS ON IN VITRO AND IN VIVO GROWTH OF CERTAIN POSTHARVEST FRUIT FUNGI. M. K. El-Kazzaz, N. F. Sommer, A. A. Kader

Exposure of certain postharvest fungi namely, *Alternaria alternata*, *Botrytis cinerea*, *Colletotrichum gloeosporioides*, *Monilinia fructicola*, *Penicillium digitatum*, *P. expansum*, *P. italicum*, *Rhizopus stolonifer* and *Thielaviopsis paradoxa*, to 1, 10, 100 and 1000 ppm C_2H_4 stimulated germination and/or germ tube elongation but did not influence final growth rates at 20°C. Ethylene concentrations up to 28,000 ppm increased the total dry weight of *B. cinerea*, as determined by glucosamine content, after 4 days at 20°C. Ethylene treatments did not influence lesion diameter on Navel oranges inoculated with *P. italicum* before C_2H_4 exposure was initiated. However, when oranges were treated with C_2H_4 for 3 days at 20°C before inoculation, continued exposure to C_2H_4 reduced lesion diameter in proportion to its concentration.

CALCIUM INFILTRATION OF 'GOLDEN DELICIOUS' APPLES AND ITS EFFECT ON POLYPHENOL OXIDASE ACTIVITY, TOTAL PHENOLIC CONTENT, AND DECAY. W. S. Conway and C. E. Sams, Hort Crops Quality Lab, HSI, ARS, S&E, U.S. Dept. of Agriculture, Beltsville, MD 20705.

'Golden Delicious' apples were treated with 0, 2, 4, 8 or 12% solutions of calcium chloride by dipping, vacuum infiltration (250 TORR) or pressure infiltration (68.95 kPa). The apples were then placed in storage (0°C). After 5 months, the fruit were removed from storage, wounded on two sides and inoculated with a conidial suspension of *Penicillium expansum*. Following additional storage of 20°C for 7 days, the apples were rated for decay severity by measuring the diameter of the decayed area at the inoculation sites. Fruit tissue was analyzed for calcium content and those apples pressure infiltrated with calcium chloride were also analyzed for total phenolic content and polyphenol oxidase (PPO) activity. Decay reduction was most evident in those apples pressure infiltrated with the higher levels of calcium chloride solution. However, phytotoxicity was greatest at these levels as well. Total phenolic content and PPO activity was not affected by an increase in calcium content of the fruit.

RESIDUES AND CONTROL OF *PENICILLIUM DIGITATUM* WITH POSTHARVEST APPLICATIONS OF IMAZALIL TO ORANGES. G. E. Brown and S. Nagy, Florida Department of Citrus, AREC, 700 Experiment Station Road, Lake Alfred, FL 33850.

Imazalil is an imidazole fungicide effective for control of green mold caused by *Penicillium digitatum*. Imazalil was applied either in water or resin solution water wax using a non-recovery spray applicator to oranges revolving on horsehair brushes saturated with the treating solution. Applications in water resulted in higher residues than applications in water wax. Residues from water treatments were also enhanced by increased time on the brushes. Injured peel of treated fruit contained higher residues than surrounding non-injured areas. Residues on fruit washed after treatment were reduced only slightly. Control of sporulation by *P. digitatum* was better with imazalil applied in water. Injuries occurring after treatment were less susceptible to infection if imazalil was applied in water rather than resin solution wax.

IN VIVO DETERMINATION OF FUNGI RESISTANT TO POST HARVEST FUNGICIDES IN THE CITRUS INDUSTRY. Petrie, J.F. and B. A. Dave, Pennwalt Corp., 1713 S. Calif. Av., Monrovia, CA 91016.

In an effort to determine if a fungicide or combinations of fungicides are effective in controlling decay in citrus packinghouses, the Decco Tiltbelt Research Laboratory applied the following method beginning in 1971. This simple method consists of exposing scratched lemons, with the scratch side up, on a table near the packout dump at a lemon packinghouse to expose the scratch to the fallout of spores at the dump. The uniqueness of this method is the utilization of the actual spore load in the commercial packinghouse to inoculate the lemons, rather than the standard method of making up an inoculum from laboratory cultures. A check lot determines whether the fallout adequately inoculated the lemons. Two other lots are marked and put through the normal wash and packout treatments. Others are dipped in test fungicide solutions for experimental and comparison use.

A SELECTIVE MEDIUM FOR *PENICILLIUM DIGITATUM* (SACC.), CITRUS GREEN MOLD. J. L. Smilanick and J. W. Eckert. Department of Plant Pathology, University of California, Riverside, CA 92521.

