

Disease Notes (continued)

- * **First Report of *Botryosphaeria dothidea* Basal Canker of Pistachio Trees in South Africa.** W. J. Swart and J. T. Blodgett, Department of Plant Pathology, University of the Free State, Bloemfontein 9300, South Africa. Plant Dis. 82:960, 1998; published on-line as D-1998-0611-01N, 1998. Accepted for publication 10 June 1998.

Pistachio (*Pistacia vera*) cultivation is a relatively new industry in South Africa with tremendous economic potential. However, disease problems could develop in time. The pathogens of pistachio found in other parts of the world plausibly will spread to South Africa and new pathogens never recorded on this host may develop here. In the summer of 1998, 2-year-old rootstocks of *P. atlantica* and *P. integerrima* displaying basal cankers and discolored phloem and xylem were observed in the Prieska district of the Northern Cape province. The objective of this study was to identify the causal agent(s). Cankered stems were split and small samples of discolored wood were aseptically placed on corn-meal agar containing streptomycin and incubated for 5 days. Fungi that grew into the agar were transferred to 1.5% water agar dishes containing pine needles to aid sporulation. Species isolated from stems included *Fusarium* spp. (44%), *Fusicoccum aesculi* (anamorph of *Botryosphaeria dothidea*) (23%), a *Cytospora* sp. (19%), a *Chaetomium* sp. (3%), and several nonsporulating mycelial fungi (11%). Inoculum of suspected pathogens (*F. aesculi*, the *Cytospora* sp., and two *Fusarium* spp.) were prepared by culturing fungi in petri dishes on potato dextrose agar overlaid with sterile cheesecloth strips (approximately 15 × 25 mm) until the strips were completely colonized. Greenhouse inoculations involved wounding *P. atlantica* stems by removing the bark (approximately 3 × 6 mm) approximately 10 cm above the soil, and wrapping the colonized cheesecloth followed by Parafilm around the stems at the wound site. Eight plants per isolate and eight wounded and nonwounded (untreated) control plants were used. Noncolonized cheesecloth was applied to wounded controls but not to nonwounded controls. Treatments were assigned randomly. Eight weeks after inoculation, the surrounding bark was removed from all treated shoots and the cambium was examined for discoloration. The length of each cambial lesion was measured and stem sections were surface disinfested and transferred to 1.5% water agar dishes, and the presence of the inoculated fungi was confirmed. Only *F. aesculi* was pathogenic to *P. atlantica* and produced cankers on all stems. The fungus was recovered from all tissues sampled 3 cm beyond the wound sites, 88% of those sampled from the wound sites, and none of the control treatments. Discoloration of the phloem and xylem was similar to that observed in the field. The mean canker length was 41 mm on trees with a mean stem diameter of 7 mm. The appearance of *B. dothidea*, in addition to *B. obtusa* (1), on pistachio justifies the need for establishing a disease management program for pistachio in South Africa.

Reference: (1) W. J. Swart and W.-M. Botes. Plant Dis. 79:1036, 1995.

***Thlaspi arvense*, a New Host for *Alternaria brassicicola*.** A. C. Cobb and H. R. Dillard, Department of Plant Pathology, New York State Agricultural Experiment Station, Cornell University, Geneva 14456. Plant Dis. 82:960, 1998; published on-line as D-1998-0617-01N, 1998. Accepted for publication 12 June 1998.

A leaf spot was observed on cruciferous weeds growing in a cabbage field located in Geneva, NY, on 1 August 1996. The leaf spots on the weeds were dark gray to black in color and varied in size from pinpoints to 1 mm in diameter. The cabbage (*Brassica oleracea* L. var. *capitata* L.) was infected with *Alternaria brassicicola* (Schwein.) Wiltshire, the cause of *Alternaria* leaf spot. The weeds were identified as *Thlaspi arvense* L., a winter annual commonly referred to as field pennycress, stinkweed, or fanweed depending on geographic location. Isolations from the diseased weed tissue yielded *A. brassicicola* (2). The numerous conidia occurred in chains of 10 or more, ranged in size from 14 to 53 µm in length, were 5 to 18 µm wide, contained from 1 to 6 transverse septa with rare longitudinal septa, and were olivaceous in color. An apical beak was absent. On potato dextrose agar (PDA) the colony was dark olive-green to black in color and velvety. Seed was collected from the *T. arvense* plants in the spring of 1997. One hundred seeds were placed in petri plates containing PDA amended with 0.01% of chloramphenicol and streptomycin sulfate. *A. brassicicola* was not isolated from the seeds. A different area of the field was planted to cabbage in 1997 and the cruciferous weeds were allowed to grow. The 1997 population of *T. arvense* consisted of plants from the previous season that flowered early and plants from seeds that germinated late in the season but did not flower. *A. brassicicola* was isolated from nonflowering weeds in September and from flowering

