Additions to the Host Range of *Fusarium oxysporum* f. sp. *radicis-lycopersici*

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ABSTRACT

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Forty-seven species and 11 varieties within five of those species from 15 botanical families were inoculated by a root-dip technique with four isolates of *Fusarium oxysporum* f. sp. *radicis-lycopersici*. Random sections from roots and crowns of symptomless plants and root and crown sections displaying lesions from infected plants were surface-sterilized and placed on Komada's *Fusarium*-selective medium. Colonies formed on the selective medium were identified as *F. o.* f. sp. *radicis-lycopersici* with the use of a tomato seedling test on water agar. Thirty-seven of the 53 plant species and varieties were infected by at least one of the four *F. o.* f. sp. *radicis-lycopersici* isolates. Highly susceptible species, including *Lycopersicon esculentum*, were observed from the Solanaceae, Leguminosae, Cucurbitaceae, and Chenopodiaceae. Several symptomless hosts were identified.

Fusarium crown and root rot of tomato (FCRR) was first identified on tomatoes grown in greenhouses in Japan in 1969 (17). The causal agent was identified as a new race (J3) of *Fusarium oxysporum* Schlecht. f. sp. *lycopersici* (Sacc.) Snyd. & Hans., which causes Fusarium wilt of tomato (17,20). However, Jarvis and Shoemaker (7) proposed that the causal agent of FCRR was not a new race of *F. o.* f. sp. *lycopersici* but a new forma specialis of *F. oxysporum* and designated the fungus as *F. oxysporum* Schlecht. f. sp. *radicis-lycopersici* Jarvis & Shoemaker. They based their conclusions on the following characteristics: 1) the FCRR organism has distinctly different symptoms than those caused by *F. o.* f. sp. *lycopersici*, 2) *F. o.* f. sp. *lycopersici* rapidly moves through the host vascular tissue, while *F. o.* f. sp. *radicis-lycopersici* lacks movement through the vascular system of the stem, and 3) the optimum temperature for FCRR is around 18 C while Fusarium wilt of tomato is favored by temperatures around 27 C. Further differences between the FCRR organism and *F. o.* f. sp. *lycopersici* include: 1) *Meloidogyne incognita* (Kofoid & White) Chitwood predisposes tomato plants to wilt caused by *F. o.* f. sp. *lycopersici* (2,10) but has no association with *F. o.* f. sp. *radicis-lycopersici* (6), 2) nitrate suppresses wilt symptoms of *F. o.* f. sp. *lycopersici* as compared to ammonium (11,19) but nitrate and ammonium similarly affect the incidence of *F. o.* f. sp. *radicis-lycopersici* (8), 3) residues on the surface of microconidia of *F. o.* f. sp. *lycopersici* and *F. o.* f. sp. *radicis-lycopersici* differ (3), and 4) the host range of *F. o.* f. sp. *radicis-lycopersici* is larger than for *F. o.* f. sp. *lycopersici* (15,20). When 17

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plant species were inoculated with different isolates of the FCRR organism, various species of the Leguminosae, as well as *Lycopersicon esculentum* Mill. (cultivar Ohio MR-13), were infected (15). *F. o.* f. sp. *lycopersici* is specific to *Lycopersicon* spp. (18).

The objectives of this study were to confirm that *F. o.* f. sp. *radicis-lycopersici* is pathogenic to species of the Leguminosae as well as *Lycopersicon esculentum* (15) and to further study its host range.

MATERIALS AND METHODS

Four isolates of *F. o.* f. sp. *radicis-lycopersici* obtained from different greenhouses in different years in British Columbia and southern Ontario, Canada, were used in these experiments.