Media varying in nutrients, pH, and inhibitory compounds were evaluated for selective development of colonies of *P. digitatum* and suppression of fungal contaminants in plates exposed to the atmosphere of citrus groves and packing houses. Thirteen species of common air-borne fungi were replica-plated to the suppressive media at 25°C. o-Phenylanisole (OPA) and 2,3,4-6 tetrachloroanisole (TCA) were the most selective agents evaluated when added to potato dextrose agar enriched with 2g/l each of peptone and yeast extract (PDAPYE), while pentachloronitrobenzene (PCNB) and acidification to pH 3.5 were less effective. The PDAPYE medium (pH 5.5) amended with 500 µg/ml OPA and 100 µg/ml PCNB restricted colony size of *P. digitatum* and greatly reduced contamination for more efficient enumeration of air-borne spores of this pathogen.

A COMPARATIVE STUDY OF GROWTH CHARACTERISTICS AND PATHOGENICITY OF *MUCOR PIRIFORMIS* ISOLATES CAUSING DECAY OF PEACHES AND NECTARINES. T. J. Michailides and J. M. Ogawa. Department of Plant Pathology, University of California, Davis 95616

Mucor piriformis isolated for the first time from decaying peaches and nectarines from California (CA) and Chile (CH) showed almost identical microscopic features. Both isolates exhibited similar mycelial growth rates on synthetic mucor agar (SMA) at temperatures ranging from 0 to 26°C. They both showed occasional columellar germination and septation; yet the CA isolate developed higher mycelial turf. The CH isolate showed higher spore germination percentages on SMA at 0, 6, and 20°C. Spores from both isolates preincubated at 27°C for 2 to 10 days showed a reduction in their ability to germinate on SMA at 21°C after 24 hr, though this reduction was greater for the CH isolate. The CH isolate was more pathogenic than the CA isolate based on lesion size, amount of tissue decayed, and pectinase production on peach fruit when inoculated and incubated at 0, 6, and 20°C.

EFFECT OF TRITON X77 ADDITION TO SEVERAL FUNGICIDES ON D'ANJOU PEAR DECAY CONTROL. R. A. Spotts, OSU Mid-Columbia Experiment Station, Hood River, OR 97031.

Wounded d'Anjou pear fruits were immersed 5 min in 0.32% sodium orthophenylphenate tetrahydrate (SOPP), 0.32% SOPP+0.5% Triton X77, or water at 0, 10, and 20°C. Each solution contained 1.0, 2.0, or 10.0 x 10⁴ conidia/ml of *Penicillium expansum*, *Botrytis cinerea*, or *Mucor piriformis*, respectively. SOPP reduced *B. cinerea* but not *M. piriformis* or *P. expansum* decay. Addition of Triton X77 to SOPP increased decay of all fungi. Solution temperature did not affect decay. Addition of Triton X77 to captan or iprodione increased decay of *M. piriformis* and *P. expansum*, and Triton X77 addition to captan or etaconazole increased *B. cinerea* decay. In 2 packinghouses, treatment of wound-inoculated pear fruit prior to inoculation with a commercial detergent containing 4.4% SOPP resulted in increased *M. piriformis* and *P. expansum* decay. SOPP treatments did not reduce fungal spore germination and appeared to stimulate *M. piriformis* germ tube growth. Results were attributed to fungistatic rather than fungicidal nature of the tested fungicides.

INTERACTION AND CONTROL OF *ALTERNARIA* STEM DECAY AND BLUE MOLD IN ANJOU PEARS. J.W. Sitton, USDA-ARS, Dept. Pl. Pathology, Washington State University, Pullman, WA 99164-6430 and C.F. Pierson, USDA-ARS, TERC, Wenatchee, WA 98801.

Decay of stems of d'Anjou pears by *Alternaria alternata* is found in fruit treated with a benomyl postharvest drench for control of *Penicillium expansum* and subsequently held in storage for prolonged periods. Benomyl favors *A. alternata*, a slower growing and less competitive fungus than *P. expansum*, at the osmotic potential (-22 bars) and temperature (near 0°C) conditions of pears in storage. Prochloraz, chlorothalonil, iprodione, and triadimenol, in combination with benomyl were effective in controlling both *Alternaria* stem decay and blue mold. The most effective control of both *Alternaria* stem decay and blue mold was obtained with 1000 ppm prochloraz combined with 500 ppm benomyl. Reduction of linear growth of *A. alternata* on potato dextrose agar amended with prochloraz was a useful method for determining the most effective concentration for disease control.

EFFECT OF TEMPERATURE AND RELATIVE HUMIDITY ON GROWTH OF MOLDS IN HARVESTED WALNUTS. J. E. Barton, M. N. Schroth, A. R. Weinhold. Department of Plant Pathology, Berkeley, CA 94720.

