Effects of *Fusarium* Inoculum Density and Root-Knot Nematodes on Wilt Resistance in Summer Squash

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ABSTRACT

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Resistance of summer squash (*Cucurbita pepo* var. *melopepo*) cultivars to *Fusarium oxysporum* f. sp. *niveum* (*F. o. niveum*) was dependent on the concentration of initial inoculum and, in one cultivar, on the presence of root-knot nematode (*Meloidogyne incognita*). Four cultivars (Hyfric, Golden Eagle, Straightneck, and Early Prolific Straightneck) showed increasing susceptibility to Fusarium wilt as the concentration of *F. o. niveum* increased from 10^3 to 10^6 microconidia per milliliter. Cultivars Crookneck and Goldneck were unaffected by increasing levels of *F. o. niveum*. With the moderately wilt-resistant cultivar Early Prolific Straightneck, concomitant infection with *M. incognita* increased both the earliness of symptom development and the total wilt incidence. Resistance in the highly Fusarium wilt-resistant cultivar Goldneck was unaffected by the root-knot nematode. All squash cultivars tested were equally and highly susceptible to *M. incognita* on the basis of degree of root-galling and egg-mass production.

Several cultivars of summer squash (*Cucurbita pepo* var. *melopepo* Alef.) are susceptible to a vascular wilt caused by *Fusarium oxysporum* Schlecht. f. sp. *niveum* (E. F. Sm.) Snyd. & Hans. (*F. o. niveum*), incitant of Fusarium wilt of watermelon (10). Reactions of squash cultivars to *F. o. niveum* range from nearly immune to highly susceptible (10).

Although resistance to *F. o. niveum* in watermelon cultivars has been reported (3,6,12), this resistance is affected by the *F. o. niveum* inoculum concentration and by concomitant infection by the root-knot nematode (*Meloidogyne incognita* (Kofoid & White) Chitwood). Resistance to Fusarium wilt in watermelon cultivars with slight to moderate levels of wilt resistance can be overcome with increasing *F. o. niveum* concentration (11,13). Infection of *F. o. niveum*-resistant watermelon cultivars by *M. incognita* also resulted in increased susceptibility of those cultivars to Fusarium wilt (14). Resistance in certain cultivars classified as highly resistant, however, was unaffected by increased concentrations of *F. o. niveum* inoculum (11).

The objectives of this study were to determine, independently, the effects of increasing concentrations of *F. o. niveum* inoculum on wilt resistance in selected squash cultivars and the effects of concomitant infection by *M. incognita* on wilt resistance. Additionally, although squash has been reported as a host for *M. incognita* (15), only a limited number of cultivars have been tested directly for resistance. Therefore, selected squash cultivars varying in susceptibility to *F. o. niveum* were also tested for their reactions to *M. incognita*. An abstract of a portion of this work has been published (5).

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MATERIALS AND METHODS

The population of *M. incognita* used was originally isolated from cotton and maintained on Rutgers tomato. The *F. o. niveum* inoculum was originally obtained from the American Type Culture Collection (ATCC 18467) and subsequently maintained in sterile soil culture. Seed of the six squash cultivars were obtained from commercial sources.

The effect of *F. o. niveum* inoculum on resistance to wilt was tested in greenhouse experiments as described previously (10). Seed of the six squash cultivars tested were germinated in flats containing a pasteurized loamy sand-peat-perlite soil mix (1:1:1, v/v). At the first-true-leaf stage, uniform seedlings were uprooted, root-dip-inoculated with *F. o. niveum*, and transplanted in 3.8-L pots (five seedlings per pot) containing the same soil mix. *F. o. niveum* inoculum was predominantly microconidia obtained from liquid broth cultures (7) adjusted to

