## Pathogenic and Nonpathogenic Verticillium Species from South Central Washington

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## ABSTRACT

Among ten Verticillium isolates from five host species, the four V. dahliae isolates were pathogenic on eggplant and cantaloupe and one each on tomato and mint. The four V. tricorpus and the two V. nigrescens isolates were not pathogenic. Phytopathology 61:435-436.

Additional key words: Solanum, Cucumis, Lycopersicon, Mentha.

Verticillium wilt is a major disease in the irrigated areas of south central Washington. During investigation of this disease on mint (Mentha piperita L.), sweet cherry (Prunus avium L.), and cantaloupe (Cucumis melo L.), a number of isolates of Verticillium were obtained. Most isolates produced hyaline mycelium and microsclerotia, and were identified as V. dahliae Kleb. Other morphological forms, however, were noted. Cultures sent to Ivor Isaac, University College of Swansea, Swansea, Wales, were identified as V. tricorpus (Isaac) and V. nigrescens Pethybr. (1). Isaac previously had found Australian isolates of V. nigrescens to be pathogenic to potato, Solanum tuberosum L., and tomato, Lycopersicon esculentum L. (2, 4, 6). In 1952, he described V. tricorpus (5) and reported it pathogenic to tomato and nonpathogenic to potato. Verticillium nigrescens also has been isolated from several hosts in Canada (1), but pathogenicity studies were not reported.

The purpose of this paper is to present data from pathogenicity tests of morphologically different isolates of Verticillium. A preliminary report has been presented (11).

Cultures were grown on prune extract agar for macroscopic and microscopic observations (12). Inoculum was prepared by growing the isolates in a modified liquid medium (7) in shake culture. Four g of yeast extract, 2.0 g KH2PO4 and 1.0 g K2HPO4 were added per 1,000 cc of water. All isolates produced abundant spores on this media. The flasks were incubated 10-14 days at room temperature.

Seedlings of cantaloupe C. melo, 'Hales Best 45'; eggplant, Solanum melongena L. 'Black Beauty'; tomato, Lycopersicon esculentum L. 'Bonny Best'; and freshly rooted cuttings of mint, M. piperita 'Mitcham' were inoculated by dipping the roots in the fungus suspension which contained a heavy concentration of spores (approx  $52 \times 10^5/\text{cc}$ ), and transplanted into a metal flat containing a mixture of one-third sand and

1. Pathogenicity of Verticillium spp. to peppermint, eggplant, cantaloupe, and tomato

			Pepi	Peppermint	Egg	Eggplant	Cant	Cantaloupe	To	Tomato
	Origin	п		Standard		Ctondond		Ctondond		C+1
Isolate	Host	Cultivar	Mean	error	Mean	error	Mean	error	Mean	error
Verticillium dahliae	Cucumis melo L.	Hales Best 45	9.6a	0.33	0 804	0.20	3.75	1.06 <sup>b</sup>	15.0c	4.6
V. dahliae	Mentha piperita L.	Mitcham	2.00	1.04	3.69	1.55	4.08	0.58	0.69	11.0
V. dahliae	Prunus avium L.	Bing	9.83	0.17	0.36	0.23	5.05	0.32	62.3	7.9
V. dahliae	P. avium L.	Bing	6.67	0.33	1.05	0.51	5.93	0.94	66.7	8.9
V. tricorpus	Solanum	Russet (tuber) d	9.73	0.27	8.55	0.76	8.65	0.46	60.7	9.9
	tuberosum L.									
V. tricorpus		Russet (tuber) d	9.50	0.50	8.86	0.38	9.45	0.33	66.3	6.6
V. tricorpus	I.	Mitcham	9.00	1.00	8.89	0.70	9.23	0.43	67.0	13.5
V. tricorpus	C. melo L.	P.I. 262169	9.83	0.17	9.20	0.80	8.45	1.18	0.09	1.7
V. nigrescens	P. avium L.	Bing	9.50	0.50	7.70	0.95	8.70	0.76	71.3	13.3
Noninoculated control			9.50	0.50	9.35	0.31	09.6	0.43	56.3	1.3
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Disease index based on healthy plants, 10 plants/replicate, 4 replicates. Disease index based on 10 = healthy, 8 = stunted, 3 = wilted, and 1 = dead. Disease index based on growth in mm. Isolated by W. B. Hoyman. Number of experiments.

two-thirds Warden fine sandy loam. The experiments were conducted in soil temperature tanks maintained at about 15 C, and in an air temperature of approx 21 C. The experimental design was a randomized complete block with four replicates of 10 plants each. Each test was repeated 3-5 times. Disease data were taken after 8 weeks on mint and after 4-7 weeks on tomatoes, eggplants, and cantaloupes.

In these experiments, pathogenicity was determined using a disease index based on healthy plants for mint and eggplants, growth in millimeters of tomato plants, and the assignment of a numerical score of 1 = dead, 3 = wilted, 8 = stunted, and 10 = the cantaloupe plants.

The mycelium of *V. dahliae* isolates was hyaline, and formed microsclerotia which varied in size, shape, and numbers. No particular morphological character could be related to isolates from a given host. Some isolates, upon repeated subculturing, lost the ability to produce microsclerotia.

The hyphal cells of *V. nigrescens* rounded up and formed chlamydospores singly or in chains.

Verticillium tricorpus produced microsclerotia, dark mycelium, and chlamydospores. Initially these isolates produced a yellow pigment, but after they had been kept in culture they lost this ability.

The data from these experiments are summarized in Table 1. Only Verticillium dahliae (No. 2) from mint was pathogenic to mint. This isolate came from a field in which the mint was severely diseased. Verticillium tricorpus (No. 7) was nonpathogenic even though isolated from a mint plant showing typical but mild symptoms of Verticillium. No other infected plants were observed in this field.

Some variation occurred in experiments with eggplant. All of the *V. dahliae* isolates, however, including the one from mint, were consistently pathogenic to eggplant and cantaloupe.

Verticillium dahliae (No. 1) isolate from cantaloupes was the only one pathogenic to tomatoes. Tomatoes inoculated with this isolate remained stunted and some died. Verticillium tricorpus and V. nigrescens were essentially nonpathogenic to the hosts used in these tests.

Pathogenic variation among isolates of the microsclerotial types of *Verticillium* has been reported from various areas (3, 10) including Washington (9). As far as is known, however, the dark mycelial type of *Verticillium albo-atrum* Reinke & Berth. has not been reported from Washington, and this constitutes the first report of the occurrence of *V. tricorpus* and *V. nigrescens* from this state.

The potential pathogenicity of these latter two spe-

cies needed to be determined because *V. tricorpus* was isolated from peppermint plants being certified "free of *Verticillium*" and from cantaloupe lines being tested for field resistance to *Verticillium dahliae*.

In these experiments, V. tricorpus and V. nigrescens were weak or nonpathogenic, and were not capable of causing losses such as those caused by V. dahliae, which usually kills peppermint, eggplant, and cantaloupe. Verticillium tricorpus and V. nigrescens were reisolated occasionally from eggplant, but this is not surprising since Verticillium dahliae has been isolated from several hosts upon which the fungus had no adverse effects (8). Verticillium nigrescens was also isolated from spearmint, Mentha cardiaca L. 'Scotch', and was nonpathogenic to M. piperita (data not included). They were morphologically stable in culture during the period of this study except for loss of ability by V. tricorpus to produce the yellow pigment.

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