Field-collected, mature walnuts with average moisture content of 19% were placed into chambers with ten different combinations of relative humidity and temperature to determine the best conditions for drying and storage. Relative humidities were between 80% and 100% and temperatures were between 15C and 27C. Predominant genera of mold fungi were *Penicillium*, *Aspergillus*, and *Alternaria*. First evidence of mold development occurred after 2 days. There was a higher percentage of walnuts exhibiting mold at the higher humidity/temperature regimes. All walnuts were moldy after 10 days at 100% RH at all three temperatures. At 90%, RH the percentages of moldy walnuts after 10 days were 40, 63, & 77 at 15, 21, & 27C, respectively. At 80% RH there was little or no mold at any of the temperatures. Dried and non-dried walnuts were tested to determine whether previous drying had an effect on the amount and timing of mold formation. Results indicated that there was a factor in non-dried walnuts that inhibited mold formation.

POSTHARVEST DISEASES OF JICAMA. B. D. BRUTON, Crop Protection and Production Research Unit, USDA-ARS, Weslaco, TX 78596.

Jicama (*Pachyrhizus erosus*) is a tropical root crop grown primarily in Mexico and to a limited extent in the Rio Grande Valley of Texas. Average losses in excess of 10% have been observed during storage in packinghouses. Mechanical injury during the harvesting and marketing process is prerequisite to infection by *Cladosporium* sp., *Penicillium* spp., and *Rhizopus stolonifera*. Inoculation of each fungus alone or in combination on nonwounded roots resulted in no infection. Of 126 jicama roots examined at retail food stores, 44% had blemishes of which 34.1% were incited by *Cladosporium*, 9.5% by *Penicillium* and 0.8% by *Rhizopus*. *Cladosporium* and *Penicillium* were primarily superficial colonizers; whereas, *Rhizopus* caused internal decay. Losses of 100% have been observed in jicama stored at 7 C and 80% RH for 28 days. Effective control measures include post-harvest dip of roots in 2000 ppm ClO₂ at room temperature for 1 min, followed by drying and storage or retail display at 20-25 C and 65-70% RH.

BACTERIAL SOFT ROT IN POTATO TUBERS AFTER VARIOUS WASHING AND DRYING PROCEDURES. J. A. Bartz and A. Kelman, Dept. of Plant Pathology, University of Wisconsin, Madison, WI 53706.

Bacterial soft rot incidence (BSRI) and severity (surface area decayed = SAD) in potato tubers (cv. Russet Burbank) from commercial storage were determined after different washing and drying treatments followed by incubation in a mist chamber for 4 days at 20 C. When tubers were immersed briefly (<5 sec) in water, the BSRI and SAD were 26 and 1%, respectively. In contrast, for tubers immersed in suspensions of *Erwinia carotovora* pv. *carotovora* (Ecc) under various experimental conditions, the BSRI and SAD ranged from 60 to 100% and 4 to 77%, respectively. The SAD increased as length and depth of immersion, CFU of Ecc in suspension, and surfactant (Triton X-100) concentration increased; the BSRI was usually 100% whenever surfactants were present or inoculum concentration exceeded 5×10^3 CFU/ml. However, when air-dried at 22 C for 2 or 4 hrs before incubation, tubers inoculated in absence of surfactant had a BSRI and SAD equal to those of non-inoculated tubers.

COMPARATIVE STUDIES WITH TWO GEOTRICHUM SPECIES CAUSING POST-HARVEST DECAY ON TOMATO FRUIT. H. E. Moline, Hort. Crops Quality Lab., HSI, ARS, S&E, U.S. Dept. of Agriculture, Beltsville, MD 20705.

Growth rates of *Geotrichum candidum* and *G. penicillium* were compared on mature-green and red tomato fruit, and potato dextrose agar (PDA) at 5, 10, 15, 20 and 25 C. The growth rate of *G. penicillium* on green fruit, red fruit, and PDA was less than that of *G. candidum* at all temperatures. *G. penicillium* appears to be a slightly less aggressive pathogen and has a significantly slower growth rate on PDA at all temperatures below 20 C than does *G. candidum*. The fungicidal effects of a number of chemicals were studied on green and red fruit inoculated with suspensions of spores of the 2 species. Sodium hypochlorite, Benlate, Topsin M, and Ronilan slightly inhibited the growth of *G. penicillium* on green fruit, and Imazalil and Ferbam slightly reduced growth of this fungus on red fruit. Sodium bicarbonate and potassium sorbate slightly inhibited *G. candidum* growth on green fruit, and Topsin M and sodium bicarbonate slightly reduced its growth on red fruit.

