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Abstracts

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Alphabetized by first author's last name.

PRODUCTION OF MONOCLONAL ANTIBODIES TO *STREPTOMYCES SCABIES*. I. Arias and R. Loria. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Monoclonal antibodies were produced to homogenized and sonicated cells of *Streptomyces scabies*, causal agent of common scab of potatoes. Initial selection of hybridoma clones was based on positive reactions in indirect-ELISA with *S. scabies*. Polyclonal antibodies and pre-immune mouse serum were used as positive and negative controls. Clones with high affinity for *S. scabies* were recloned and tested for cross-reaction with *S. acidiscabies*, the acid scab pathogen, and several nonpathogenic *Streptomyces* strains. A clone with high affinity for *S. scabies* which did not cross-react with *S. acidiscabies* was identified. However, this clone did cross-react with several of the nonpathogenic strains tested. This information is consistent with morphological and physiological data which suggests that *S. scabies* is more closely related to several nonpathogenic species than to *S. acidiscabies*.

EVALUATION OF RED SPRUCE IN WEST VIRGINIA FOR NUTRIENT DEFICIENCIES. D. E. Audley, J. M. Skelly, and L. H. McCormick, The Pennsylvania State University, University Park, PA 16802, and W. A. Jackson, USDA-Forest Service, Asheville, NC 28802.

Nutrient deficiency has been implicated as a factor in spruce declines. A survey to evaluate nutrient deficiencies of red spruce (Picea rubens Sarg.) growing on the Monongahela National Forest was conducted during summer and fall of 1990. In forty stands, three plots per stand were selected and the three dominant or co-dominant trees closest to each plot center sampled. Total tree height, ht. of live crown, and dia. at breast ht., were measured. Increment cores were taken to determine tree age at breast height. Percent discoloration and defoliation, as observed from the ground, was estimated by 5% classes. Values ranged from 0-30% (median=5%) and 0-60% (median=10%) for discoloration and defoliation, respectively. Upper crown foliar samples were collected and needles from the 1988-1990 internodes were analyzed for nutrient condition. Soil samples were collected and analyzed for pH, CEC, and % base saturation of K, Mg, and Ca to assess their relationship to tree condition.

ROLE OF POLLINATION AND INOCULATION ON ERGOT DEVELOPMENT IN SORGHUM. R. Bandyopadhyay[1], L. K. Mughogho[2], and G. Tegegne[3]. [1]Department of Plant Pathology, Cornell University, Ithaca, NY 14853; [2]ICRISAT Patancheru P.O., A. P. 502 324, India; [3]IAR, Nazret, Ethiopia.

Ergot is a tissue replacement disease involving pistils of cereal crops and grasses. It is generally believed that pollinated flowers resist infection and are not colonized by the fungus. We studied ergot (*Sphacelia sorghi*) development in sorghum by inoculating flowers at different time intervals before or after pollination. Pollination and infection related events were monitored by fluorescence microscopy. When inoculated 1-4 d before pollination, 52-95% spikelets were infected, but only 4-10% spikelets had infection if inoculated 1-4 d after pollination. Pollination and fertilization reduced ergot because the fungus failed to grow or grew slowly in pistils inoculated after pollination. Similarly pollen tube growth was retarded in spikelets inoculated before pollination. About 2% spikelets had grains with stroma growing at their basal end. Such grains probably resulted from pistils in which infection hyphae and pollen tube were observed growing simultaneously, each succeeding in its respective function -- colonization and fertilization.

Partially purified protein extracts isolated from American and Chinese chestnut have antifungal properties against the chestnut blight pathogen *Cryphonectria parasitica*.
Janna L. Beckerman and William A. Powell, Dept. of Environmental and Forest Biology, SUNY College of Environmental Science and Forestry.

American and Chinese chestnut *(Castanea dentata and C. mollissima,* respectively) differ greatly in susceptibility to the chestnut blight fungus, *C. parasitica.* The poorly understood resistance mechanism operating in Chinese chestnut that inhibits the invasion of the fungus is lacking in the American chestnut. To determine differences in the defense response, we have isolated partially purified protein extracts from challenged and unchallenged woody tissues of both species. Utilizing SDS-PAGE, we detected increases of specific proteins in the extracts of the challenged tissue as compared to the unchallenged tissue. We have also found that protein extracts from unchallenged tissue from both species exhibit antifungal properties. We are continuing to look for differences in the role proteins play in the defense response, as well as looking for potential inhibitory proteins in other organisms which could be genetically engineered into the American chestnut.

FEEDING PREFERENCE OF THE ASTER LEAFHOPPER AS AFFECTED BY ACREMONIUM-INFECTED TURFGRASS SPECIES. R.J. Buckley and P.M. Halisky, Department of Plant Pathology, NJAES, Cook College, New Brunswick, NJ 08903.

Populations of aster leafhoppers, Macrosteles fascifrons, (ALH) were subjected to feeding trials on perennial ryegrass, tall fescue and Chewings fescue to determine feeding preference or deterrence. The plant samples consisted of genetically identical clones of these three grasses that, were either infected (EI) or free from infection (EF) by their respective endophyte, viz., A. lolii, A. coenophialum or A. typhinum. The results indicate that, when given a choice, ALH preferred to feed on EF tall fescue rather than on EI tall fescue. In contrast, ALH preferred to feed on EI rather than on EF Chewings fescue. Tests with perennial ryegrass showed no

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clear-cut preference for either EI or EF ryegrass plants. These results suggest that an antifeedant component is associated with A. coenophialum-infected tall fescue, and that it may be responsible for the feeding deterrence of M. fascifrons. Such components associated with A. lolii-infected perennial ryegrass and A. typhinum-infected Chewings fescue, if present, appear to have little or no effect on feeding by M. fascifrons.

DETECTION OF THE BACTERIALIKE ORGANISM (BLO) ASSOCIATED WITH CITRUS GREENING DISEASE (CG) USING BLO-SPECIFIC OLIGONUCLEOTIDE SEQUENCES FROM RIBOSOMAL RNA GENE. K.H. Chen and T.A.Chen, Department of Plant Pathology, Rutgers University, New Brunswick, N.J. 08903

The 16S ribosomal RNA gene (rDNA) from CG-BLO was selectively amplified via PCR using a set of synthetic oligonucleotides, Primer A and F', which are broadly homologous to conserved eubacterial 16S rDNA. Comparisons of 16S or 18S rDNA sequences of the BLO, plant chloroplast, plant mitochondria, and eubacteria were made with the aid of the Biovax program. Oligonucleotide sequences specific in BLO 16S rDNA were then chosen as primers for PCR to amplify and detect BLO, but not plant nucleic acids. Two oligonucleotides, oligo 1, a 21-base pair and oligo 2, a 20-base pair, were designed from the variable portion of the BLO rDNA. Using oligo1 and primer F', a 900-bp DNA fragment was amplified from crude extracted or purified DNA from BLO infected periwinkle plants. The same two primers have successfully amplified a DNA fragment of the expected size from several different BLO-DNA preparations. No PCR products could be demonstrated when DNA from healthy periwinkles and E. coli were used as templates.

DETERMINING VECTOR STATUS OF GREEN APPLE APHIDS FOR ERWINIA AMYLOVORA TO APPLE SHOOTS UNDER FIELD CONDITIONS. G. G. Clarke, K. D. Hickey, and J. W. Travis, Dept. of Plant Pathology, Pennsylvania State University, Univ. Park, PA, 16802.

Colonies of apterous green apple aphids, Aphis pomi, were established on shoots of 'Red Yorking'/MM111 apple to determine their ability to vector Erwinia amylovora under field conditions. Once aphids were established the colony-supporting shoots were syringe-inoculated with E. amylovora at $1x10^8$ cfu/ml and then caged together with noninoculated, aphid-free shoots. All inoculated shoots blighted within four days and aphids migrated to the noninoculated shoots. None of the noninoculated shoots developed blight, indicating that aphids may be unable to transmit sufficient quantities of bacteria from blighting shoots to healthy shoots by their feeding. Aphids fed a solution containing E. amylovora through Parafilm membranes were also unable to transmit blight to shoots in the field. A parallel syringe inoculation test indicated that between $1x10^3$ and $1x10^4$ cells were needed for establishment of typical shoot blight symptoms. In a separate test the presence of aphids did not result in an increased number of blighted shoots when shoots with and without active aphid colonies were atomized with a suspension of E. amylovora.

RFLP ANALYSES AND DOT HYBRIDIZATIONS OF CHROMOSOMAL DNA DISTINGUISH TWO MYCOPLASMALIKE ORGANISMS (MLOS) ASSOCIATED WITH GRAPEVINE YELLOWS DISEASE. R. E. Davis, E. L. Dally, A. Bertaccini[1], R. Credi[1], R. Osler[2], V. Savino[3], L. Carraro[2], B. Di Terlizzi[4], M. Barba[5], and I. M. Lee. Agricultural Research Service, USDA, Beltsville, MD; [1]Universita degli Studi, Bologna, Italy; [2]Universita di Udine, Italy; [3]Universita di Bari, Italy; [4]Istituto Agronomico Mediterraneo di Valenzano, Italy; [5]Istituto Sperimentale per la Patologia Vegetale, Rome, Italy.

DNA based methods were employed in a study of flavescence doree (FD), a yellows disease of grapevine. A MLO (FDB) associated with FD in southern Italy and a FD MLO (FDU) from northern Italy were distinguished from one another on the basis of results from restriction fragment length polymorphism (RFLP) analyses and dot hybridizations using cloned DNA from FDU and other MLOs. The findings indicate that at least two genetically different MLOs are associated with this grapevine yellows disease.