Forty-seven plant species and 11 varieties within five of the species from 15 botanical families were inoculated with the *F. o.* f. sp. *radicis-lycopersici* isolates (Table 1). Seeds of each species were surface-sterilized for 5 min in 0.5% of NaOCl, followed by two 5-min rinses in sterile, distilled water, and then planted in 10-cm-diameter plastic pots filled with sterile sawdust. The sawdust originated from a mixture of Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco) and western hemlock (*Tsuga heterophylla* (Raf.) Sarg.) wood which is recommended for use as a growth medium for greenhouse vegetables in British Columbia (1). The sawdust was autoclaved twice, for 1 hr each time to sterilize the medium. The seedlings were watered as needed with a tomato feeding formula for sawdust culture (1). When the seedlings reached the first true leaf stage, they were gently removed from the sawdust and the roots were dipped in 100 ml of a 10^5 microconidial suspension of *F. o.* f. sp. *radicis-lycopersici* per milliliter of water for a minimum of 30 min. The seedlings were then replanted in their pots with the original sawdust media. Larger pots and additional sterilized sawdust were used when necessary. Three to five seedlings of each plant species were inoculated with each of the four isolates of *F. o.* f. sp. *radicis-lycopersici* unless otherwise noted in Table 1. The inoculated seedlings were incubated in the greenhouse at 20–30 C. Because all of the plant species could not be tested simultaneously, tests were conducted on sets of five to 15 plant species at a time, with tomato seedlings of a susceptible cultivar included in each set. Four wk after inoculation, the inoculated plants were gently uprooted and the crown and roots examined for signs of infection. Visible lesions were cut from the roots and crowns, surface-sterilized for 5 min in 0.5% of NaOCl, and placed on Komada's selective medium for *Fusarium* spp (14). If lesions were not observed on the roots and crowns, random parts of the root and crown sys-

Table 1. The compatibility of four isolates of *Fusarium oxysporum* f. sp. *radicis-lycopersici* with different plant species

Scientific classification of plant species	Common name Cultivar[b]	Isolate number[a] 1	2	3	4
Solanaceae					
Lycopersicon esculentum Mill.	Tomato				
	Dombello	*	*	o	*
	Larma	−	o	o	−
	Dombito	*	*	*	*
	Oregon II	*	*	*	*
	W848139	*	*	*	*
	OE2797F1VF	*	*	*	*
Solanum tuberosum L.	Potato	−	−	−	−
Capsicum frutescens L.	Pepper				
	Lattini	+	+	o	+
Leguminosae					
Vicia faba L.	Broad bean				
	Small Pod	*	*	*	*
	Banner	*	*	*	*
Phaseolus vulgaris L.	String bean				
	Green	*	*	*	*
	Burgundy	*	*	*	*
	Black Wax	*	*	*	*
	Pinto bean	*	*	*	*
Pisum sativum L.	Pea				
	Little Marvel	−	+	+	+
	Sugar Snap	+	+	+	+
Trifolium pratense L.	Red clover	*	*	*	*
Trifolium repens L.	White clover	*	*	*	*
Melilotus alba Medik.	White sweet-clover	−	−	−	−
Cruciferae					
Brassica juncea L.	Taisei greens	+	−	+	+
B. napus L.	Canola	−	−	−	−
B. napus L. var. *napobrassica* (L.) Rchb.	Rutabaga	−	−	−	−
B. oleracea L. var. *botrytis* L.	Cauliflower				
	Dok Elgon	+	−	−	+
	Winter	+	+	−	−
B. oleracea L. var. *capitata* L.	Cabbage				
	Stokes II EC Chinese	+	+	−	+
	Sphinx	−	−	−	−
	Fornax	−	−	−	s
B. oleracea L. var. *gemmifera* DC.	Brussels Sprouts				
	Porter	+	+	+	+
	Rampart	+	+	+	−
B. oleracea L. var. *gongylodes* L.	Kohlrabi	+	+	+	+
B. oleracea L. var. *italica* L. Plenck	Broccoli				
	Cape Queen	−	−	−	−
	Bonanza	s	−	−	−
Capsella bursa-pastoris (L.) Medik.	Shepherd's-purse	−	−	−	s
Raphanus sativus L.	Radish				
	Miura Cross	+	−	+	+
	April Cross	−	+	+	−
Umbelliferae					
Anethum graveolens L.	Dill	−	−	−	−
Apium graveolens L. var. *dulce* (Mill.) Pers.	Celery				
	Hurst	+	−	+	+
	Tall Utah 5270R	+	+	+	+
	Tall Utah 5270HK	−	−	−	−
Daucus carota L.	Carrot				
	Dess Dan	−	−	−	−
	Chautenay	+	−	+	+
Foeniculum vulgare Mill.	Fennel				
	Domino	−	−	−	−
	Zefa Tardo	−	−	−	−
Plantaginaceae					
Plantago lanceolata L.	Narrow Leaved Plantain	s	s	o	o

(*continued on next page*)