ELICITATION OF TERPENOID ALDEHYDE PHYTOALEXINS IN *VERTICILLIUM DAHLIAE*-INFECTED COTTON STEM. M. E. Mace

Terpenoid aldehyde (TA) phytoalexins occur in scattered paravascular parenchyma cells appressed to vessel walls in *Verticillium*-infected cotton stems. These cells may be biochemically specialized for TA synthesis; however, they may represent random sites of *Verticillium*-induced injury. Thus, stem xylem of infected *Verticillium*-resistant *Gossypium barbadense* was examined histochemically in detail for localization of TA phytoalexins. TA phytoalexins were found in cells located several cells away from the pathogen isolated in the vessel lumen at 1 week after inoculation. Physical contact of these specialized parenchyma cells and the pathogen, therefore, is not required for TA synthesis. Furthermore, phytoalexin elicitor(s) produced in vessels by *Verticillium* or by *Verticillium*-injured parenchyma cells appressed to vessel walls apparently migrate from these sites and elicit TA synthesis in specialized cells well removed from the infected vessel.

INCIDENCE OF STORAGE FUNGI IN FARM-STORED GRAIN. D. B. Sauer and C. L. Storey, U.S. Grain Marketing Research Laboratory, USDA, ARS, 1515 College Avenue, Manhattan, KS 66502.

When 100 surface-disinfected kernels were plated from each of 2500 samples taken from farm bins in 1980, *Aspergillus glaucus* was found in 84% of the corn, 70% of the oats, and 37% of the wheat samples. Corn, oats, and wheat samples with *A. glaucus* had averages of 26, 20, and 14% of the kernels invaded, respectively. Fungal contamination varied little in corn from four crop years. Wheat from the most recent (1979) crop year was low in storage fungi and high in field fungi compared to earlier crop years. Storage fungi averaged higher in samples with higher moisture contents as received, even though most moisture contents were too low to permit mold growth. *A. glaucus* was also higher in northern compared to southern states in the central plains and was higher in samples with insects. *A. flavus* was found in an average of 1.2% of all corn kernels and in less than 0.1% of the oats and wheat. We gratefully acknowledge USDA's Agricultural Stabilization and Conservation Service for providing the samples.

SUPPRESSION OF THE BIOLOGICAL CONTROL AGENT *TRICHODERMA HAMATUM* ON SEEDS BY SOIL-BORNE *PSEUDOMONAS* SPP. J. P. Hubbard, Y. Hadar, and G. E. Harman, Agrigenetics Corp., 1120-220th St. W., Farmington, MN 55024 (1st author) and Dept. of Seed & Veg. Sci., New York State Agricultural Experiment Station, Geneva, NY 14456.

Trichoderma hamatum, applied as conidia to pea seeds, may colonize the seed coats and protect germinating seeds from attack by seed rot organisms in some soils, but not in others. The erratic performance of *T. hamatum* as a biological control agent may be due in part to competition and/or antagonism from fluorescent pseudomonads indigenous to the soil. In soil with reduced populations of pseudomonads (pasturized), *T. hamatum* operates successfully as a biocontrol agent, but if fluorescent pseudomonad isolates are inoculated on *T. hamatum* treated seeds immediately prior to planting, effectiveness is decreased. However, in field soils in which *T. hamatum* successfully protected seeds and in which it failed, the seed coat pseudomonad population was similar both qualitatively and quantitatively. Evidence is discussed suggesting that iron chemistry affects the interaction of fluorescent pseudomonads with *T. hamatum*.

INTEGRATION OF FLUID DRILLING AND BIOCONTROL PRACTICES TO AVOID *PYTHIUM* INFECTION OF SEEDS AND SEEDLINGS. Y. Hadar, A. G. Taylor and G. E. Harman, New York State Agricultural Experiment Station, Geneva, NY 14456.

Seeds were germinated in aerated water until radicle emergence (pregermination) and then transferred to a 1% Polysurf C gel and planted in field soil. No disease occurred when pregerminated cucumber seeds were planted in *Pythium*-infested soil at 25°C. The efficacy of pregermination in reducing seed rot decreased as soil temperature decreased, but at all temperatures more pregerminated seedlings survived. *Pythium* infection increased when dead cucumber or pea seeds were planted together with pregerminated cucumber seeds. Apparently, pregerminated seeds did not stimulate *Pythium* since the seed exudates were removed during imbibition. Gels used in fluid drilling can be used as a delivery system for biocontrol agents to soil. Spores from *Trichoderma koningii*, isolated from NY soil, were mixed in the gel in levels of 10^8 spores/ml. This mixture, when applied to *Pythium* infested soil, reduced infection in both cucumbers and peas.

EFFECT OF METHAM-SODIUM ON GROWTH AND CONIDIAL GERMINATION OF *TRICHODERMA* SPP. J. A. Lewis and G. C. Papavizas. USDA, ARS, Beltsville, Maryland 20705.