IN VITRO PRODUCTION OF THE PHYTOTOXIN THAXTOMIN A BY *STREPTOMYCES SCABIES* AND *S. ACIDISCABIES*. L. M. Delserone, R. A. Bukhalid, and R. Loria, Cornell University, Dept. of Plant Pathology, Ithaca, NY 14853

Thaxtomin A is a phytotoxin produced by *Streptomyces scabies* and *S. acidiscabies*. Where purified toxin is applied to immature tubers, initial symptoms of potato scab are produced, suggesting that thaxtomin A may be a pathogenicity determinant. Thaxtomin production *in vitro* has not been reported. We tested three strains each of *S. scabies* and *S. acidiscabies* for production of thaxtomins in a variety of liquid media. Thaxtomin was detected and identified after three days growth by thin-layer chromatography, spectrophotometry and bioassays of culture filtrate extracts. All pathogenic strains of *S. scabies* and *S. acidiscabies* produced thaxtomin in culture, while three nonpathogenic strains did not. The data indicate that thaxtomin production is not host-induced and is correlated with pathogenicity.

JOINT ACTION EFFECTS OF FUNGICIDES USED FOR THE CONTROL OF BOTRYTIS LEAF BLIGHT OF ONION IN NEW YORK. M. S. DeMilia and J. W. Lorbeer, Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Mixtures of mancozeb with iprodione, vinclozolin or chlorothalonil, fungicides used for control of Botrytis leaf blight of onion in NY, were analyzed for joint action effects. Six spray trials were conducted on commercial onion fields during 1989 and 1990. Each trial included 25 treatments comprising combinations of five levels of two fungicides. Fungicide joint action effects were determined by methods based on the multiplicative survival model and the additive dose model (Weed Sci. 26:58-71. 1978), and on regression analysis. Mancozeb and iprodione exhibited synergistic joint action effects, while mancozeb and vinclozolin exhibited additive joint action effects. Mancozeb and chlorothalonil exhibited additive joint action effects except with respect to harvest yield, which exhibited antagonistic joint action effects as a result of yield inhibition at high (but not low or intermediate) dosage levels of chlorothalonil.

PARTIAL CHARACTERIZATION OF A PHYTOTOXIC COMPOUND PRODUCED BY *PYTHIUM ULTIMUM*. H. Desilets, P. Blais, and R. R. Bélanger. Département de phytologie, Université Laval, Québec, Qc G1K 7P4.

In a previous experiment, it was demonstrated that culture filtrates of the non-specific fungus *Pythium ultimum* could induce typical infection symptoms on geranium (Desilets and Bélanger, 1991). This study was thus initiated in an attempt to characterize the active phytotoxic compound(s). Fractions were obtained from *Pythium* culture filtrates by sequential filtration on Sephadex G-75 and G-25 columns and were assayed on geranium tissue-cultured plantlets and seedlings. Of all the isolated fractions, one was found to induce typical symptoms of root necrosis and root hair breakdown on geranium. In vitro plantlets also suffered a significant growth reduction on both shoots and roots when exposed to the isolated fraction. The putative toxic compound was heat stable, water soluble and of low molecular weight (150-300 d). Dilution of the fraction (1:5) induced an apparent auxinic effect on the growth of the plantlets, which was characterized by an abundant production of roots. However, these roots were thin and lacked root hairs.

INCREASES IN ROOT COLONIZATION BY RHIZOBACTERIA AND ASPARAGUS YIELD FOLLOWING NACL APPLICATIONS W. H. Elmer, Dept. of Plant Pathology and Ecology, The Connecticut Agricultural Experiment Station, Box 1106, New Haven, CT 06504.

Granular applications of NaCl (560 kg/ha) were applied in April to one half of experimental asparagus plots in Hamden, Madison, and Windsor, CT that were affected by Fusarium crown and root rot. All plots received normal fertilizer. Spears were harvested, trimmed, and weighed every 2-3 days for 1-3 weeks depending on the age of the planting. Soil cores were taken 15 cm away from the crown to a depth of 22.5 cm before and after NaCl treatment. Roots were washed and surface-disinfested, and 0.5-1.0 g were agitated in saline buffer. The buffer was then serially diluted onto neutral PDA. The mean colony-forming-units of bacteria/g root (CFU) increased between sampling in all plots in each field, but the NaCl-treated roots had 3.9, 5.6, and 4.1 times more CFU than nontreated roots in the three plantings. The majority (67-95%) of the strains were fluorescent pseudomonads. Applications of NaCl significantly increased both yield and CFU when compared to untreated plants, and there were positive correlations between CFU and yield. Increased CFU by NaCl may contribute to the suppression of Fusarium crown and root rot of asparagus.

LOCALIZATION OF FREE AND CONJUGATED FORMS OF SALICYLIC ACID IN LEAVES OF TMV-INOCULATED TOBACCO. Alex J. Enyedi, Nasser Yalpani and Ilya Raskin, Center for Agricultural Molecular Biology Rutgers University, New Brunswick, NJ, 08903-0231

The incompatible reaction between tobacco (cv. Xanthi-nc NN) and TMV triggers the *de novo* synthesis of salicylic acid (SA) and a hypersensitive response. Elevated levels of SA also elicit the accumulation of pathogenesis-related proteins and possibly induce systemic acquired resistance. Tobacco leaves typically contain 0.05 µg/g fresh weight SA which increases to 20 µg/g SA six days after TMV inoculation. SA is preferentially synthesized in and around lesions. The highest concentration of SA is consistently detected within 3.5 mm of the center of a lesion. In contrast, the concentration of free SA is 27-fold less in tissue located 6 to 10 mm away from the lesion. Chemical and enzymatic hydrolysis of TMV-infected leaf extracts revealed the presence of SA conjugates. ß-Glucosidase treatment suggests the conjugate is ß-*O*-D-glucosylsalicylic acid. The SA conjugate is detected only in leaves which contain local lesions. No SA conjugate is found in phloem exudates or in uninfected leaves of TMV-inoculated plants. The conjugation of SA may represent one mechanism which regulates the level of free SA in the plant.

PRESENCE OF ALGINATE GENE SEQUENCES AMONG MEMBERS OF THE PSEUDOMONAS rRNA HOMOLOGY GROUPS I-IV. W. F. Fett, C. Wijey and E. R. Lifson; USDA, ARS, Eastern Regional Research Center, Philadelphia, PA 19118

Total genomic DNA of thirteen bacterial strains representing pseudomonad rRNA homology groups I-IV were screened for sequences homologous to four Pseudomonas aeruginosa alginate (alg) genes by Southern hybridization. Biotinylated probes for three structural genes (algA, algC and algD) and one regulatory gene (algRl) were prepared. Genomic DNA of strains representing group I (P. syringae pv. glycinea, P. viridiflava and P. corrugata) strongly hybridized with all four gene probes. P. corrugata has not been reported to be capable of alginate production. Genomic DNA from the representatives of groups II-IV gave no or very weak hybridization with the probes except for algC. This study suggests that the ability to produce alginate among the pseudomonads is restricted to members of rRNA homology group I in agreement with earlier physiological studies.

REACTION OF BEAN LINES TO *MELOIDOGYNE INCOGNITA* AND *FUSARIUM OXYSPORUM* F. SP. *PHASEOLI*. R. A. France and G. S. Abawi. Dept. of Plant Pathology, NYSAES, Cornell University, Geneva, NY 14456

The reaction of 16 lines to the Fusarium wilt pathogen was determined by utilizing 15-day-old bean seedlings grown in sterilized sand. The distal 1 cm of root tips were cut off and the roots were dipped for 5 min in a suspension of 10^6 conidia/ml of the pathogen. Seedlings were transplanted into pots filled with pasteurized soil. Severity of wilting was recorded weekly; vascular discoloration and dry weight were determined 6 wk after transplanting. Reaction of the same lines to the root-knot nematode was determined by inoculating bean seeds (3 seeds/10 cm pot) with 10,000 eggs placed around the seeds in pasteurized soil. Root galling, eggs/root system, and dry weight were determined after 8 wk. Nemasnap and line A 107 were resistant to both pathogens, whereas IPA 1, Ica Pijao, Labrador, and Isabella were susceptible to both pathogens. The other 10 lines evaluated were resistant to only one of the two pathogens.

CONTROL OF FUNGAL DISEASES OF GRAPEVINE BY SHORT-WAVE ULTRAVIOLET LIGHT. David M. Gadoury, R.C. Pearson, R.C. Seem, T. Henick-Kling*, L.L. Creasy**, and A. Michaloski, Departments of Plant Pathology, Food Science and Technology*, and Fruit and Vegetable Science**, Cornell University, Geneva, NY 14456.

Short-wave ultraviolet light (UV) from germicidal fluorescent lamps killed hyphae within powdery mildew (PM) colonies on grapevine seedlings at doses below those that severely injured plant tissue. Exposure of conidia of *Uncinula necator* and sporangia of the downy mildew pathogen (DM), *Plasmopara viticola*, to UV doses above 100 mWs/cm^2 inhibited germination of these structures, but did not prevent resporulation of *P. viticola*. A field unit bearing 48 UV lamps arranged in two banks was used to treat a vineyard of cv Catawba in 1990, and vineyards of cv Delaware, Chancellor, and Rosette in 1991. In 1990, weekly exposure of vines to 200 mWs/cm^2 reduced foliar PM severity from 32% to 6.5%, and the fruit DM incidence from 10% to 0. Similar reduction of fruit and foliar PM, but not DM, was observed in 1991. Some berry russet was observed on Chancellor and Rosette, but not on Delaware or Catawba. Net photosynthesis was unaffected by the UV doses used in our study. Levels of the phytoalexin resveratrol were not related to disease control in UV-treated vines. The mechanism of action of UV appears to be direct damage to exposed propagules and mycelia.

