

# Fusarium Wilt of Chrysanthemum: Pathogen-Free Rooted Cuttings and Susceptibility of New Cultivars

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## ABSTRACT

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Rooted cuttings of 57 cultivars of chrysanthemums from a commercial propagator were culture-indexed and found to be free of the Fusarium wilt pathogen *Fusarium oxysporum* f. sp. *chrysanthemi*. The vast majority of cultivars were highly resistant to both *F. oxysporum* f. sp. *chrysanthemi* and f. sp. *tracheiphilum*; however, Excel, Foxy, Luv, and Fortune were highly susceptible and Applause, Circus, Remarkable, and Tempter were susceptible to *F. oxysporum* f. sp. *chrysanthemi*. Foxy was highly susceptible and Luv was susceptible to *F. oxysporum* f. sp. *tracheiphilum*.

Serious outbreaks of Fusarium wilt of chrysanthemum (*Chrysanthemum morifolium* (Ramat.) Hemsl.) caused by *Fusarium oxysporum* f. sp. *chrysanthemi* Litt., Armst., & Armst. were reported recently (3). The pathogen was found in 1.3–9.6% of rooted cuttings of cultivars both susceptible and resistant to Fusarium wilt. This was very alarming, since most growers rely on the availability of clean cuttings from a culture-indexing program (1) for control of this disease. The serious recurrence of this disease (2) has stimulated renewed interest in the development and use of resistant cultivars to control Fusarium wilt (3,5) and emphasizes the need for specialist chrysanthemum propagators to review

and improve their programs to produce cuttings free of vascular wilt pathogens.

The objectives of this study were to determine incidence of *F. oxysporum* f. sp. *chrysanthemi* in rooted cuttings and disease reactions of new chrysanthemum cultivars to the two Fusarium wilt pathogens, *F. oxysporum* f. sp. *chrysanthemi* and *F. oxysporum* f. sp. *tracheiphilum*.

## MATERIALS AND METHODS

Twenty rooted cuttings of each of 57 cultivars were examined by culture-indexing to determine if the pathogen was present in rooted cuttings. After the roots were removed, sections of lower stems were surface-sterilized with 0.5% sodium hypochlorite for 6 min, then cut into four 1-cm subsections and placed on sterile absorbent paper. Sections were placed on acidified cornmeal agar (ACMA), incubated at 24 C, and observed daily for 7 days (5). Isolates of the suspected pathogen were obtained and increased on ACMA, and 10 rooted cuttings of chrysanthemum cultivar Cirbronze were inoculated for each isolate by the root-dip technique described later for proof of pathogenicity. In addition, macroconidia from each section were examined for conformity to the characteristics of *F. oxysporum* (3).

To determine susceptibility of chrysanthemums to *F. oxysporum* f. sp.

*chrysanthemi* (FOC), cultivars were inoculated with a mixture of four isolates of FOC, two from North Carolina (NC 510 and NC 513) and two from Florida (FRC-O-693 and FRC-O-950). To determine susceptibility to *F. oxysporum* f. sp. *tracheiphilum* (E. F. Sm.) Snyder & Hans. race 1 (Armst. & Armst.) (FOT), cultivars were inoculated with two isolates of FOT (FRC-O-924 and FRC-O-1084) (5). Isolates were grown at 24 C for 2 wk on potato-dextrose agar (PDA) made from fresh potatoes in petri plates. The inoculum was prepared by blending contents of equal numbers of plates per isolate in sterile distilled water for 1 min. The inoculum was used immediately.

Cuttings, as received from the propagator, were inoculated by dipping the roots in a suspension of the inoculum containing about  $6 \times 10^5$  viable propagules per milliliter for 5 sec and transplanting one cutting per 10-cm-diameter clay pot into Metro Mix 220 (W. R. Grace Co., Cambridge, MA). Roots of control plants were dipped in sterile PDA suspension. Plants were misted for 5 sec each hour from 0800 to 1700 hours the first 2 days after transplanting to reduce the shock of transplanting. Each plant received Peters 20-20-20 soluble fertilizer (Robert Peters Co., Inc., Allentown, PA) each week, beginning 1 wk after transplanting. A 15:1 proportioner was used to apply the fertilizer solution (25 g/L = 350 ppm of nitrogen). In addition, 2 wk after transplanting, each plant received 5 cm<sup>3</sup> of 18-8-13 Osmocote controlled-release fertilizer (Sierra Chemical Co., Milpitas, CA). Plants were grown in the greenhouse from mid-August to mid-November without supplemental light and without pinching and were watered as needed with overhead sprinklers. Day and night temperatures averaged 34.1 and 20.5 C, respectively. Pots were set on benches of corrugated transite covered with wire

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screens to prevent spread of the pathogen during watering.

Plants were rated weekly for 8 wk for symptoms of *Fusarium* wilt according to the following severity index: 0 = apparently healthy; 1 = slight stunting (15–25% reduction of plant height) or slight distortion of foliage or flowers or slight chlorosis; 2 = slight wilt (one or two leaves affected) or moderate stunting (26–50% reduction of plant height) or obvious distortion of foliage or flowers or moderate, general chlorosis; 3 = wilt, moderate (three leaves to half of plant affected) or severe stunting (>50% reduction of plant height) or severe chlorosis (entire plant yellowed); 4 = wilt, severe (more than half of plant affected); and 5 = dead, whole plant irreversibly wilted, stems necrotic (5).

The heights of inoculated plants were measured and compared with those of uninoculated plants of the same cultivar as an index of stunting. Ten replicates of each cultivar-isolate combination were used in a completely randomized block design.