The effect of vapors from metham-sodium (MS) used at 0, 10, 25, 50, 100, 350, 700, and 1000 µg (a.i.)/ml, and of direct contact of the fumigant (at the same concentrations) on radial growth of 42 *Trichoderma* isolates and on conidial viability and germination of six isolates of the antagonist were studied in vitro. Vapors from 1000 µg of MS did not prevent growth of 41 out of 42 isolates, although the rate of growth decreased with increasing amount of the fumigant. Vapors from 500 µg of MS did not reduce conidial germination in *T. viride* (isolates T-1, T-1-R4, T-1-R9) or *T. hamatum* (isolates Th-20, Tri-4, 489-13). Contact of conidia with 100 µg/ml of MS prior to germination reduced viability of most isolates less than 20%. All isolates were viable to some extent after contact with 350 and 700 µg/ml for 1 hr, but germination was slow. Viability and germination were reduced in most isolates after 6 hr. T-1-R4 was less affected than other isolates by MS at 350 µg/ml suggesting a differential sensitivity by *Trichoderma* isolates to the fumigant. Direct contact of spores during germination with 10 µg/ml of MS reduced germination of T-1, T-1-R4, Tri-4, and Th-20 by 60, 80, 35, and 100%, respectively.

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FACTORS AFFECTING SUSCEPTIBILITY OF *SCLEROTIUM ROLFSSII* SCLEROTIA TO *TRICHODERMA HARZIANUM* IN NATURAL SOIL. Y. Henis and G. C. Papavizas. USDA, ARS, Beltsville, MD 20705.

Fresh, naturally- and PDA-produced sclerotia of *S. rolfssii* (Sr-3) were exposed to drying, washing, NaClO (1% solution), metham-sodium, and heat treatments, and immersed in 1.5 X 10⁷/ml spore suspension of *T. harzianum* (WT-6). Samples of sclerotia were incubated on natural soil and PDA supplemented with 10 µg/ml of 8-hydroxyquinoline (8-HQ) which selectively inhibited *Trichoderma* growth. Sclerotia were examined for: (i) Their ability to germinate on PDA + 8-HQ and on soil; (ii) their ability to allow sporulation of WT-6 on their surface; and (iii) for their eventual degradation. Both naturally- and artificially-produced fresh sclerotia did not germinate and were not attacked by WT-6 in natural soil. However, exposure of the sclerotia to a relative humidity of < 30% for 24-48 hr, to ≥ 100 µg/g of metham-sodium in soil for 1-4 hr, and to partial heat treatment of 90 C for 15 sec increased their germination in soil as well as their susceptibility to attack by WT-6. A synergistic relationship between metham-sodium and WT-6 was demonstrated with 80-1000 µg/g for 1-4 hr and with 20 µg/g for 16 hr in natural soil.

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DEVELOPMENT OF CHLAMYDOSPORES OF *TRICHODERMA* SPP. IN CULTURE AND IN SOIL. S. D. Cohen, J. A. Lewis, G. C. Papavizas, and G. A. Bean. USDA, ARS, Beltsville, MD 20705 and Dept. of Botany, Univ. of Maryland, College Park, MD 20742.

Development of chlamydospores from conidia of *T. hamatum* [Tri(4)], *T. viride* [T-1-R9], and *T. harzianum* [Th-7] was observed on dialysis membranes placed on agar containing either soil extract, malt-yeast-peptone (MYP), molasses-corn steep liquor or glucose-ammonium tartrate. Conidia produced germ tubes after 1 day on all media. Terminal and intercalary chlamydospores were observed on developing hyphae on all media tested. Chlamydospore formation began on MYP agar after 2 days and on other media after 3 days. Chlamydospores were also formed on hyphae emerging from conidia after 3-4 days on membranes placed in autoclaved soil with and without alfalfa tissue and in nonautoclaved soil with alfalfa. Chlamydospores did not form from conidia in natural, nonamended soil. This is the first report of chlamydospore formation in natural, amended soil.

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CONTROL OF PEA SEEDLING DISEASES WITH HOST RESISTANCE, *TRICHO-* *DERMA*, AND FUNGICIDES. John M. Kraft and George C. Papavizas, USDA-ARS-WR, P.O. Box 30, Prosser, WA 99350, and USDA-ARS-NER, Room 274, Bldg. O11A, BARC-West, Beltsville, MD 20705.