EVALUATION OF TWO METHODS FOR ASSESSMENT OF FIELD RESISTANCE TO POTATO LEAFROLL VIRUS. L. Greenspan Gallo, S. A. Slack, and R. Loria. Plant Pathology Department, Cornell University, Ithaca, NY 14853.

Ten potato cultivars (cv) known to differ in levels of PLRV resistance were evaluated under conditions of uniform inoculum pressure. Treatments (T) contained 11 plants with T1 having 5 cv plants either side of a central PLRV-infected Russet Burbank and with T2 having 11 cv plants covered by a floating row cover (Agryl P10) to exclude aphids. In T1 natural aphid colonization was permitted and in T2 viruliferous aphids were introduced. Survey results showed aphid populations to be uniformly distributed among cv in T1, but not in T2 due to inadvertent natural colonization. Disease incidence, determined on plants from tubers harvested from T1, ranged from 0-93% and was consistent with expected field resistances. Significant cv. groupings were (most susceptible to most resistant): Russet Burbank, Sebago, B 9922-11 > Katahdin, Red Pontiac, F 100-1, Irish Cobbler, Norland > Abnaki, BelRus. Agreement between visual assessment and ELISA on plants from harvested tubers was ≥90% except for Irish Cobbler, BelRus and Norland (40-60%).

EFFECT OF ADJUVANTS ON SURVIVAL OF AN ANTAGONISTIC STRAIN OF *Pseudomonas putida* (PSU831) ON ALFALFA LEAVES. Y. Guevara, F. Lukezic, and K.T. Leath. Department of Plant Pathology, The Pennsylvania State University, University Park, Pa 16802.

Different adjuvants (nutrients, U.V.light protectants, osmoprotectants and wetting agents) were added to bacterial suspensions that were applied by immersion or droplet techniques onto alfalfa plants of different cultivars. After treatment the plants were kept in a dew chamber for 48 h, and then transferred to greenhouse conditions. Samples were taken at selected time intervals. Leaflets were shaken for 1/2 h in a washing buffer, serial dilutions were made and plated on a selective medium containing 300 ppm Rifampicin in order to recover the labeled antagonist PSU831. After incubation, colony forming units were counted. All data were transformed logarithmically and were subjected to analysis of variance using the SAS computer package. Bacterial survival was enhanced with the addition of nutrients (King's B broth), osmoprotectants (betaine and inositol) and U.V. light protectants (folic acid and congo red). The best combination was King's B broth + 10g dextrose/l +betaine (0.35 g/l) + folic acid (0.5g/l). A bioassay was conducted in order to determine the effect of the treatments on the antagonistic ability of PSU831 against Phoma sp.

CLONING OF CHROMOSOMAL DNA OF THE MYCOPLASMALIKE ORGANISM (MLO) ASSOCIATED WITH GRAPEVINE YELLOWS. J. R. Guo, T. A. Chen, N. Loi and R. C. Pearson[1]. Dept. Pl. Path., Rutgers Univ., New Brunswick, NJ 08903 and [1]Dept. Pl. Path., Cornell Univ., NYSAES, Geneva, NY 14456.

Chromosomal DNA of the MLO associated with grapevine yellows (GY) was extracted from infected periwinkle plants by CsCl buoyant density gradient centrifugation. GY-MLO DNA was digested with EcoR I restriction enzyme. The fragments were ligated with EcoR I digested pUC19 and transformed into E. coli DH5α cells. Two recombinant plasmids specific to GY-MLO that contained inserts of 9.0 and 1.6 kb were selected. Using a dot blot assay, recombinant plasmids labeled with ^{32}P-dCTP hybridized with total DNAs from periwinkle plants infected with GY-MLO and not with healthy or diseased plants infected with 6 other MLO diseases. The cloned DNA probes could detect as little as 10 ng total DNAs from infected periwinkle plants and grapevine samples showing grape yellows symptoms collected from Geneva, New York.

RAPID DETERMINATION OF RESTRICTION FRAGMENT LENGTH POLYMORPHISMS (RFLP) WITHIN THE MITOCHONDRIAL AND NUCLEAR GENOMES OF FUSION PROGENY IN *TRICHODERMA HARZIANUM*. Chris Hayes, Gary Harman, Thomas Stasz, Sheri Woo, Matteo Lorito, Antonio Di Pietro, Department of Horticultural Sciences, Cornell University/NYSAES, Geneva, NY 14456

Certain strains of *Trichoderma harzianum* have been identified as potential biocontrol agents against a variety of plant pathogenic fungi. Alone, these few strains do not possess the ability to be effective biocontrol agents. By combining the beneficial traits of these strains, a superior biocontrol strain could be formed that would be as efficient as chemical fungicides. Since most of these biocontrol strains are asexual, it would be difficult to combine beneficial traits of several strains. Hence, protoplast fusion has been used to combine the nuclei of different strains in an attempt to isolate recombinant strains that contain the biocontrol attributes of both parents. However, during protoplast fusion the mitochondrial genomes, in addition to the nuclear genomes, may also fuse. Monitoring the genetic elements obtained in both genomes has been possible by purifying the genomes to homogeneity on a CsCl density gradient. A more rapid approach has been devised that allows for identification of parental origin of both the mitochondrial and nuclear genomes within a total DNA extract utilizing RFLP.

KARYOTYPING OF *TRICHODERMA HARZIANUM* UTILIZING TRANSVERSE ALTERNATING FIELD ELECTROPHORESIS (TAFE). Chris Hayes, Gary Harman, Sheri Woo, Maria L. Gullino, Department of Horticultural Sciences, Cornell

University/NYSAES, Geneva, NY 14456

Recent techniques have been devised that allow the resolution of intact chromosomes. This new technology has been employed in an attempt to electrophoretically karyotype two strains of *Trichoderma harzianum* and a progeny formed by protoplast fusion of the two strains. Intact chromosomes were obtained by embedding viable protoplasts of parental strains, T12 and T95, and a fusion strain, 1295-22, into agar blocks. The protoplasts were lysed within the agar blocks, which greatly reduced shearing of the chromosomes. Three diverse run parameters were identified that allowed for visualization of chromosomes ranging in size from 100 Kilobase pairs (Kb) up to 6.0 Megabase pairs (Mb). The parental strains differed in the number and size of chromosomes. The fusion progeny had a karyotype that was similar to parental strain T12, with chromosomes of all strains ranging in size from 2.0 to 6.0 Mb.

CHLOROTHALONIL EFFICACY REDUCED BY SIMULTANEOUS APPLICATION OF SAFER® INSECTICIDE CONCENTRATE. H. Hill, School of Forest Resources, and N. G. Wenner, and W. Merrill, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802

Flowable chlorothalonil formulations are widely used to control needlecasts of Scots pine because of their 4-6 week residual activity. With increasing interest in the use of integrated pesticide applications and bio-rational insecticides, the object of this study was to determine the effect of applying insecticidal soap with the Bravo® 720 chlorothalonil formulation. Bravo® 720 was applied at 3.2 l f.p./ha with and without Safer® insecticide concentrate at 18.75 l f.p./ha to Scots pine Christmas trees during the spring infection period for Cyclaneusma needlecast. Efficacy was determined by direct isolation of C. minus from the needles of the 1990 complement (400 needle isolations per treatment per sample date) prior to treatment on 12 April 1991 and at weekly intervals thereafter for 6 weeks. Despite abnormally low rainfall during this period, simultaneous application of the two compounds significantly reduced the efficacy of the fungicide ($P < 0.0005$).

REACTIONS OF WINTER WHEAT CULTIVARS TO BARLEY YELLOW DWARF VIRUS (BYDV) INFECTION. T. K. Hoffman, M. L. Risius, and F. E. Gildow, Dept. of Agronomy and Dept. of Plant Pathology, Pennsylvania State University, University Park, PA 16802.

Field plots of five winter wheat cultivars, Titan, Fillmore, FL 302, Tyler, and 2555 were inoculated with the PAV isolate of BYDV to evaluate effects on components of yield and on virus replication. Plots, established in Centre County, PA, were infested in the fall of 1988 and 1989 when plants were at the four leaf stage. Grain yield reductions attributed to BYDV-PAV were significant for all cultivars in both years, and cultivars were ranked as listed above. Titan exhibited the greatest percentage yield reduction (67 %) and 2555 the least (27.5 %). Other significant changes in BYDV infected plants included increases in spikes per meter of row, reductions in the number of kernels per head and reductions of kernel weight. Increased levels of virus in leaf tissue were correlated with larger yield reductions. Data suggest that among the cultivars tested tolerance is not very strong, but may be associated with some resistance to virus replication.

IMPORTANCE OF BUTTRESS ROOT AND TAPHOLE WOUNDS AS INFECTION COURTS FOR THE SUGAR MAPLE (ACER SACCHARUM) SAPSTREAK PATHOGEN, CERATOCYSTIS COERULESCENS. David R. Houston, USDA Forest Service, 51 Mill Pond Road, Hamden, CT 06514.

