

**Comparison of *Verticillium dahliae* isolates from cruciferous and non-cruciferous hosts.** R. G. BHAT and K. V. Subbarao. UC Davis, 1636 E. Alisal St., Salinas, CA 93905. *Phytopathology* 89:S7. Publication no. P-1999-0042-AMA.

Representative isolates of *Verticillium dahliae* from four crucifer and 11 non-crucifer hosts and an isolate of *V. albo-atrum* from alfalfa were compared for morphology of conidia and microsclerotia, pathogenicity, ability to produce *nit* mutants, and random amplified polymorphic DNA (RAPD) markers. Crucifer isolates produced a low number of large conidia on PDA compared with *V. albo-atrum* and non-crucifer isolates, which produced abundant, small conidia. Microsclerotia of crucifer and non-crucifer isolates were irregular in shape. Crucifer isolates produced more linear microsclerotia on PDA compared with non-crucifer isolates that produced more globular microsclerotia. Crucifer isolates were severely pathogenic only on crucifer hosts with some exceptions and caused mild infection on few other hosts. Non-crucifer isolates and *V. albo-atrum* isolate were destructive on their host of origin and showed varying degrees of infection on other hosts tested. Only crucifer isolates did not produce *nits*. RAPD marker profiles of crucifer isolates were distinct from others. Many of the markers present in crucifer isolates were not present either in *V. albo-atrum* or non-crucifer isolates, indicating that crucifer isolates were not the result of parasexual hybridization between *V. dahliae* and *V. albo-atrum*. Thus, crucifer isolates belonged to a separate group in *V. dahliae*.

**Pathogenicity of *Verticillium dahliae* isolates on plants other than their own hosts.** R. G. BHAT and K. V. Subbarao. University of California, Davis, 1636 E. Alisal St., Salinas, CA 93905. *Phytopathology* 89:S7. Publication no. P-1999-0043-AMA.

Pathogenicity of *Verticillium dahliae* isolates from artichoke, bell pepper, cabbage, cauliflower, chili pepper, cotton, eggplant, horseradish, lettuce, mint, oilseed rape, potato, strawberry, tomato, and watermelon was tested on four crucifers (bok choy, chinese cabbage, radish and oilseed rape) and three non-crucifers (bean, carrot and sugarbeet). Spore suspensions containing approximately  $10^7$  conidia/ml were used for root-dip inoculation of 30-day-old seedlings in the greenhouse. Plant height, percent disease incidence and disease severity were recorded eight weeks after inoculation. Isolates from crucifer hosts and watermelon caused severe disease on crucifers with the exception of horseradish isolate on chinese cabbage. Isolates from cotton, lettuce, and strawberry were non-pathogenic to crucifers whereas the remaining isolates differentially interacted. Carrot and sugarbeet plants were resistant to all the isolates tested. Bean plants, which have been considered as non-hosts of *Verticillium*, were susceptible to nine isolates which included the isolates from artichoke, bell pepper, tomato and strawberry among others. Data support the previous conclusion that *V. dahliae* isolates from various hosts have a host range specificity.

**In search of the *Ralstonia solanacearum* siderophore gene.** G. BHATT and T. P. Denny. University of Georgia, Athens, GA. *Phytopathology* 89:S7. Publication no. P-1999-0044-AMA.

*R. solanacearum*, a soil borne phytopathogen that causes wilt disease in a wide variety of plants, produces the siderophore schizokinen. The goal of this project is to isolate and characterize the siderophore biosynthetic genes and determine the role of iron acquisition in pathogenesis. Mutants of the global virulence regulator, PhcA, show increased siderophore production. Downstream of *phcA* is the ferric uptake regulator (*fur*) gene, which suggests that iron may be involved in virulence. Random Tn5 mutants were screened for reduced production of schizokinen using the CAS universal siderophore indicator medium, which was modified for *R. solanacearum* by reducing the PIPES to 0.3% and using various non-deferrated complex amino-acid mixtures. The first siderophore mutant appeared to have Tn5 inserted in the sulfite reductase gene, and is thus a cysteine auxotroph. This mutant is too pleiotropic, for our purposes. At least three other mutants (which do not seem to be amino acid auxotroph ht. The siderophore mutants will be tested for changes in virulence and survival.

***Antrodia sinuosa* is significant in brown heartwood rot of living lemons in Arizona.** D. M. BIGELOW (1), M. E. Matheron (2), and R. L. Gilbertson (1). (1) University of Arizona, Tucson, AZ; (2) University of Arizona, Yuma, AZ. *Phytopathology* 89:S7. Publication no. P-1999-0045-AMA.