Integrated control of pea seedling diseases (primarily *Pythium* spp.) was studied at Prosser, WA, in 1980 and 1981. In both years, seeds of a root rot susceptible cv, Dark Skin Perfection and root rot-resistant breeding lines were treated with either fungicides, spores of a benomyl-tolerant mutant of *Trichoderma viride* (T-1-R4), or a combination of *T. viride* spores plus a fungicide. In both years, the root rot-resistant breeding lines outyielded the susceptible cultivar regardless of treatment. In 1981, the highest plant stands and seed yields with either the susceptible or resistant pea lines occurred when seed was treated with *T. viride* spores alone. By using a selective agar medium containing benomyl, it was determined that *T. viride* was established in pea plant rhizospheres sampled at both 2 weeks after emergence and at full bloom.

1010 PHYTOPATHOLOGY

EFFECT OF NITROGENOUS ORGANIC AMENDMENT MATERIALS ON SUPPRESSION OF PHYTOPHTHORA SOIL POPULATIONS. Leslie A. Bower and P.H. Tsao. Department of Plant Pathology, University of California, Riverside, CA 92521.

Urea, chicken manure, cottonseed, hoof and horn or feather meal were placed at 3 perimeter sites on the surface of moist artificially *Phytophthora*-infested soil (~500 propagules/gm soil) in petri dishes. After 7 or 14 days incubation, *Phytophthora* was assayed 0-12mm(A), 13-25mm(B) and 26-38mm(C) from the amendment site. All materials tested significantly lowered the *Phytophthora* populations at site A by day 7 and sites A and B by day 14. Chicken manure and feather meal completely prevented *Phytophthora* recovery at all sites by day 14. *P. cinnamomi* populations were reduced up to 87% at site C with cottonseed meal and up to 70% with hoof and horn meal at site B by day 14. *P. parasitica* was similarly affected. These tests confirm that nitrogenous organic amendment materials extend inhibitory effects to a considerable distance from the amendment material and suppression may be achieved in soils of uneven amendment distribution and at sub-eradicator dosages.

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BIOCONTROL OF *PYTHIUM APHANIDERMATUM* ON CUCUMBER BY MICROBIAL ISOLATES FROM MEXICAN SOILS. R. D. Lumsden, USDA, ARS, Beltsville, MD, 20705; G. Frias-T., CSAT, Cardenas, Mexico; R. Garcia-E., Colegio Postgraduados, Chapingo, Mexico; and M. T. Dunn, Dept. of Botany, Univ. of MD, College Park, 20742.

Damping-off of cucumber caused by *P. aphanidermatum* was much less in a natural Beltsville sandy loam soil amended with sand-bran cultures (1.0% dry wt.) of antagonistic fungi than in nonamended soil. Of 130 fungal isolates from five Mexican soils, three soils of which were naturally suppressive to *P. aphanidermatum*, 95 isolates decreased disease compared to the *pythium* control. Of these, 63 isolates controlled disease as well as or better than an Arasan[®] seed treatment. The major fungal species inducing the best disease control were *Fusarium* spp., *Trichoderma* spp., *Paecilomyces* spp., *Penicillium* spp., *Gliocladium* spp., and *Verticillium chlamydosporium*. Others that decreased disease were species of *Hemicella*, *Farrowia* and *Aspergillus*. Only one of about 30 isolates of *Streptomyces* spp. and five of about 50 isolates of bacteria controlled disease when applied as seed treatments. The five bacterial isolates were identified as *Pseudomonas cepacia*.

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EFFECT OF *LAETISARIA ARVALIS* ON POPULATION DYNAMICS OF *PYTHIUM* *ULTIMUM* IN PASTEURIZED AND NATURAL FIELD SOIL. S. B. Martin, H. C. Hoch, and G. S. Abawi, New York State Agricultural Experiment Station, Cornell University, Geneva, NY 14456.

The antagonist, *Laetisaria arvalis* (LA), was grown on corn leaf meal (CLM) or spent sugar beet pulp (BP) and incorporated (10% v/v) into natural and steam-pasteurized (60 C, 30 min) beet field soil. Pasteurized soil infested with *Pythium ultimum* and natural soil were placed in 10-cm pots and left fallow, or planted with the table beet cv. Ruby Queen and incubated in a greenhouse at 18-25 C. Propagule densities of *Pythium* spp. and LA were monitored using selective media. In one test, mean populations of *P. ultimum* at 0, 7, 21, and 35 days were 72, 467, 4,507 and 5,978 in nonamended pasteurized soil, whereas in LA-amended pasteurized soil, counts were 51, 60, 318 and 290 prop./gm. Populations of *Pythium* spp. were 455, 757, 1,468 and 1,939 in nonamended natural soil and 280, 325, 537 and 399 prop./gm in LA-amended natural soil. Populations of *Pythium* spp. remained statistically the same in fallow treatments, whereas populations of LA remained high in amended soils.