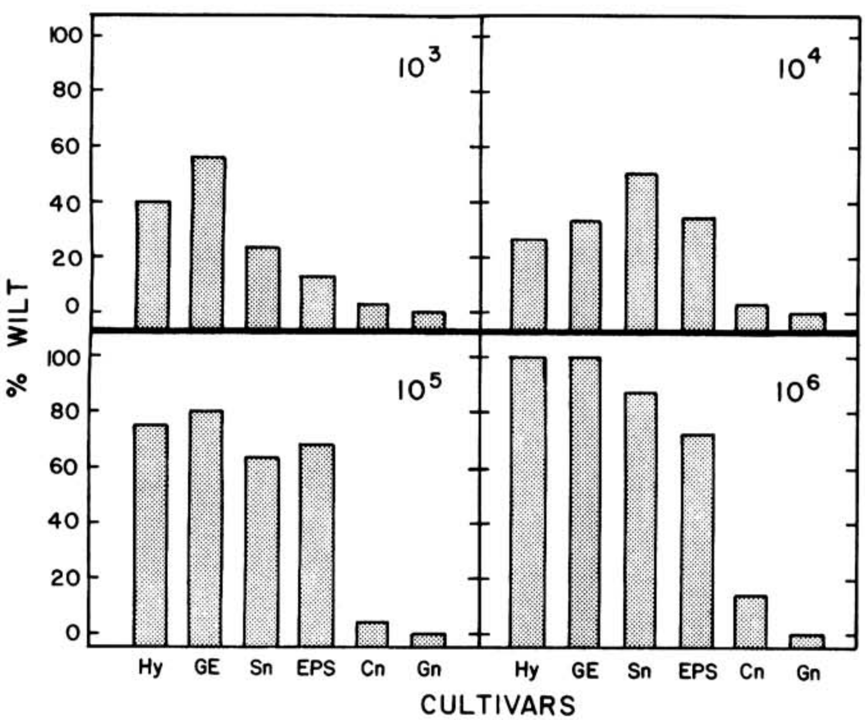

Fig. 1. Percent wilt of summer squash cultivars 3 wk postinoculation with increasing concentrations (10^3–10^6 microconidia per milliliter) of *Fusarium oxysporum* f. sp. *niveum*. Hy = Hyrific, GE = Golden Eagle, Sn = Straightneck, EPS = Early Prolific Straightneck, Cn = Crookneck, and Gn = Goldneck.

10^3, 10^4, 10^5, or 10^6 microconidia per milliliter. Wilt symptom development was monitored for 3 wk and the plants were evaluated for resistance as described previously for watermelons (11): highly resistant (≤20% wilt), moderately resistant (21–50% wilt), slightly resistant (51–80% wilt), and susceptible (>80% wilt). There were six replicated pots of each inoculum concentration for each cultivar tested.

Four summer squash cultivars were evaluated for susceptibility to *M. incognita*. Five seeds of each cultivar were planted in 1.6-L pots filled with a pasteurized loamy sand-peat soil mix (6:1, v/v), and seedlings were thinned to three per pot. Ten days after emergence of seedlings, each pot was infested with 10^4 *M. incognita* eggs by pipetting a suspension of eggs into plastic tubes (0.5 mm i.d.) inserted in the soil at the base of each seedling (two tubes per seedling). Nematode inoculum was collected from infected tomato roots by the NaOCl extraction method (8). Plants were maintained in a greenhouse after inoculation. After 6 wk, roots were harvested, washed gently with tap water, and rated for root-galling and egg-mass production. A scale of 0–5 was used to independently rate both galling and egg masses: 0 = no galling or egg masses and 5 = severe galling or > 100 egg masses per root system (2). Each cultivar treatment was replicated five times.

The effect of concomitant root-knot nematode infection on Fusarium wilt resistance was tested on the moderately wilt-resistant cultivar Early Prolific Straightneck and the highly wilt-resistant cultivar Goldneck. Squash seeds were germinated in flats, and 10 days after emergence, two seedlings were transplanted into 1.6-L pots containing the loamy sand-peat soil mix. Soil was infested with 10^4 *M. incognita* eggs by pouring a suspension of eggs into the transplant depressions in the soil before transplanting. *F. o. niveum* treatments were obtained by mixing 5 g of an *F. o. niveum*-infested sand-cornmeal medium (16) with each liter of the loamy sand-peat-soil mix before filling the pots. Dilution-plate counts established that the infested sand-cornmeal medium contained about 74,000 *F. o. niveum* colony-forming units per gram. Treatment combinations evaluated were 1) controls (sterile water plus sterile sand-cornmeal medium), 2) *M. incognita* alone, 3) *F. o. niveum* alone, and 4) *M. incognita* plus *F. o. niveum*. Plants were grown in the greenhouse and observed daily for wilt symptom development. There were 10 two-plant replicates of each treatment.