In spring of 1984, groups of 10 maple trees were wounded and inoculated with C. coerulescens (Cc). Wounds were (A) 2 tapholes, 1 open, 1 with metal spout; (B) 2 tapholes with plastic tubing; and (C) 2 buttress root bruises. Tapholes (A) were inoculated once, when made (3/13 or 4/17) or at weekly intervals until 5/31, or (B) on 5/2 or 5/31 when spouts were pulled. Bruises were inoculated once when made (weekly, from 3/13 to 6/15). Trees (5 per group) were cut in 1989 or 1990. Columns of discoloration about (A) tapholes were significantly longer than (B); (A) tapholes made 3/13 had longer columns than those made 4/17. Inoculation with Cc did not affect column length. Only 1 of 50 (A) and 1 of 22 (B) inoculated trees, and 1 of 20 (A) control trees had stain patterns suggestive of sapstreak and these were strongly limited. In contrast, some tree groups with inoculated buttress wounds were severely infected: 3 of 5 (5/2), 4 of 5 (5/31) and 1 of 5 (6/15).

THE VICTORIN RECEPTOR SITE IN OATS: 1. AUTORADIOGRAPHICAL LOCALIZATION OF A VICTORIN C DERIVATIVE. H. W. Israel, V. Macko, and T. J. Wolpert, Department of Plant Pathology and Boyce Thompson Institute for Plant Research, Cornell University, Ithaca, NY 14853.

In search of the putative victorin receptor site, we incubated seedling leaf pieces of victorin-susceptible and -resistant oats in a ^{125}I labeled Bolton-Hunter derivative of victorin C, processed these for high-resolution autoradiography, and analyzed them for cytological distribution of radioactivity. Susceptible oats bound three times more toxin than the resistant, the epidermis/mesophyll victorin binding ratio in the susceptible line was twice that of the resistant, and the toxin bound preferentially by six-fold to cell walls of both lines. Unexpectedly, vascular tissues of both oats were found to bind more than ten times the victorin bound by all other tissues combined. These results, which thus far have not qualified specificity competitively, indicate that cell walls and several other organelles of most oat leaf tissues contribute to victorin binding and processing in complex ways yet to be discovered.

THE VICTORIN RECEPTOR SITE IN OATS: 2. IMMUNOCYTOCHEMICAL LOCALIZATION OF A VICTORIN BINDING PROTEIN. H. W. Israel, V. Macko, and T. J. Wolpert, Department of Plant Pathology and Boyce Thompson Institute for Plant Research, Cornell University, Ithaca, NY 14853.

High-resolution immunocytochemistry with protein A-gold complex-labeled antibody (I_gG), raised against a 100 kd protein from oats that binds the ^{125}I Bolton-Hunter derivative of victorin C, was used to localize the toxin receptor site in seedling leaves. Labeling was uniform and exclusive to mesophyll mitochondria of both Victoria blight-susceptible and -resistant oats with and without toxin treatment. Epidermal and vascular mitochondria were unlabeled. These results, coupled with those given in the preceding abstract (1. Autoradiographical Localization of a Victorin C Derivative), foreshadow a complex mechanism(s) of both disease development and host susceptibility that will require extensive additional molecular-cytological work for resolution.

Infection of conifer seedlings with tomato mosaic virus. V. Jacobi and J.D. Castello. SUNY, College of Environmental Science & Forestry, 1 Forestry Drive, Syracuse, NY 13210.

Tomato mosaic virus (ToMV) has been detected in water and red spruce on Whiteface Mt., NY. To prove the infectivity of ToMV, a water isolate was purified and used to inoculate the roots of potted red and black spruce and balsam fir seedlings obtained from the nursery and placed in a coldframe. Needles and roots of inoculated and noninoculated control seedlings were tested by enzyme-linked immunosorbent assay (ELISA) and immunoelectron microscopy (IEM) for ToMV 12 and 18 mo post inoculation (PI). ToMV was detected in the roots of inoculated seedlings 12 mo PI and in the roots of both inoculated and control seedlings 18 mo PI. Because ToMV was not detected in 160 additional control seedlings, mulch, and soil from the nursery, the infection of control seedlings most likely resulted from virus movement from inoculated seedlings in the coldframe.

EVALUATING PERCENT WEIGHT LOSS FOR MEASURING ROOT COLD HARDINESS OF CONTAINER-GROWN BLACK SPRUCE. J.H. Jian & W.H. Livingston, Dept. of Forest Biol., Univ. of Maine, Orono, ME 04469.

Measuring root cold hardiness is essential for assessing a seedling's resistance to cold temperature damage. Percent weight loss (PWL) and a triphenyltetrazolium chloride(TTC) method were evaluated as quick measures of root cold hardiness for container-grown black spruce. After 1-yr old seedlings (360) were exposed to 5, -5, -15, -25 C for 3 hr, corresponding PWL values were 7.02, 7.19, 12.99, and 17.94, while absorbance at 475 nm after TTC incubation was 5.07, 5.57, 2.89, and 1.30. PWL and TTC methods detect freezing damage in roots similarly ($P<0.01$), and the results were consistent with seedling new-root-numbers after 3 wk incubation in the greenhouse. The PWL method was the quickest and easiest to use. Root washing prior to freezing was used to decrease the time and space needed for freezing seedlings (630 seedlings used). PWL of washed roots was highly correlated with seedling new-root-numbers in the greenhouse for both washed roots ($r=-0.873$) and roots in a soil plug ($r=-0.780$). The PWL method can be used to quickly evaluate the root cold hardiness of black spruce seedlings.

INFLUENCE OF SOIL FUMIGATION AND METALAXYL ON THE INCIDENCE OF PYTHIUM BLIGHT ON BELL PEPPERS. S.A. Johnston, Rutgers University, Bridgeton, NJ 08302

The experiment was conducted in a field continuously cropped to peppers and arranged in a split plot design with 4 replications. Whole plots included soil fumigants, either applied through drip irrigation or chisel injection, and a surfactant transplant water solution. Half of each plot received metalaxyl (0.1 kg/m^2) drenched onto seedlings prior to transplanting and through drip irrigation (1.12 kg/ha) 13, 46, and 75 days after transplanting. Pythium blight (P. aphanidermatum) appeared 10 days after transplanting and resulted in plant death. Soil fumigation resulted in a significant reduction in disease incidence and the surfactant treatment resulted in greater disease incidence than the nontreated check. A significant interaction occurred between the whole plots and subplots. For each of the soil fumigants (Telone C-17, Vapam, and Vorlex), the addition of metalaxyl did not decrease disease incidence, however, metalaxyl did result in less disease in the surfactant and nontreated check plots compared to the nonmetalaxyl treated subplot.

ESTIMATION OF CLOVER ROOT CURCULIO INJURY IN NEW YORK ALFALFA FIELDS AND CORRELATION WITH FUNGAL ROOT AND CROWN ROT. D. W. Kalb and G. C. Bergstrom, Dept. of Plant Pathology, Cornell University, Ithaca, NY 14853.

Sixty-one randomly selected alfalfa fields in five regions of New York were surveyed for injury by the clover root curculio (CRC) (*Sitona hispidulus*) and fungi associated with root and crown rot. Roots were rated for deep feeding wounds (DFW) and surface feeding caused by CRC, then split longitudinally and rated for root and crown rot. The number of DFW per root (tap and main secondary) ranged from 0 to 45; means for 2-, 3-, and 4-yr roots were 2.6, 5.5, and 6.1 DFW, respectively. Levels of CRC injury and root and crown rot were lowest in two northern counties. DFW were positively correlated with root and crown rot (R = 0.67). Fungi were isolated from randomly chosen roots and identified. Fusarium spp. accounted for 60% of the isolations: *F. oxysporum* (25%), *F. solani* (21%), *F. avenaceum* (4%), other (3%), nonidentified (7%). Other potential pathogens included species of *Phoma* (12%), *Stemphyllium* (2%), *Thielaviopsis* (2%), and *Pythium* or *Phytophthora* (2%).

RELATIONSHIP BETWEEN FOLIAR ELEMENTS AND SYMPTOMS ON NORWAY SPRUCE. J. Ke and J. M. Skelly, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802

Surveys in 1987 to 1990 of Norway spruce within 12 plantations across 4 NE states found symptoms of yellowing on a site-specific basis. Foliar N, K, and Ca concentrations of most of the sampled trees were above deficiency ranges, while foliar Mg concentrations of most symptomatic trees were below the deficiency range. Although knowledge of nutrient deficiency ranges may help diagnose foliar symptoms, their exclusive use may overly simplify relationships between foliar symptoms and concentrations of foliar elements. Principal component regression of the data provided assessment of interactions and balances among foliar elements and possible influences on crown discoloration and defoliation. Crown discoloration and defoliation were not only associated with foliar concentrations of individual elements, such as Mg, Ca, K, Al, Mn, B, Pb, Sr, N, P, Fe, Zn, and Cu, but also associated with interactions and balances between and among these elements.

RELATIONSHIP BETWEEN SOIL ELEMENTS AND FOLIAR SYMPTOMS ON NORWAY SPRUCE. J. Ke and J. M. Skelly, Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802

As part of a study to determine etiology of symptoms observed on Norway spruce surveyed in 1987 to 1990 in 12 plantations across 4 NE states, soil pH, exchangeable Mg, K, Ca, and corresponding percent saturations in soils were lower, while soil Al concentrations were higher for most of the symptomatic trees in comparison to the healthy trees. Foliar concentrations of Mg, Ca, K, P, Al, and Fe were positively correlated with concentrations of corresponding soil elements. Principal component regression of the data provided a comprehensive assessment of soil elements and their molar ratios and related influences on crown discoloration and defoliation. Molar ratios between soil elements, such as Mg/Al, Ca/Al, K/Al, K/Mg, and Mn/Mg were indicators for discoloration and defoliation.