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ISOLATION OF INHIBITORS OF PHYTOPHTHORA CINNAMOMI FROM COMPOSTED HARDWOOD BARK. J. H. Wilton, L. M. Basham, H. A. J. Hootink, and R. W. Doskotch. Dept. of Plant Pathology, Ohio Agricultural Res. and Devel. Center, Wooster and Dept. of Medicinal Chemistry and Pharmacognosy, The Ohio State University, Columbus 43210.

A 60.33 kg batch of 4-month-old composted hardwood bark (mostly oak, ash and hickory) was exhaustively extracted with 95% ethanol and yielded 624 gm residue. Further fractionation by solvent/solvent partitioning produced hexane, 90% methanol, ethyl acetate and water solubles which were tested for inhibitory activity in *Phytophthora cinnamomi* and *P. citrophthora* sporangium and zoospore bioassays. The slight amount of activity found in the hexane solubles was largely due to sulfur. Most activity was in 90% methanol solubles, which were fractionated further by precipitation in various solvents, followed by acid/base partitioning and chromatography. Several synergistically-active compounds have been isolated, whose structures are currently be-

ing investigated. Major activity of the ethyl acetate fraction has been attributed to small amounts of the same compounds found in the 90% methanol fraction. Water solubles were inactive.

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THE FUNGUS FLORA OF HARDWOOD BARK COMPOSTS SUPPRESSIVE AND CONDUCTIVE TO RHIZOCTONIA DAMPING OFF. G. A. Kuter, E. B. Nelson and H. A. J. Hoftink, Dept. of Plant Pathology, Ohio Agric. Res. and Devel. Center, Wooster 44691.

Fungal populations were isolated from batches of hardwood bark compost of varying age and suppressiveness to Rhizoctonia damping off. Consistent differences were observed in the species composition of populations isolated from conducive vs. suppressive batches. *Trichoderma* spp. (*T. hamatum*, *T. harzianum*, and *T. koningii*) were among the predominant species in the mature (>23 wk old) suppressive composts but were absent or less abundant in the green (3 wk old) conducive composts. Furthermore, these *Trichoderma* spp. were not recovered from composts made conducive by heat (60C) treatment or from conducive peat. Details of the mycoflora isolated from conducive and suppressive composts will be presented.

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INDUCTION OF SUPPRESSIVENESS TO RHIZOCTONIA DAMPING-OFF BY SPECIES OF TRICHODERMA AND GLIOCLADIUM VIRENS FROM COMPOSTED HARDWOOD BARK. E. B. Nelson, G. A. Kuter, and H. A. J. Hoftink, Department of Plant Pathology, Ohio Agricultural Res. and Devel. Center, Wooster 44691 and The Ohio State University.

Fungal isolates from composted hardwood bark were tested for their ability to suppress Rhizoctonia damping-off. Spore suspensions of selected isolates were added to hardwood bark composts made conducive by heat-treatment (60C). Composts were infested with *R. solani* and percentage disease determined after 7 days using a radish seedling assay. Isolates of *Trichoderma* spp. (*T. hamatum*, *T. koningii*, and *T. harzianum*) and *Gliocladium virens* from batches of suppressive mature (>23 wk old) composts were most effective in suppressing damping-off. Fungal isolates from green (3 wk old) conducive and mature conducive composts did not induce suppressiveness. Rhizoctonia populations declined more rapidly in suppressive mature composts and composts amended with *Trichoderma* spp. than in conducive composts. *Trichoderma* spp. and *G. virens* were among the most abundant fungi recovered from *R. solani* propagules incubated in suppressive composts.

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CUNNINGHAMELLA ELEGANS LEDNER, AN ANTAGONIST TO RHIZOCTONIA SOLANI KUHN. M. M. Joshi, G. R. Goss, and S. N. Hillebrener. Kalo Ag Chemicals, Inc., 525 Kentucky Street, Quincy, IL 62301.

Cunninghamella elegans Ledner (CE) was isolated from *Rhizoctonia solani* Kuhn (RS) sclerotia and mycelial sections after incubation in either Seaton-Urban silt loam or Wakeland sand for three weeks at 30C and -0.33 bars water potential. Antagonism to RS was monitored by observing seedling survival in white sand flats, seeded with Williams soybeans, uninoculated, or inoculated with either CE, RS or both. After planting, flats were incubated for one week under incandescent and fluorescent light at 32±5C and 95±5% relative humidity, at which time, the percentage of healthy emerged seedlings was determined. When flats were inoculated with CE either at planting or two days prior, significant control (p=0.05) of RS damping-off was not observed. Inoculation with CE four days prior to planting caused a significant increase in seedling survival over the RS treated flats. *C. elegans* appears to be antagonistic to *R. solani*, but to control damping-off, an established population of *C. elegans* must already be present.