## RESULTS AND DISCUSSION

*Fusarium* spp. with macroconidia and microconidia were isolated from 29 of the 57 cultivars examined (Table 1). Frequency of isolation within a cultivar ranged from 5 to 60% of the cuttings indexed. None of the isolates obtained was pathogenic on cuttings of Cirbronze. Inoculations with all four FOC isolates described resulted in severe wilt and death of test plants. The cuttings of these cultivars were free of FOC, indicating that specialist propagators have improved their culture-indexing programs during the past few years (5). However, these cultivars could not be determined to be free of FOT since the test cultivar for pathogenicity was Cirbronze, which is highly resistant to FOT (5). Plans are to assay chrysanthemum cultivars every few years for presence of the *Fusarium* wilt pathogens.

Cirbronze, Promenade, Puritan, Royal Trophy, Tuneful, and Yellow Delaware were evaluated earlier for susceptibility to *Fusarium* wilt (5), but they were included in this study to confirm the reliability of the inoculation method. Ratings of these six were similar to those reported earlier, except the height reduction of Royal Trophy and Yellow Delaware in this study (48 and 31%, respectively) was much less than reported previously (100 and 97%, respectively) (5). This difference may have been due to the differences in the day lengths when the tests were conducted. The previous test was performed during long days, whereas this test was initiated under long days and terminated under short days, which induced flowering of plants.

Of the 51 new cultivars examined for *Fusarium* wilt resistance, Excel, Foxy,

**Table 1.** Frequency of isolation of nonpathogenic *Fusarium* spp. from rooted cuttings of chrysanthemum cultivars and reactions of the cultivars to inoculation with *F. oxysporum* f. sp. *chrysanthemi*

Cultivar	Frequency of isolation of nonpathogenic <i>Fusarium</i> spp. (%)	Disease rating 8 wk after inoculation <sup>a</sup>	Susceptibility rating <sup>b</sup>
Accord	0	0.0	VR
Alert	0	0.0	VR
Applause	0	3.4	S/T
Aspen	0	0.0	VR
Ballerina	0	0.0	VR
Cambria	5	0.0	VR
Camelot	5	0.0	VR
Cavalcade	10	0.0	VR
Champ	0	0.0	VR
Charm	5	0.0	VR
Chardunnay	60	0.0	VR
Charlie	0	0.0	VR
Cirbronze	0	4.4	VS
Circus	0	3.9	S/T
Cymbals	0	0.0	VR
Debonair	5	0.0	VR
Dragon	10	0.0	VR
Echo	0	0.0	VR
Eclipse	20	0.0	VR
El Charo	45	0.0	VR
Excel	5	4.8	VS
Favor	0	0.0	VR
Fortune	30	4.1	VS/T
Foxy	5	4.6	VS <sup>c</sup>
Goldmine	5	0.0	VR
Grenadine	40	0.8	VR
Heirloom	0	0.0	VR
Luv	0	4.1	VS/T <sup>c</sup>
Marmalade	0	0.0	VR
Matador	0	0.0	VR
Mellow	0	0.0	VR
Pink Nova	0	0.0	VR
Promenade	5	1.1	R
Puritan	35	0.0	VR
Rawhide	0	0.0	VR
Red Belair	0	0.0	VR
Remarkable	20	2.6	S/T
Riot	0	0.0	VR
Ritz	55	0.0	VR
Royal Trophy	45	4.1	VS/T
Sequest	40	0.0	VR
Sequoia	35	0.0	VR
Shamrock	5	0.0	VR
Songster	0	1.5	R
Splendor	15	0.0	VR
Starfire	5	0.0	VR
Surf	60	0.0	VR
Surfine	5	0.0	VR
Tempter	15	3.8	S/T
Tuneful	0	0.2	VR
Venture	25	0.0	VR
Viking	0	0.0	VR
White Marble	0	0.0	VR
White Nova	0	0.0	VR
Yellow Cambria	0	0.0	VR
Yellow Delaware	5	3.1	S/T
Yellow Nova	0	0.0	VR

<sup>a</sup>Disease severity rated on a scale of 0–5, where 0 = apparently healthy; 1 = slight stunting (15–25% reduction of plant height) or slight distortion of foliage or flowers or slight chlorosis; 2 = slight wilt (one or two leaves affected) or moderate stunting (26–50% reduction of plant height) or obvious distortion of foliage or flowers or moderate, general chlorosis; 3 = wilt, moderate (three leaves to half of plant affected) or severe stunting (>50% reduction of plant height) or severe chlorosis (entire plant yellowed); 4 = wilt, severe (more than half of plant affected); and 5 = dead, whole plant irreversibly wilted, stems necrotic.

<sup>b</sup>Susceptibility was based on the following disease severity ratings: 0.0–1.0 = very resistant (VR), 1.1–2.0 = resistant (R), 2.1–4.0 = susceptible (S), and 4.1–5.0 = very susceptible (VS). In addition, cultivars rated S or VS but with less than 50% reduction in height were also rated tolerant (T). Values are the average of 10 replicates.

<sup>c</sup>Foxy (4.7) and Luv (2.8) were also rated very susceptible and susceptible, respectively, to *F. oxysporum* f. sp. *tracheiphilum*.

Luv, and Fortune were rated highly susceptible and Applause, Circus, Remarkable, and Tempter were rated susceptible to FOC (Table 1). Foxy was rated highly susceptible, Luv was rated susceptible, and all other cultivars were rated highly resistant to FOT. Since Foxy was unique in being very susceptible to both FOC and FOT, this cultivar should be used in future pathogenicity studies with unknown *Fusarium* isolates from

chrysanthemum.

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