EFFECTS OF OZONE ON THE VEGETATION OF ACADIA NATIONAL PARK Paul S. King, Robert Kohut, and John Laurence, Boyce Thompson Institute for Plant Research, Tower Road, Ithaca, NY 14853

Summer ambient ozone (O3) concentrations at Acadia National Park in Maine often exceed the national air quality standard of 120 ppb, prompting concern over possible effects on plant communities within the park. In 1990 a 3-year research project was initiated by the National Park Service in an effort to evaluate the response of native plants to O3. In the first year, 14 species were exposed in open-top chambers to one of four levels of O3 (charcoal-filtered, non-filtered, and 1.5 or 2.0 times ambient). Exposures occurred 12 hours daily for 101 days. Extent of foliar injury and rates of photosynthesis were determined monthly. Big leaf aster, gray birch, jack pine and bunchberry developed foliar O3 injury at or slightly above ambient concentrations with significant injury present at higher levels. The higher O3 treatments significantly decreased rates of photosynthesis for aster and gray birch and tended to decrease rates in paper birch and red oak.

VIABILITY OF EGGS AND INFECTIVITY OF VERMIFORM STAGES OF *PRATYLENCHUS PENETRANS* FROM SELECTED MARIGOLDS. M. Ko and M. Arevalo-Guerra, Department of Plant Pathology, University of Hawaii, Honolulu, HI 96822 and Cornell University, Ithaca, NY 14850.

Viability (Vi) of eggs and infectivity (Inf) of vermiform stages (rVS) of *P. penetrans* (Pp) recovered from *Tagetes erecta* cv. 'Pumpkin Crush' (PC) and 'Gold Galore' (GG) were investigated. Those from Wando pea served as control. Seedlings were inoculated with 10,000 Pp vermiform stages. Eggs or rVS were subsequently extracted from roots at selected intervals for 3 or 5 weeks, respectively. Inf of rVS was determined by a 10-day bioassay with 6-day-old pea seedlings as % initial inoculum of rVS remaining in their roots, and Vi of eggs as % hatch in water after 12 days. Inf of rVS declined with prolonged residence inside all roots. Inf of rVS from GG during the 5 week period ranged from 25% to 100% of those from PC or 14% to 33% of those from pea. Few eggs were recovered from GG and none hatched. Viabilities of eggs from PC decreased from 40% at day 10 to 8% at day 20, but those from pea were 57% at the same times. Direct exposure of the eggs/rVS to root extracts of the marigold cultivars during the same period resulted in 100% mortality within 12 hours.

PENETRATION AND REPRODUCTION OF *ROTYLENCHULUS RENIFORMIS* AS INFLUENCED BY SOIL TEXTURE AND PARTICLE SIZE. M. Ko and D. P. Schmitt, Department of Plant Pathology, University of Hawaii, Honolulu, HI 96822.

Reproduction of *Rotylenchulus reniformis* was investigated under greenhouse conditions in: (1) soil textures ranging from silty clay to 100% sand of 500 μm particle size, and (2) silica sand ranging from 180 μm to 1200 μm in particle size. Tomato or cowpea seedlings were inoculated with a total of 2,000 eggs plus vermiform stages. Number of nematodes penetrating the roots at day 10 was highest in silty clay soil and lowest in sand with particle size of 500 μm or larger (coarse sands). Nematode population density at week 6 was also highest in silty clay soil and in fine sands with particle sizes of 280 μm or smaller. Roots from silty clay soil and fine sands harbored more kidney-shaped females and also 25 times as many eggs as those from coarse sands. However, numbers of eggs produced per female from all soil textures or particle sizes were not significantly different. Thus, the low nematode population densities in coarse textured soils or sands may in part be attributed to few initial root penetrations by this nematode.

ANALYSIS OF GENETIC DIVERSITY WITHIN VEGETATIVE COMPATIBILITY GROUPS OF *CRYPHONECTRIA PARASITICA* USING DNA FINGERPRINTING. Y.-C. Liu and M. G. Milgroom. Dept. of Plant Pathology, Cornell University, Ithaca, NY 14853.

DNA fingerprinting was used to analyze the genetic variability within and between v-c groups from populations of *Cryphonectria parasitica* on chestnut. The proportions of bands shared within v-c groups from Michigan varied approximately from 0.7 to 1.0 with an average close to 0.9. In West Virginia the proportions ranged from 0.3 to 0.9 with an average of approximately 0.6. The proportion was not different within and between v-c groups in the West Virginia population. These results show that DNA fingerprints identify clonal lineages within v-c types in Michigan where *C. parasitica* populations are thought to be clonal. In contrast, there was very little correlation between fingerprints and v-c groups in the West Virginia population where sexual reproduction is important.

IN VITRO BIOLOGICAL EFFECTS OF *TRICHODERMA* CHITINASES ON DIFFERENT PLANT PATHOGENIC FUNGI. Matteo Lorito, Gary Harman, Antonio Di Pietro, Chris Hayes, Dept. of Hort. Sci., Cornell University/NYSAES, Geneva, NY, 14456.

Trichoderma is well known for is ability to control plant pathogenic fungi and many authors have suggested that chitinases play a role in this biocontrol ability. Chitinases degrade polymeric chitin and therefore should weaken the cell walls of chitin-containing fungi thereby inhibiting mycelial growth and/or spore germination. We purified an endochitinase from culture filtrate of *T. harzianum* and obtained another fraction that contained primarily chitobiase along with a small amount of N-acetylglucosaminidase and one other protein. Effects of these enzymes on *Fusarium* spp., *Botrytis sp., Ustilago sp.* and *Pythium sp.* were examined using a microscopical assay in which spores of the target fungi were incubated in PDB and exposed to different enzyme concentrations. Inhibition of spore germination was detected for *Botrytis sp.* and *Fusarium* spp. Moreover, cell replication appeared to be significantly reduced in *Ustilago* sp. while, as expected, the enzyme solution had no effect on spores and mycelium of the Oomycete *Pythium sp.* The different chitinases solutions showed differential levels and types of antifungal activity.

INFECTION OF POTTED ASPEN PLANTS WITH ASCOSPORES OF HYPOXYLON MAMMATUM. P.D. Manion and Y. Yuan, SUNY College of Environmental Science and Forestry, Syracuse, NY 13210

The conditions for infection of aspen by H. mammatum ascospores have eluded most researchers for more than 50 years, until Belanger et al. (Phytopathology 79:315-317) demonstrated infection and necrosis of unwounded moisture-stressed aspen in tissue culture. We initiated 13 ascospore inoculation experiments on nine aspen clones to confirm these results on moisture-stressed, potted trees in the growth chamber. Recovery of H. mammatum from surface-sterilized tissues 4-6 months post inoculation demonstrated ascospore infection of unwounded, needle-puncture wounded, and scalpel-wounded stem tissues. H. mammatum was recovered only from asymptomatic green tissues, suggesting that the fungus has an endophytic phase prior to its pathogenic cankering phase.

USE OF *BRASSICA* SPP. AS GREEN MANURE FOR BIOLOGICAL CONTROL OF *PRATYLENCHUS PENETRANS*. W. McFadden[1], J. Potter[2], J.E. Brandle[3]. [1]Horticultural Research Institute of Ontario, Vineland Station, Ontario, Canada, L0R 2E0; [2]Agriculture Canada Research Station, Vineland Station, Ontario, Canada, L0R 2E0; [3]Agriculture Canada Research Station, Delhi, Ontario, Canada, N4B 2W9.

With increasing public concern about the use of pesticides in agriculture, sustainable alternatives to fumigation for control of soil-borne pests are being sought. The active ingredient in one of the most widely used soil fumigants is methyl isothiocyanate. Glucosinolates present in tissues of *Brassica* spp. are hydrolyzed to forms of isothiocyanate by the enzyme myrosinase when leaf tissue is crushed in the presence of water. These products have been shown to affect insects, fungi and nematodes. Extracts of leaves from different *Brassica* species and cultivars were evaluated *in vitro* for their effects on *Pratylenchus penetrans*. Leaf extracts from some of species tested immobilized nematodes only after 24 hr exposure. Extracts from the mustard cultivar Domo were the most effective, immobilizing nematodes within 10 minutes. Preliminary field trials were conducted in 1991 to evaluate the effect of incorporating a green manure of *Brassica* species on populations of *P. penetrans*.

CABBAGE REMAINED FREE OF BLACK ROT WHEN PLANTED IN CRUCIFEROUS DEBRIS INFESTED WITH *XANTHOMONAS CAMPESTRIS* PV. *CAMPESTRIS*. M. T. McGrath and M. S. Ghemawat, Dept. of Plant Pathology, Long Island Horticultural Research Laboratory, Cornell University, Riverhead, N.Y. 11901-1098

Crop rotation is a recommended practice for managing black rot of crucifers; however, research is lacking that has evaluated its importance. Brussels sprout seed naturally infested with *Xanthomonas campestris* pv. *campestris* (Xcc), was planted in a 790 m² area on 18 July 1990. All plants (2580) developed symptoms of black rot. Crop debris overwintered above ground. After moldboard plowing, 35 stem pieces were collected on 18 April 1991. Extracts obtained from surface sterilized subsamples were plated onto FS medium. Xcc was obtained from 26 pieces; pathogenicity of isolates was confirmed. Xcc-free cabbage seed was planted into the Xcc-infested site on 14 June 1991. Because none of the 1935 plants developed symptoms of black rot before reaching marketable size (23 August 1991), crop rotation is less important than use of disease-free seed for managing black rot of cabbage.