[a]Isolates of *Fusarium oxysporum* f. sp. *radicis-lycopersici* obtained from different greenhouses in different years in southern Ontario and British Columbia. Rating system: * = *F. o.* f. sp. *radicis-lycopersici* recovered from root system, lesions large and roots severely affected; + = *F. o.* f. sp. *radicis-lycopersici* recovered from root system, lesions small with only a slight discoloration of the roots; s = *F. o.* f. sp. *radicis-lycopersici* recovered from root system, lesions not evident; − = *F. o.* f. sp. *radicis-lycopersici* not recovered from root system, lesions not evident; and o = *F. o.* f. sp. *radicis-lycopersici* infection not attempted.

[b]Indented names under the common name of the plant are the names of the cultivars tested, if known.

Table 1. *(continued from preceding page)*

Scientific classification of plant species	Common name Cultivar[b]	Isolate number[a] 1	2	3	4
Cucurbitaceae					
Cucumis metuliferus E.H. Mey ex. Naudin	Horned Cucumber	*	*	*	*
C. sativus L.	Cucumber				
	Stokes 56NK	*	*	*	*
	Wisconsin MR18	*	*	*	*
	Forona	–	–	o	–
Cucurbita pepo L.	Zucchini				
	Select	–	–	–	–
Compositae					
Galinsoga ciliata (Raf.) Blake	Hairy Galinsoga	+	+	+	+
Lactuca sativa L.	Lettuce				
	Kelly's	–	s	–	s
	NY12	–	–	–	s
	Butter Crunch	–	–	o	–
Rudbeckia hirta L.	Rudbeckia	–	s	–	–
Senecio vulgaris L.	Common Groundsel	–	–	–	–
Tagetes patula L.	French Marigold	–	–	–	–
Caryophyllaceae					
Stellaria media (L.) Vill.	Chickweed	+	+	o	o
Spergula arvensis L.	Corn Spurry	+	+	o	o
Polygonaceae					
Rumex crispus L.	Curly Dock	+	+	+	+
Polygonum convolvulus L.	Wild Buckwheat	+	+	o	+
Chenopodiaceae					
Beta vulgaris L. subsp. *cicla* L.	Chard	*	*	*	*
B. vulgaris L.	Beet				
	Detroit	*	*	*	*
	Monopoly	*	*	*	*
Chenopodium album L.	Lamb's-quarters	–	–	–	–
Spinacia oleracea L.	Spinach				
	Melody	*	*	*	*
Amaranthaceae					
Amaranthus retroflexus L.	Redroot Pigweed	–	+	+	+
Actinidiaceae					
Actinidia deliciosa (A. Chev.) C.F. Liang et A. R. Ferguson var. *deliciosa*	Kiwi				
	Hayward	–	–	–	–
Liliaceae					
Asparagus officinalis L.	Asparagus				
	Viking	+	+	+	+
Allium ampeloprasum var. *porrum* L.	Leek				
	Titan	–	–	–	–
	Durabel	–	–	–	–
Allium cepa L.	Onion				
	Norstar	+	+	+	+
	Klondike	+	+	+	+
Poaceae					
Avena sativa L.	Oat				
	Cascade	+	–	–	–
Echinochloa crusgalli (L.) P. Beauv.	Barnyard grass	–	–	–	–
Festuca ovina L.	Sheep Fescue				
	Bighorn	–	–	s	–
Hordeum vulgare L.	Barley				
	Klondike	+	+	+	+
Lolium perenne L.	Perennial Ryegrass				
	Palmer	–	–	–	–
Secale cereale L.	Fall Rye				
	Musketeer	+	–	+	–
Triticum aestivum L.	Wheat				
	Norstar	–	+	–	+
Zea mays L.	Corn				
	Dekalb	–	–	–	–
	Pioneer	–	–	–	–

tems were surface-sterilized and placed on Komada's medium. Colonies formed on Komada's medium were identified as *F. o.* f. sp. *radicis-lycopersici* through a seedling test on water agar (16).