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EFFECT OF SYNTHETIC IRON CHELATES ON POPULATION DENSITIES OF FUSARIUM OXYSPORUM AND PSEUDOMONAS PUTIDA IN SOIL. Marcella Dupler, Fran M. Scher, and Ralph Baker, Dept. of Botany and Plant Pathology, Colorado State Univ., Fort Collins, CO 80523.

Fusarium wilt incidence was reduced by addition of FeEDDHA and *Pseudomonas putida* to pathogen-infested soil. The mechanism of control was thought to be due to a reduction in iron available to the *Fusarium*. Addition of FeEDTA and FeEDDHA to *Fusarium oxysporum* f. sp. *conglutinans*-infested soil did not significantly affect population densities in host and non-host rhizo-

sphere soils. Addition of 100-1000µg FeEDDHA/g soil increased population densities of rifampin-resistant *P. putida* in fallow and rhizosphere soils; however, 100µg FeEDTA/g soil resulted in no increase. Stimulation of biological activity in soil by the addition of water, 1 wk prior to introduction of *P. putida*, decreased subsequent rhizosphere colonization when compared with equivalent bacterial additions to air-dry soil. FeEDDHA increased root colonization by *P. putida* under both conditions.

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SPREAD OF FUSARIUM OXYSPORUM F. SP. MELONIS IN FUMIGATED FIELD SOIL. J. J. Marois, M. T. Dunn, and G. C. Papavizas. 1st and 3rd authors: Soilborne Diseases Lab, USDA, Beltsville, MD 20705, 2nd author: Dept. of Botany, Univ. of Maryland, College Park, MD 20742.

A field with a history of Fusarium wilt of muskmelon was strip fumigated with 1,2-dichloropropane and 1,3-dichloropropene and other related compounds plus methyl isothiocyanate (Vorlex®) at 280 L/ha in 72 cm wide beds and mulched with clear plastic. Pathogen recolonization was monitored every 2 wk by taking nine soil cores at 8 cm intervals across the bed from edge to edge. Each core was divided at 5 cm increments to a depth of 25 cm and the pathogen population density of the sample determined by dilution plates and pathogenicity tests. The pathogen was established in the outer edges of the beds 32 days after fumigation. By 74 days after fumigation, the pathogen population density was higher in the deeper soils than in the shallow top 5 cm of soil. At 102 days the pathogen had reinvaded the fumigated soils completely. An experimental model was developed to simulate the spread of the pathogen through fumigated soils.

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BIOLOGICAL CONTROL OF FUSARIUM WILT OF CARNATION WITH DISEASE SUPPRESSIVE SOILS AND RHIZOBACTERIA. G. Y. Yuen, A. H. McCain and M. N. Schroth. Dept. Plant Pathology, University of California, Berkeley, CA 94720.

Suppressive soils reduced the severity of carnation wilt caused by *Fusarium oxysporum* f. sp. *dianthi* when added to pathogen-infested conducive soil at rates ranging from 1% w/w to 10% w/w. These treatments resulted in a decrease in disease severity averaging 30%. Dipping roots of carnation cuttings into suspensions of these suppressive soils prior to planting resulted in similar decreases. Bacteria isolated from the rhizosphere of carnations grown in one suppressive soil were effective in reducing wilt severity by more than 40% when applied as single strain suspensions to cuttings prior to planting. Wilt reduction by bacterial treatments remained effective up to 4 months, while treatments with suppressive soil suspensions were effective for over 7 months. The duration of wilt protection in each bacterial treatment corresponded to the time that the applied strain was detectable in the rhizosphere of treated plants.

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INHIBITION OF PHYTOPHTHORA CACTORUM BY BACTERIAL ISOLATES AND EFFECTS OF CHEMICAL FUNGICIDES ON THEIR GROWTH AND ANTAGONISM. R.S. Utkhede, Agriculture Canada, Research Station, Summerland, British Columbia VOH 1Z0

Five bacteria isolated from soil produced diffusible antibiotics on corn meal agar (CMA) which were antagonistic to the growth of *Phytophthora cactorum*, a causal agent of crown rot of apple trees. The growth of one bacterial isolate B8, showing the largest zone of inhibition on CMA, was not significantly affected by 50 and 100 ppm of mancozeb and 50, 100, 200, and 400 ppm of Aliette but showed significant increase in growth by 50 ppm of Ridomil. The mycelial growth of *P. cactorum* was completely inhibited on CMA containing 50 and 100 ppm by Ridomil, mancozeb or 600 ppm by Aliette. The growth and antagonistic ability of bacterial isolate B8 on unamended CMA was not significantly affected even after 4 weeks of growth on 50 and 100 ppm of Ridomil, mancozeb or 50 to 1000 ppm of Aliette. These results indicate that it may be possible to use this bacterial isolate individually or in combination with chemical fungicides for the control of crown rot of apple trees.