Outbreak of a Stem Necrosis on Chrysanthemum Incited by *Pseudomonas cichorii* in Florida

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ABSTRACT

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An outbreak of a stem necrosis on chrysanthemum was observed in two commercial fields in Florida in the winter of 1982. About 2.25 acres were lost to the disease in one field. No leaf symptoms were associated with the disease. Symptoms overlapped those incited by *Fusarium solani*. *Pseudomonas cichorii* was consistently isolated from diseased tissue and was demonstrated in subsequent inoculation tests to incite the disease.

In the winter of 1982, a dark-colored stem necrosis was observed on maturing chrysanthemum (*Chrysanthemum morifolium* Ramat.) plants in two fields in Florida. No foliar symptoms were observed; symptoms overlapped those associated with Fusarium stem rot (3). In one field, about 2.25 acres were lost as a result of the stem necrosis. The cultivars Florida and Blue Marble appeared to be the most susceptible. In some beds, 90% of these cultivars were lost as a result of the stem necrosis.

A green fluorescent bacterium was isolated from the margins of the stem lesions. The objectives of this study were to identify the bacterium and to attempt to reproduce the symptoms.

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MATERIALS AND METHODS

Isolation. Diseased stems were cut into small sections at the interface of brown discolored areas and green tissue. For bacterial isolation, chips were triturated in sterile distilled water and incubated for 20 min. The liquid was streaked on medium B (KMB) of King et al (10). After 24 hr of incubation at 24 C, fluorescent colonies were streaked on KMB to obtain pure colonies. For fungal isolation, chips were surface-sterilized with 0.54% sodium hypochlorite for 3 min and plated on potato-dextrose agar and on galactose agar for identification.

Identification. The scheme of Hildebrand and Schroth (7) was used to identify the fluorescent bacterial colonies. All strains were checked for arginine

dihydrolase (18), oxidase activity (12), and induction of a hypersensitive reaction on Early Cal Wonder pepper by the procedure of Klement et al (11). Levan production was tested by the procedure of Lelliott et al (13). Utilization of mannitol, sorbitol, erythritol, D-(−)-tartrate, and DL-lactate were determined with Misaghi and Grogan's (16) mineral medium, using Noble agar in place of Oxoid agar No. 3. Formation of β-glucosidase and sucrose utilization was also tested (8). Pectate degradation at pH 5 and 8.5 was determined by Hildebrand and Schroth's procedure (7). The bacterial strains were checked for Gram stain (17) and presence of flagella (4).

Pathogenicity tests. Florida Marble, White Marble, Polaris, and Beloved chrysanthemums were grown in the greenhouse from cuttings. Leaves were inoculated by the pinprick method (6) with 48-hr-old cultures at 10^8 colony-forming units per milliliter. Stems were inoculated by pricking with insect pins laden with the bacterium. Treated plants were placed under intermittent mist (10 sec/10 min/day) for 10 days. Isolation from artificially inoculated tissue was conducted to complete Koch's postulates.

RESULTS AND DISCUSSION

Symptoms generally became prominent 3 wk before harvest. Lesions frequently developed at the nodes (Fig. 1A) and extended up and down the stem. Early lesion development was characterized by water-soaking (Fig. 1A), whereas mature lesions were dark blue to black (Fig. 1B). Older tissue (lower stem area) was more heavily discolored than younger tissue but streaking did not necessarily extend to the crown. Severely affected plants died; others developed streaking that did not affect growth or flower development. The latter were harvested.

A fluorescent *Pseudomonas* sp. that induced a hypersensitive reaction in experimental plants was consistently isolated from the stem lesions of diseased field plants. It induced a hypersensitive reaction in pepper and was oxidase positive and arginine dihydrolase negative. Based on biochemical and nutritional tests (Table 1), the stem lesion isolates were identified as *Pseudomonas cichorii* (Swingle) Stapp and were biochemically indistinguishable from *P. cichorii* isolates from leaves. *Fusarium* spp. and other fungi were isolated inconsistently from the lesions.

The *P. cichorii* isolates from stems induced typical leaf spot symptoms on chrysanthemum leaves (Table 2, Fig. 2A). Stem inoculations resulted in duplication of the field symptoms (Table 2, Fig. 2B) with resulting water-soaking and slight browning of the tissue. The stem lesions later became dark brown to black.