weeds in October. Nonflowering plants were removed from the field in November, planted in pots, and placed in the greenhouse to induce flowering. Identity of both plant populations was confirmed as *T. arvense* (Warren Lamboy, Cornell University, Geneva, NY). Pathogenicity of *A. brassicicola* isolates from *T. arvense* was demonstrated on cabbage and *T. arvense* by following Koch's postulates. Conidia (10^5) from a 5-day-old culture isolated from *T. arvense* grown on PDA were atomized onto field pennycress and cabbage plants with a Preval sprayer. The plants were enclosed in plastic bags and put under lathe shading in the greenhouse. The pathogen was reisolated from symptomatic tissue of both plants after 5 days. This weed could serve as a potential source of *A. brassicicola* inoculum because it is not controlled by herbicides used in crucifer production systems. *Alternaria raphani* has been reported on *T. arvense* in Canada (1). This is believed to be the first report of *A. brassicicola* on *T. arvense*.

References: (1) K. Mortensen et al. Can. Plant Dis. Surv. 73:129, 1993. (2) P. Neergaard. 1945. Danish Species of *Alternaria* and *Stemphylium*. Oxford University Press, London, pp. 137-138.

Subgroup Determination of Bulgarian Isolates of Cucumber Mosaic Virus and the Presence of Satellite RNAs. V. A. Mavrodieva, Plant Protection Institute, 2230 Kostinbrod, Bulgaria; and D. J. Barbara and N. J. Spence, Horticulture Research International, Wellesbourne, Warwick, CV35 9EF, UK. Plant Dis. 82:960, 1998; published on-line as D-1998-0528-01N, 1998. Accepted for publication 27 May 1998.

Cucumber mosaic virus (CMV) is one of the most important viruses in Bulgaria, causing severe losses to agriculture, but little is known about the occurrence and distribution of subgroups within the country or the presence of satellite RNAs (satRNAs). Samples showing typical symptoms (mild to severe mosaic, vein clearing, vein necrosis, leaf deformation, stunting, and fruit necrosis) on several important crops (tomato, cucumber, pepper, bean, courgette, and tobacco) were collected from the main agricultural regions of the country. Isolates were maintained by sap inoculation in tobacco plants. Total RNAs were isolated from 38 samples (including two from bean) and used in reverse transcription-polymerase chain reaction (RT-PCR) assay with primers corresponding to the coat protein (CP) gene of RNA3 (3). A single strong band, 870 bp in length, was produced from all these samples. Amplified products were analyzed for subgroup differentiation by digestion with the restriction endonucleases *MspI* (3), *PvuII*, and *EcoRI*. The *MspI* subgroups 2 and 1 designated by Rizos et al. (3) according to their restriction endonuclease digest data correspond to the subgroups I and II widely used in the literature and based on serology, sequence data, and other properties. In this report, the subgroups are referred to as I and II for the sake of clarity. Isolates in both subgroups were found in all the main regions of Bulgaria. A few variations in *MspI* and *EcoRI* digestion patterns were seen, indicating some variability between isolates within subgroups. Only five samples, three from tomato and two from pepper, were found to be subgroup II. Subgroup I isolates were found in all the crops sampled. The PCR product from one representative isolate of each subgroup was cloned and sequenced by standard procedures. Alignment of the nucleotide and predicted amino acid sequences with published sequences of the CMV CP gene confirmed that the amplified products were derived from CMV. A further eight samples from bean gave only weak amplification and digestion of the products suggested they were likely to be subgroup II. However, these samples were unusual in not inducing symptoms in inoculated tobacco and in being difficult to propagate. The nature of these virus isolates is therefore unclear. Only a single occurrence in Bulgaria of satRNA of CMV has been reported (4) but in this study satRNAs were detected by RT-PCR (1) in total plant RNA extracts of 21 of the 38 samples tested. Amplified products of two of them, NB and 146D, were sequenced; comparison with published sequences confirmed that they were derived from CMV satellite. As expected from the symptoms induced by these isolates, a sequence homologous to the domain of satRNA Y responsible for bright yellow mosaic on tobacco (2) was identified in satRNA NB but not in satRNA 146D. satRNAs were not detected in the eight bean samples that had given only weak amplification with the CMV CP gene primers. The results presented here clearly demonstrate the presence of both subgroups of CMV in Bulgaria. Although CMV in Bulgaria has been studied previously by serological methods, no evidence had been found for the presence of subgroup II.

References: (1) F. Grieco et al. Virology 229:166, 1997. (2) C. Masuta and Y. Takamami. Plant Cell 1:1165, 1989. (3) H. Rizos et al. J. Gen. Virol. 73:2099, 1992. (4) E. Stojmenova. J. Cult. Collect. 1:45, 1995.