Brown heartwood rot has been observed in all mature lemon trees sampled in the Yuma area. Our research has shown brown rot in Yuma lemon orchards is due mainly to two basidiomycetes, *Coniophora eremophila* Lindsey & Gilb.

and *Antrodia sinuosa* (Fr.) P. Karst. *A. sinuosa* was isolated from heartwood rot samples from 63% of the 40 lemon orchards checked to date. Heterogenic incompatibility tests pairing 26 isolates of *A. sinuosa* from 11 orchards indicated that they are different genotypes. This implies that infection resulted from airborne basidiospores. Mean length of decay column in inoculated 4-cm diameter lemon branches after six months was 20.7 cm for *A. sinuosa* compared to 4.7 cm for *C. eremophila*. Fruiting bodies of *A. sinuosa* have been found in orchards each month since the survey was started in September 1998. *C. eremophila* has not been found fruiting on lemon trees. *A. sinuosa* is potentially more destructive than *C. eremophila* because it fruits readily in the field and grows more rapidly.

**Effect of soil moisture on tomato seed-to-seedling transmission of *Clavibacter michiganensis* subsp. *michiganensis*.** C. M. Biggerstaff, M. L. GLEASON, and E. J. Braun. Department of Plant Pathology, Iowa State University, Ames, IA 50011. *Phytopathology* 89:S7. Publication no. P-1999-0046-AMA.

To determine the effect of soil moisture on seed-to-seedling transmission of *Clavibacter michiganensis* subsp. *michiganensis* (*Cmm*) in tomatoes, *Cmm*-infested tomato seeds (cv. Heinz 1916) were germinated in two different media (one composed of 20% soil, 50% peat and 30% perlite, the other a commercial mix of peat and perlite) in sealed plastic cups at six different matric potentials from near saturation to  $-0.1$  MPa. Cups were placed in a growth chamber at 27 C and 75% RH under 24 h light. All seedlings in a treatment were harvested when 40% of the seedlings had fully expanded cotyledons. After noting presence or absence of attached seed coats, seedlings (including seed coats if attached) were individually macerated in buffer and plated on selective media. There were no significant differences in percent transmission of *Cmm* among moisture treatments, although there was a trend toward greater occurrence of *Cmm* on seedlings germinated at intermediate matric potentials. *Cmm* transmission was highly correlated with seed coat retention; 94% of 119 *Cmm*-positive seedlings retained their seed coat.

**An improved ELISA for detection and quantitation of neomycin phosphotransferase II in plants.** W. O. BLISS, J. Q. Xia, and C. L. Sutula. Research Dept., Agdia, Inc., Elkhart, IN 46514. *Phytopathology* 89:S7. Publication no. P-1999-0047-AMA.

An improved ELISA has been developed to detect and measure neomycin phosphotransferase II (NPT II) in transgenic plants. The test is based on a polyclonal antibody and a monoclonal antibody specific for NPT II protein. The microtiter plate is coated with the polyclonal antibody and incubated with sample. NPT II protein captured on the plate is then detected with the monoclonal antibody and an antimouse antibody-enzyme conjugate. Test conditions were optimized by comparing different buffers, surfactants, blocking reagents, and washing and incubation times. Specificity of the resulting ELISA is much improved compared to currently available NPT II tests. This test can be used with crops such as corn, potato, tobacco, cucurbits and others. NPT II protein can be detected at 0.4 ng/ml, or 40 pg per test well. This improved ELISA provides a reliable way to detect and quantitate NPT II in transgenic plants.

**\* Variation in aggressiveness of *Sphaeropsis sapinea* RAPD marker groups on several conifers.** J. T. BLODGETT and G. R. Stanosz. University of Wisconsin, Madison, WI. *Phytopathology* 89:S7. Publication no. P-1999-0048-AMA.

Diseases caused by *S. sapinea* can result in extensive losses in Christmas tree and ornamental plantings. Hosts with particular value as ornamental or Christmas trees include Scots varieties East Anglia and Austrian Hills, red, and mugho pines, Colorado blue spruce, Douglas-fir, and balsam fir. Seedlings of these species/varieties were wounded and inoculated with isolates of the two RAPD marker groups (A and B) of *S. sapinea*. Symptom severity (distance from the inoculation site at which necrotic needles were present) resulting from inoculation with group A isolates exceeded that of B isolates on all hosts except blue spruce. The hosts varied considerably in responses to A group isolates. Based on symptom severity, East Anglia Scots pine was most susceptible and balsam fir was least susceptible when inoculated with A isolates. The pathogen was recovered from symptomatic and asymptomatic seedlings inoculated with isolates of both groups. This study emphasizes the importance of characterizing the group(s) of *S. sapinea* encountered and the potential to compare resistance to both groups among coniferous genera, species, and/or varieties.