P. cichorii is generally considered a leaf pathogen. It causes a leaf spot on geraniums (2), Gerbera (15), and lettuce (5) along with several other hosts and induces a leaf spot and bud blight of chrysanthemum (1,14). The bacterium has previously been shown to cause stem lesions and a bud blight of chrysanthemum (1,9,14). In two reports (1,14), however, the stem lesions were found to be of minor importance and were only observed in association with leaf spotting

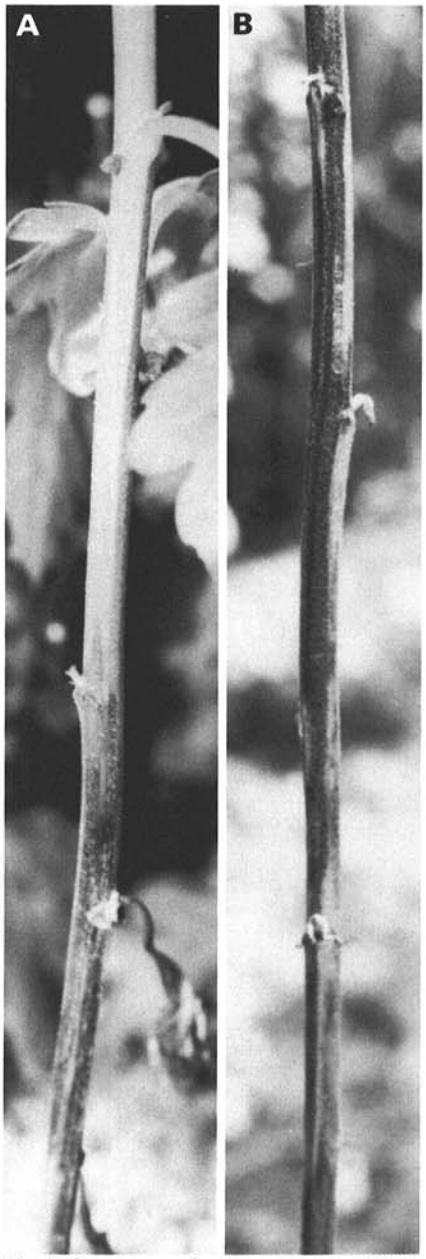

Fig. 1. Symptoms of stem necrosis on field-grown chrysanthemums incited by *Pseudomonas cichorii:* **(A)** water-soaking toward the margin of the lesion and **(B)** dark lesion development as the lesion matures.

Fig. 2. Symptoms on greenhouse-grown chrysanthemums inoculated with *Pseudomonas cichorii* isolated from chrysanthemum stems: **(A)** typical leaf spot development and **(B)** dark discoloration of the stem tissue.

Table 1. Physiological reactions of *Pseudomonas cichorii* strains from leaves and stems of chrysanthemum

Tests	Number of strains positive	
	Leaf[a] (3 strains tested)	Stem[b] (9 strains tested)
Oxidase	3	9
Arginine dihydrolase	0	0
Levan	0	0
Sucrose	1	2
β-Glucosidase	3	9
Pectate degradation		
pH 5	0	0
pH 8.5	0	0
Utilization of sorbitol	0	0
Erythritol	0	0
DL-Lactate	3	9
D-(−)-Tartrate	3	9
Mannitol	3	0

[a]Provided by J. Miller.
[b]One strain was provided by A. Chase.

Table 2. Pathogenicity of *Pseudomonas cichorii* strains on chrysanthemum

Strain	Origin	Cultivar inoculated	Virulence[a]	
			Stem	Leaf
5281-1	Stem	Florida Marble	+	+++
5264	Stem	White Marble	+++	+++
		Beloved	++	+++
5281-2	Stem	Florida Marble	+++	+++
		Polaris	++	+++
5303-1	Stem	Florida Marble	+++	+++
		Beloved	+++	++
5275-1	Stem	Florida Marble	+++	+++
		Polaris	+	+
5280-4	Stem	White Marble	++	+++
		Yellow Manatee	+++	+++
5280-2	Stem	Florida Marble	++	++
5280-3	Stem	White Marble	+++	++
		Yellow Manatee	+	+++
081-3263	Leaf	White Marble	++	++
		Polaris	+	++

[a]+ = Weakly virulent reaction; +++ = highly virulent reaction.

or bud blights where *P. cichorii* moved down the leaf petiole into the stem or down the peduncle into the stem. This report is the first of a stem necrosis observed without the associated bud blighting or leaf spotting.

It is not clearly understood why an epidemic of bacterial spot of chrysanthemums occurred where stem necrosis was the only observable symptom. One factor that might have been important in the epiphytotic was the unseasonably warm winter. Optimum temperature for disease development is 28 C (J. B. Jones, *unpublished*). Strain variation may have been responsible for the stem necrosis. Although an isolate from leaves of chrysanthemums induced stem lesions, its ability to cause stem lesions was less effective when compared with stem lesion isolates.

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