RESULTS AND DISCUSSION

Thirty-seven of the 53 species and varieties tested in these experiments were infected by at least one of the four isolates of *F. o.* f. sp. *radicis-lycopersici* (Table 1). Ten species were highly susceptible, including *L. esculentum* from the Solanaceae and species from the Leguminosae, Cucurbitaceae, and Chenopodiaceae. Some of the species that were infected by *F. o.* f. sp. *radicis-lycopersici* in this study were not observed to be infected in previous host range studies (15,20). *Cucumis sativus* L. was not infected by the isolates of *F. o.* f. sp. *radicis-lycopersici* used in previous studies, but the two field cultivars of *C. sativus* tested in our experiments were highly susceptible to the four isolates of *F. o.* f. sp. *radicis-lycopersici* (Table 1). However, the long English greenhouse cucumber cultivar Forona was not infected by *F. o.* f. sp. *radicis-lycopersici* in our experiments. Differences in cucumber cultivar susceptibility may explain differences in the results of our work and the earlier studies (15,20). The *F. o.* f. sp. *radicis-lycopersici* isolates used in our study also infected cabbage (*Brassica oleracea* L. var. *capitata* L.), radish (*Raphanus sativus* L.), and wheat (*Triticum aestivum* L.) (Table 1), but these plants were not observed to be infected by *F. o.* f. sp. *radicis-lycopersici* in earlier studies (15,20). These differences between studies may also be attributable to differences in cultivars or inoculation techniques. The host range studies of Rowe (15) were conducted by soaking seeds of the host range plants in microconidia suspensions of 10^6/ml of each isolate of *F. o.* f. sp. *radicis-lycopersici* and then planting the seeds in soil infested with 10^5 to 10^7 chlamydospores per gram of soil. A comparison of Rowe's inoculation technique and our root-dip technique was not conducted.

The presence of small lesions with only a slight discoloration of the roots on the plant species with the "+" designation in Table 1 reflect, at best, a weak pathogenesis by *F. o.* f. sp. *radicis-lycopersici*. It may be that these plant species would not have been observed to be susceptible if lower concentrations of microconidia were used to inoculate the host range plants. However, when tomato seedlings of the cultivar Dombello were inoculated with the use of a root-dip technique and concentrations of microconidia of *F. o.* f. sp. *radicis-lycopersici* of 10^3, 10^4, 10^5, 10^6, and 10^7 per millileter of water, a concentration of 10^5 microconidia per millileter was found to be the lowest concentration that gave consistent infection of the tomato seedlings (Klaver, Menzies, and Koch, *unpublished*). Also, populations of *F. o.* f. sp. *radicis-lycopersici* can be as concentrated as 10^5 to 10^7 propagules per gram of medium when inoculated to steam-disinfested soil (15) or steam-sterilized sawdust (Menzies and Koch, *unpublished*).

It has been suggested that *F. o.* f. sp. *radicis-lycopersici* may be a low-grade pathogen associated with many plants in the field but not causing damage until it is present with plants growing under suboptimal temperatures in recently disinfested soil (15). Our results would support this statement (Table 1), and this may explain the spread of this pathogen onto susceptible crops and into new areas. The broad host range of *F. o.* f. sp. *radicis-lycopersici* containing both "symptomless" hosts and hosts displaying only mild symptoms of infection (i.e., a slight discoloration of the roots) suggests that these host species may act as reservoirs of inoculum, supporting the pathogen until highly susceptible hosts are available for infection. Also, the spread of the pathogen over short and long distances on symptomless or mildly infected transplants, in compost, (5) or in soilless media (4) would help to explain the rapid spread of this pathogen throughout the world. Symptomless hosts or carriers have also been reported for *F. o.* f. sp. *lycopersici* (13).

The infection of lettuce and spinach by our isolates of *F. o.* f. sp. *radicis-lycopersici* (Table 1) was unexpected considering that control of FCRR can be obtained by interposing a lettuce or spinach crop between successive crops of tomato in greenhouses (9). It has been suggested that the decomposition of these residues may act by releasing allelochemicals into the soil, which decreases infection in some way, rather than decreasing the population of the pathogen (9,12). It is possible that while the roots of these hosts are susceptible to *F. o.* f. sp. *radicis-lycopersici*, the decomposition of the crop residues creates an environment that is inhospitable to growth or survival of the pathogen.

The results of this host range study further support the designation of the new forma specialis, *radicis-lycopersici*, for the FCRR fungus, because unlike *F. o.* f. sp. *lycopersici*, it is pathogenic to *Lycopersicon* spp. and other host species within and without the Solanaceae (5,7,15,20,Table 1).

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