CHRYSOMYXA WEIRII ON PICEA PUNGENS IN PENNSYLVANIA. W. Merrill, J. D. Peplinski, and N.G. Wenner, The Pennsylvania State University, Department of Plant Pathology, 306 Buckhout Lab, University Park, PA 16802.

Chrysomyxa weirii Jacks., an autoecious needle rust, was found infecting Picea pungens (Colorado blue spruce) Christmas trees in western Luzerne County, PA in June 1991. Lower branches on severely infected 2.5-4 m tall, 12-15 year old trees were devoid of all needle complements except those of 1990 and 1991, whereas adjacent non-infected trees bore needle complements from 1987. Severely infected older trees (approx. 30% of the stand) on about 1 hectare lost virtually all 1990 needles. Trace to moderate levels of infection occurred on scattered younger trees throughout another 4 hectares. Source of the seedlings in the severely infected, older portion of the stand is unknown. Given the widespread movement of ball-and-burlap planting stock of P. pungens and the difficulty of inspection procedures, this pathogen poses a threat to Pennsylvania's nursery industry should it become widely established. This is the first record of this pathogen in PA, and the second report of it on P. pungens.

MELAMPSORELLA CARYOPHYLLACEARUM ON ABIES FRASERI IN PENNSYLVANIA. W. Merrill and N.G. Wenner, The Pennsylvania State University, Department of Plant Pathology, 306 Buckhout Laboratory, University Park, PA 16802

In eastern North America, yellow broom rust of Abies species, caused by Melampsorella caryophyllacearum, is distributed throughout the range of A. balsamea in Canada south through northern New England, New York, Michigan and Minnesota. The fungus has been known from PA previously only on the alternate host, Cerastium sp. Earlier searches by Kern and our own searches failed to find it in the limited native stands of A. balsamea. In May 1991 this disease was found in an A. fraseri Christmas tree plantation in Tioga County, PA. The witches' broom was at least 7 years old, and about 0.7 m in diameter. Aecia formed in mid-July; needle casting began in early August. Pathological anatomy of infected needles appeared identical to that of infected A. balsamea. This is the first report of the pathogen on Abies sp. in PA, and the first report of it attacking A. fraseri.

SUBDUE® 2E ROOT DIPS PHYTOTOXIC TO DOUGLAS-FIR. W. Merrill, N.G. Wenner. The Pennsylvania State University, Department of Plant Pathology, 306 Buckhout Lab, University Park, PA 16802.

In studies to control Phytophthora root rot in Douglas-fir transplant beds, 36 bundles of 50 seedlings each were randomly selected from the packing line. Twelve treatments consisted of soaks in water or in Subdue® 2E at 0.16 or 0.31 ml ai/liter of water for 1, 2, 4 or 8 hr. Each treatment was replicated 3 times. All treated seedlings were planted by the nursery crew using a six-row transplanting machine within 45 min of treatment on 7 June 1990. Survival was determined 14 May 1991 by counting all plants that had formed new current-year shoots. Although Subdue® 2E was not significantly phytotoxic to Douglas-fir seedlings when applied at 0.16 ml ai/liter, except with a soaking time of 8 hr, there was a tendency for reduced survival at all treatment levels. It was significantly phytotoxic when applied at 0.31 ml ai/liter with soaking times of 2 hr or longer, and extremely phytotoxic with a soaking time of 8 hr (ave. survival = 5.3 treated seedlings vs 42.7 untreated seedlings). This phytotoxicity precludes its use as a root dip.

COMPARISON OF GENETIC VARIABILITY IN THE CHESTNUT BLIGHT FUNGUS, *CRYPHONECTRIA PARASITICA*, FROM CHINA AND THE UNITED STATES. M. G. Milgroom and S. E. Lipari. Dept. of Plant Pathology, Cornell University, Ithaca, NY 14853.

Cryphonectria parasitica was introduced into the US from Asia about 90 years ago. We tested the hypothesis that genetic variability in this fungus in the US is much less than in the ancestral populations from Asia. Genetic variability in *C. parasitica* was investigated in 16 isolates from China and 17 isolates from the US using RFLPs. Southern blots were probed with 14 randomly selected single- or low-copy probes and a DNA fingerprinting probe. In the Chinese sample, eight of the 14 random probes revealed polymorphisms, while in the US sample, only two were polymorphic. Mean haplotype diversity *(H)* in the Chinese sample was 0.22 while for the US sample $H = 0.08$ for these 14 probes. Inexplicably, there was much greater variation in DNA fingerprints in the US sample than among the Chinese isolates. There were 16 unique fingerprints among US isolates, while there were only six fingerprints among the 16 Chinese isolates.

GENETIC AND PHENOTYPIC CORRELATIONS AS METHODS FOR STUDYING CROSS RESISTANCE AMONG STEROL BIOSYNTHESIS-INHIBITING FUNGICIDES. T.L. Peever and M.G. Milgroom, Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Cross resistance relationships among several sterol biosynthesis-inhibiting fungicides (SBIs) were determined by calculating genetic and phenotypic correlations in resistance using the barley pathogen *Pyrenophora teres*. Genetic correlations equal

to 1 were obtained among four SBIs tested that are known to inhibit the same demethylation step in sterol biosynthesis (DMIs). This indicates that the same genes control resistance to all four DMIs. In contrast, genetic correlations of 0 were obtained between resistance to the morpholine SBI fenpropimorph (different mode of action) and the DMIs indicating that different genes control resistance to these two classes of SBIs. Phenotypic correlations in resistance were also calculated for isolates of *P. teres* sampled from different populations. These calculations revealed that the phenotypic correlations did not necessarily equal the genetic correlations and were not the same from one population to the next.

BICARBONATE INHIBITION OF PHYTOPATHOGENIC FUNGI IN VITRO. Porter, L.L., Urbina-Reyes, R.N., and Horst, R.K., Dept. of Plant Pathology, Cornell University, Ithaca, NY 14853

Sodium and potassium bicarbonate effectively reduce powdery mildew on greenhouse-grown roses and cucurbits, and may suppress other fungal pathogens, such as species of *Alternaria* and *Fusarium*. To uncover antifungal properties of bicarbonates, we assayed germination and growth of single spores of *Alternaria brassicae (Ab)* and *F. graminearum (Fg) in vitro* over a range of concentrations (0.05 to 2.0% w/v) of three bicarbonate salts. *Ab* was inhibited by 0.4% $NaHCO_3$; 1.6 and 2.0% were fungicidal. *Ab* mycelia grew sparsely on 1.6 and 2.0% $KHCO_3$; no *Ab* spores germinated on NH_4HCO_3. Inhibition of *Fg* growth on $NaHCO_3$ or $KHCO_3$ was evident at 0.4, 1.6 and 2.0%; NH_4HCO_3 was lethal at the latter two concentrations. Results suggest that bicarbonates have a profound effect on *Ab* and *Fg*. Extensive work is underway to elucidate the reason(s) for this broad-based effect on these and other fungi.

INCORPORATING WEATHER FORECASTS INTO POTATO LATE BLIGHT DISEASE FORECASTS. R. Raposo, D.S. Wilks[1], and W.E. Fry, Dept. of Plant Pathology, and ([1]) Dept of Soil, Crop and Atmospheric Sciences, Cornell University, Ithaca, NY 14853.

The potential value of weather forecasts to improve fungicide application frequency via BLITECAST and a computer simulation forecast was evaluated using computer simulation models which accurately describe potato late blight development. The maximum contribution of a weather forecast was determined assuming perfect knowledge of future weather, one and two days in advance. Perfect knowledge of future weather (one or two days) increased the level of disease suppression achieved by a protectant fungicide for both disease forecasts. The value of actual (imperfect) weather forecasts was estimated by using a recently developed probabilistic model which identifies the relationship between weather forecasts (temperature and relative humidity) and actual weather. Preliminary analysis indicates that real forecasts were nearly as good as perfect knowledge of future weather in disease suppression.

A LOW-COST SPORE TRAP FOR SAMPLING AT MULTIPLE FIELD SITES. A. M. C. Schilder and G. C. Bergstrom. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

A simple, low-cost spore trap has been developed for use in experiments where multiple sites have to be sampled simultaneously. The trap consists of a 6-ft threaded metal rod (5/16 inch diam) on which a bulldog paper clamp (Boston clip No. 1, Hunt MFG Co, Statesville, NC) is held in place by two 1.5-inch diam metal washers flanked by 5/16-inch nuts. A microscope slide coated with silicone grease is placed in the clamp and can be changed easily at chosen time intervals. The height of the clamp can be adjusted by moving the assembly up or down along the threaded rod. A plastic picnic plate, which acts as a rain cover above the slide, is also held in place with washers and nuts. Multiple clamps can be placed on the rod to sample at different heights. The rods are placed in the soil about 1/2-1 ft deep and are quite stable in varying weather conditions. The total cost per trap is approx. $1.60. The traps have been used succesfully to study aerial dispersal of conidia of the fungus *Pyrenophora tritici-repentis* from wheat.

OZONE INDUCED EFFECTS TO HALF-SIB FAMILIES OF BLACK CHERRY. J. M. Skelly, K. R. Snyder, and J. E. Savage, Dept. of Plant Pathology, The Pennsylvania State University, Univ. Park, PA 16802.

Seven half-sib (h-s) families of Prunus serotina, Ehrh. were exposed for 7h/da, 5d/wk to 30, 60, 90, and 120 ppb ozone for 10 weeks. Exposures occurred within CSTR chambers located within a greenhouse supplied with charcoal filtered air. Half-sibs were from superior black cherry in the Monongehela (MO) and Allegheny (R.M) National Forests. Significant (p>.001) differences were noted between all treatments for all h-s families for percent leaves and percent leaf area symptomatic. Defoliation was greatest (p>.001) in the 90 and 120 ppb O_3 treatments; a difference (p>.001) was also noted in defoliation the 60 ppb vs the 30 ppb O_3 treatments. Half sib families were not different at 30 ppb O_3 for percent leaves or leaf area affected, or defoliation; results were similar at 60 ppb except that defoliation was greatest in h-s R-12 (p>.05). Half-sib family differences were noted as significant but variable at 90 and 120 ppb in all parameters; defoliation of R-12 was again highest. Overall, h-s families MO22, MO23, and R-12 were more symptomatic (all measures) than were h-s families MO17, 10, and 7, and M15.

THERAPY CYCLING TO ELIMINATE HIGH-TITERED, MULTIPLE VIRUS INFECTIONS FROM POTATOES. S. A. Slack, I. S. Dewi, and L. A. Tufford. Plant Pathology Dept., Cornell University, Ithaca, NY 14853.

A protocol for treatment of *in vitro* plantlets to eliminate systemic viruses was established (Can. J. Bot. 68:1515). Efficiency decreases, however, for selected genotypes when a virus is high titered and/or multiple viruses exist. Modified nodal cuttings (≤0.5 mm) of the specialty potatoes 'Purple Fingers' infected with potato viruses S, Y and leafroll and 'All Red' infected with PVS, PVX and PLRV were established on a medium containing ribavirin (20 mg/l) and subjected to 4-6 wks thermotherapy. This cycle was repeated for clones testing positive by ELISA for ≥1 virus. After one therapy cycle, 14% of 'Purple Fingers' and 8% of 'All Red' plantlets tested free from all viruses. System inefficiency was primarily due to low elimination of PLRV (14% from 'Purple Fingers' and 13% from 'All Red'). Virus titers were reduced 27-90% for a specific virus:clone combination with PLRV reduced 60% in 'Purple Fingers' and 72% in 'All Red'. Following the second cycle, 95% of the 'Purple Fingers' and 64% of the 'All Red' plantlets tested virus-free with 100% and 69% freedom from PLRV, respectively. A second therapy cycle is recommended to enhance system efficiency to eliminate recalcitrant viruses.

THE INFLUENCE OF APPLE CULTIVAR ON PSEUDOTHECIA AND ASCOSPORE PRODUCTION. C. A. Smith and W. E. MacHardy, Department of Plant Biology, Univ. of New Hampshire, Durham, 03824.

The influence of apple cultivar on ascospore productivity, pseudothecial density, asci per pseudothecium, and the rate of ascospore maturation of *Venturia inaequalis* (Cke.) Wint. was evaluated in 1990. Ascospore productivity, determined by trapping spores at weekly intervals from heavily infected leaves that had overwintered on the orchard floor, was highest on Rome (15,450 spores/100cm^2) intermediate on Golden Delicious, McIntosh, and Spartan (~10,000-11,400 spores/100cm^2), and lowest on Mutsu, Red Delicious, and Stayman (~5,000-8,000 spores/100cm^2). Pseudothecial density (pseudothecia/100 cm^2) was greatest on Red Delicious, Rome, and Spartan (700-900), intermediate on Golden Delicious and McIntosh (~500), and least on Mutsu and Stayman (~300). Asci/pseudothecium and the ascospore maturation rate was determined by examining squash mounts of pseudothecia through the primary scab season. Asci/pseudothecium was greatest on Spartan (84) and differed significantly only from Red Delicious (53) and Stayman (41). Within the first three weeks, 63% of the season's ascospores had matured in Stayman, ~40-50% in Golden Delicious, McIntosh, Mutsu, Rome, and Spartan, and 15% in Red Delicious.

TRANSFORMATION OF A POWDERY MILDEW, *UNCINULA NECATOR*, BY MICROPROJECTILE BOMBARDMENT. Franzine D. Smith, David M. Gadoury*, Peter R. Harpending, and John C. Sanford, Depts. of Hort. Sci. and Pl. Path.*, Cornell University, NYSAES, Geneva 14456.

A benomyl-sensitive, single-spore clone of *U. necator*, growing on grapevine seedlings, was bombarded with tungsten particles coated with plasmid pbenA3; a pUC19 derivative that contains the benA3 allele of *Aspergillus nidulans*. This gene confers a high level of resistance to benomyl in *Aspergillus*. After bombardment, seedlings were sprayed every 7-10 days with 300 mg L^{-1} benomyl; which is lethal to the wild-type pathogen. After 3 weeks, conidia from colonies that continued to grow were transferred to benomyl-treated seedlings. Controls included (i) colonies that were not bombarded, (ii) colonies that were treated with naked particles, and (iii) seedlings that were not inoculated, but were sprayed with benomyl. Up to 10 % of the colonies that were bombarded with pbenA3 became resistant to benomyl. Transformation was confirmed by isolation of DNA from the wild-type isolate and putative transformants, amplification of DNA with PCR using a primer sequence from the *Aspergillus* gene and comparison of DNA after restriction digestion. This is the first report of transformation of an obligately parasitic fungus.

Quantitative estimation of anthracnose of flowering dogwood in Connecticut. V. L. Smith, CT. Agr. Exp. Sta., New Haven, CT 06504

Naturally-occurring epidemics of anthracnose of flowering dogwood (*Cornus florida*) were assessed at 1 location in 1990 and 3 locations in 1991. Number

of lesions, lesion area, and healthy leaf area were recorded for 50 leaves taken weekly at each location, beginning at leaf emergence through late summer. For all locations in both years, lesion area and number of lesions peaked within 4 wk of leaf emergence, and did not increase significantly after that time. Lesion were larger in 1990 than in 1991. These findings indicate that dogwood anthracnose in CT appears to be a monocyclic disease, with most infections occurring early in the spring. Chemical control measures for anthracnose would therefore be most effective if applied soon after leaf emergence.

MOLECULAR CLONING AND DETECTION OF DNA FOR THE MYCOPLASMALIKE ORGANISM ASSOCIATED WITH BLUEBERRY STUNT DISEASE W. Tang & T.A.Chen, Department of Plant Pathology, Rutgers University, New Brunswick, NJ 08903

Chromosomal DNA of the mycoplasmalike organism (MLO) associated with blueberry stunt (BS) disease was extracted from infected blueberry plants and purified by CsCl buoyant density gradient centrifugation. DNA was digested with HindIII restriction endonuclease, ligated with PUC19 and transformed in E. coli DH5α. A recombinant plasmid containing a 0.5kb MLO DNA segment was selected by differential hybridization against healthy and diseased plant DNA probe. In dot blot and southern hybridization analysis, the DNA probe hybridized with DNA isolated from MLO infected blueberry but not with DNA from healthy plants. Because of the low BS-MLO titer of infected plants, at least 50 ng of total DNA from infected plant were needed for detection. The probe appeared to be specific to BS-MLO since it did not cross hybridize with nucleic acids from three other MLO infected periwinkle and lettuce plants.

EVALUATION OF NITROGEN FORM AND THE RATE OF NITROGEN AND CHLORIDE APPLICATION FOR THE CONTROL OF SUMMER PATCH ON 'FYLKING' KENTUCKY BLUEGRASS. D. C. Thompson, J. R. Heckman, and B. B. Clarke, Dept. of Plant Pathology, Cook College, New Brunswick, NJ 08903.

The influence of nitrogen form and the rate of nitrogen and chloride application was assessed on turf quality and severity of summer patch in 'Fylking' Kentucky bluegrass. Plots were inoculated with Magnaporthe poae in 1990. Varying rates of ammonium sulfate and calcium nitrate and combinations of potassium sulfate and potassium chloride were applied every three weeks from May to October in 1990 and 1991. In 1990, patch diameter and rhizosphere pH were slightly reduced by the application of ammonium sulfate. In 1991, the onset of summer patch, patch development, bulk soil and rhizosphere pH were greatly reduced by ammonium, with the greatest reduction at the highest rates of ammonium application. Chloride did not influence the severity of summer patch, turf quality, or soil pH. Turf quality was not significantly influenced by nitrogen form or rate.

SYMPTOMOLOGY, CYTOPATHOLOGY AND DISTRIBUTION OF TOMATO SPOTTED WILT VIRUS IN INFECTED GREENHOUSE ORNAMENTALS. R.J. Vali and F.E. Gildow. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

Thirteen species of commercially-grown greenhouse ornamentals, originally testing positive for the I and/or L strains of tomato spotted wilt virus (TSWV), were maintained for 1 1/2 years and periodically tested by ELISA. Decreases in virus titre to undetectable levels and corresponding remission of symptoms occurred in all plant species except *Senecio*, *Tolmiea*, *Cyclamen* and *Begonia*. Upon further evaluation of these four species, differences in distribution and titre of virions among symptomatic and asymptomatic tissues were observed by ELISA and TEM. In contrast to an earlier report (Urban et al. 1991, Phytopathology 81:525), TSWV-I was observed as mature membrane-bound virions in cytoplasmic vesicles in *Senecio*, *Tolmiea* and *Cyclamen*. Viroplasm, filamentous inclusions, or amorphous inclusions were not observed.

DERMEA PSEUDOTSUGAE STEM CANKER OF DOUGLAS-FIR IN EASTERN NORTH AMERICA. N.G. Wenner and W. Merrill, The Pennsylvania State University, Department of Plant Pathology, 306 Buckhout Laboratory, University Park, PA 16802.

In 1981 a plantation of Douglas-fir Christmas trees was established in Carbon County, PA in a field that had been in hay or field crops for about 150 years. The "soil" had been eroded and depleted of nutrients and consisted primarily of the shaley subsoil. The plantation has been severely damaged by branch and stem cankers; since 1988 approx. 10,000 1-1.5 m tall dead or dying trees have been cut and burned (approx. 50% of the stand). A complex of several canker fungi is involved in this mortality. One of these is Dermea pseudotsugae (anamorph = Foveostroma boycei), previously reported in western North America from British Columbia south to California. Conidiomata formed occasionally on canker faces throughout the fall and winter months and abundantly throughout the spring. Apothecia with asci and ascospores were found from mid-February through April. This is the first report of this pathogen in eastern North America.

LEUCOSTOMA KUNZEI CANKER OF DOUGLAS-FIR AND FRASER FIR CHRISTMAS TREES. N.G. Wenner and W. Merrill, The Pennsylvania State University, Department of Plant Pathology, 306 Buckhout Laboratory, University Park, PA 16802

Because of increasing difficulty in producing and marketing Scots pine Christmas trees, increasing acreages of Douglas-fir and Fraser fir have been planted in PA, much of it on low quality sites. Concurrently, branch and stem cankers have increased in incidence. Following the 1988 drought, a mixed planting of Douglas-fir and Fraser fir in Tioga County, PA was severely damaged. Multiple rows of trees on shallow soil over sandstone outcrops died or lost their tops due to girdling branch and stem cankers. Adjacent rows of trees on deeper soil were unaffected. Canker faces bore masses of sexual and asexual fruiting structures of Leucostoma kunzei (anamorph = Cytospora kunzei). Many trees with tops girdled in 1988 formed multiple replacement leaders in 1989 and 1990. Many of these new tops were subsequently girdled during the 1991 drought. This is the first report of L. kunzei attacking Fraser fir.

EVALUATION OF AN INTEGRATED, REDUCED-SPRAY PROGRAM USING DMI FUNGICIDES FOR CONTROL OF PRIMARY APPLE SCAB
W. F. Wilcox, D. I. Wasson*, and J. Kovach, Cornell Univ., NYS Agr. Expt. Sta., Geneva 14456 and *Cornell Coop. Extension., Albion NY 14411

A reduced-spray program for control of primary apple scab was evaluated for 3 yr in 14 commercial apple orchards in western NY. Growers were advised to apply a DMI fungicide (fenarimol, flusilazol, or myclobutanil) in combination with oil or insecticides (if needed) at the following four phenological stages, regardless of the timing of scab infection periods: (1) tight cluster (TC); (2) pink bud (P); (3) petal fall (PF); and (4) PF +10 days (1C). This schedule is predicated on low numbers of discharged ascospores prior to TC in most commercial orchards and the extended curative and suppressive activities of the DMI's when used in two sequential applications (TC + P, PF + 1C); it contrasts with a standard program of 6-7 primary scab sprays in this region. Including copper applied in some orchards at green tip to control fire blight, the mean number of fungicide applications in all test orchards was 4.1, 4.4, and 4.5 in 1988, '89, and '90, respectively. The mean incidence of fruit scab was 0.2, 1.0, and 1.6% in the same respective years, during which an average of 5.2, 14.4, and 8.8 primary infection periods occurred.

VIRULENCE OF DIPLOCARPON EARLIANA TO STRAWBERRY G. Xue, J. Zheng, and J. C. Sutton Department of Environmental Biology, University of Guelph, Guelph, Ontario, Canada N1G 2W1

Virulence of D. earliana was evaluated using 2-cm-diameter discs cut from young strawberry leaves. The discs were placed on fibreglass screen overlying moist paper towel in petri dishes. They were then sprayed with a suspension of D. earliana spores (5 X 10^4 condia/ml water), and incubated at 20 C with a 14 h photoperiod. Disease severity was estimated at 14 d after inoculation. Severity ratings for leaf discs and for attached leaves from eight strawberry cultivars inoculated with one isolate of D. earliana were significantly correlated ($r = 0.995$, $P < 0.0001$). Six pathotypes of D. earliana were differentiated when eight isolates of the pathogen were evaluated on 18 strawberry cultivars. One pathotype, composed of one isolate each from Nova Scotia and Washington State, was virulent (>5% leaf disc infected) on 17 cultivars. The five other pathotypes, composed of Ontario isolates, were differentially virulent on 4 to 16 of the cultivars. Cultivar Vibrant was resistant to all isolates and cv. Vantage was resistant to Ontario isolates only. This is the first report of variation in virulence of D. earliana.

CHARACTERIZATION OF DIVERSITY AMONG STRAINS OF *PSEUDOMONAS CEPACIA* ISOLATED FROM CLINICAL SOURCES, SOILS, AND INFECTED ONIONS. D. S. Yohalem and J. W. Lorbeer. Department of Plant Pathology, Cornell University, Ithaca, NY 14853.

Pseudomonas cepacia ex Burk. (Palleroni and Holmes) was first described as a soft-rotting pathogen of onions. Later synonomies expanded the species concept to include strains isolated from soils, fresh water, and nosocomial sources; the

variability and extreme ecological diversity of the group calls these synonomies into question. Several strategies were attempted for division of the species based on the source of strains and pathogenicity to onion. Fatty acid profiles (27 strains), isoenzymes profiles (62 strains), and two systems based on suites of variable characters (203 strains) (Zentrallbl. Bakteriol. Microbiol. Hyg. [A] 224:478 and Ann. Biol. Clin. (Paris) 39: 9) failed to distinguish groups based on *a priori* criteria. At least two clusters are apparent when using the Gower coefficient calculated for 113 characters for the 203 strains, indicating a need for taxonomic revision of the group based, however, on criteria other than source.

SIZE AND STRUCTURE OF A HIGH MOLECULAR WEIGHT DSRNA FROM "BARSOY" BARLEY. I. Zabalgogeazcoa and F. E. Gildow. Department of Plant Pathology, The Pennsylvania State University, University Park, PA 16802.

A high molecular weight dsRNA isolated by phenol extraction and cellulose chromatography from healthy barley (*Hordeum vulgare* cv. Barsoy) was characterized for size and secondary structure. The dsRNA was not associated with virus-like particles, as judged by density gradient purification and observations of leaf tissue by electron microscopy. No change in the electrophoretic mobility of the dsRNA was observed after treatment with S1 nuclease, suggesting that the entire molecule is base-paired. Observation of platinum shadowed purified dsRNA by electron microscopy revealed linear molecules comparable to dsDNA. Of 253 dsRNA molecules measured, more than 50% were 12-14 kbp long. In 1% agarose gels utilizing dsRNA standards, an estimate of 13.4 kbp was obtained for Barsoy dsRNA. However, when ds-DNA was used as a standard, the estimated size was larger (19-23 kbp) and did not correspond to the electron microscopy data.

INFLUENCE OF TEMPERATURE AND WETNESS DURATION ON INFECTION OF BLACK SPRUCE BY *Botrytis cinerea*. P.G.Zhang and J.C. Sutton, Dept. of Environmental Biology, University of Guelph, Ontario, Canada N1G 2W1.

Seedlings of black spruce (*Picea mariana* BSP.) were kept in the dark at 36 C for 96 h for predisposition to *Botrytis cinerea*, then inoculated at 20-21 C with a spore suspension of the pathogen (10^6 conidia/ml water · plus surfactant). Inoculated plants were kept in various air temperatures (1-36 C), wetness durations (0-48 h), and regimes of light and dark periods. Infection was estimated indirectly by assessing incidence of sporulation of *B. cinerea* in leaves placed on an agar medium containing paraquat. Minimum, optimum, and maximum temperatures for infection were between 4 and 12 C, near 20 C, and between 28 and 36 C, respectively. At 12, 20, and 28 C, the pathogen infected the leaves in wetness duration $\geq$12 h but not when wetness lasted $\leq$8-9 h. Incidence of infection increased sigmoidally with wetness duration at 12, 20 and 28 C. A logistic model [$Ln(y+1)/(99-y)=a+(b1+b2T+b3T^2)*WD$] was developed to describe the relation of temperature (T) and wetness duration (WD) to infection (y) (R^2 between 0.8992 and 0.9188).

A REPEATED SEQUENCE IN A NATIVE PLASMID (pCXC100) OF CLAVIBACTER XYLI SSP. CYNODONTIS. Y. P. Zhang, M.C. Metzler, & T. A. Chen. Department of Plant Pathology, Rutgers University, New Brunswick, N.J. 08903

A 51-kb plasmid (pCXC100) was isolated from a majority of isolates of Clavibacter xyli ssp. cynodon tis (CXC), which is a non-pathogenic, gram-positive bacterium inhabiting the xylem of Bermudagrass. Using eight enzymes, a restriction map was constructed for the plasmid. One PstI fragment within the plasmid (P7) exists in multiple copies in the bacterial chromosome as demonstrated by Southern blot analysis. The P7 sequence does not hybridize to DNA from Clavibacter michiganense ssp.sepedonicum, indicating that it is not related to the chromosomal repeat found on the large native plasmid of that bacterium. A 1.1-kb region of the P7 DNA fragment has been sequenced. We are currently analyzing the DNA sequence to search for characteristics of transposons such as inverted repeats or open reading frames coding for transposase.

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