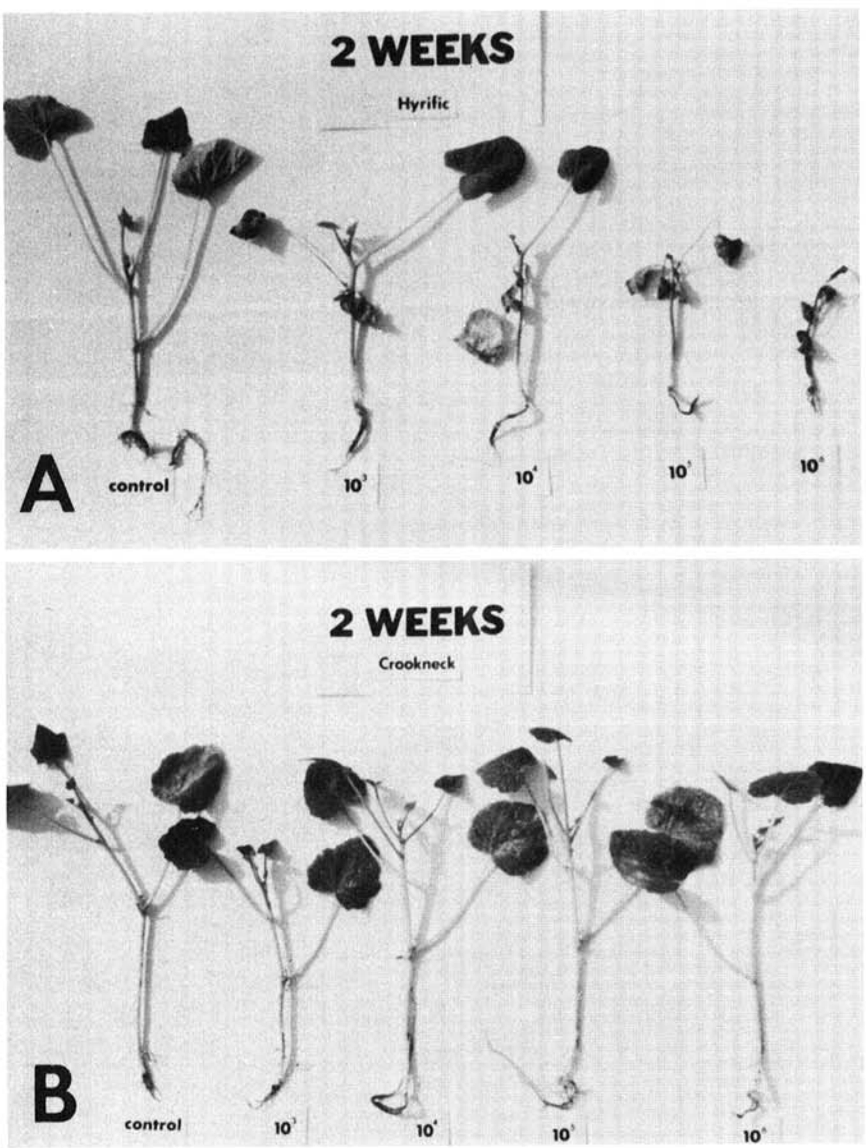

Fig. 2. Symptom development and wilt severity in **(A)** Hyfric and **(B)** Crookneck summer squash after 2 wk with increasing concentrations of *Fusarium oxysporum* f. sp. *niveum*. (Left to right) Concentration increases of 0, 10^3, 10^4, 10^5, and 10^6 microconidia per milliliter.

RESULTS

Reactions to increasing levels of *Fusarium*. Cultivars Straightneck and Hyrific were moderately resistant to *F. o. niveum* (23 and 40% wilt) and Golden Eagle was slightly resistant (58% wilt) at 10^3 microconidia per milliliter (Figs. 1 and 2). As inoculum concentrations increased, there was a general trend among these cultivars of increasing incidence of wilt development. At 10^6 microconidia per milliliter, all three cultivars were susceptible to *F. o. niveum* (>80% wilt). Early Prolific Straightneck showed a similar response to increased inoculum concentration, being highly resistant (13.3% wilt) at 10^3 microconidia per milliliter and only slightly resistant (72% wilt) at 10^6 microconidia per milliliter (Fig. 1). In contrast, resistance to *F. o. niveum* in the cultivars Crookneck and Goldneck was unaffected by increased inoculum concentration. These two cultivars remained highly resistant (≤20% wilt) at 10^6 microconidia per milliliter (Figs. 1 and 2).

Reactions to root-knot nematode. Cultivars Straightneck, Early Prolific Straightneck, Crookneck, and Goldneck all were susceptible to the nematode, showing root-gall indices of 3.5–3.8. Egg-mass indices were ≥4.0 for each cultivar.

Reactions to *Fusarium* and root-knot nematode. When the moderately resistant Early Prolific Straightneck was grown in the presence of both *F. o. niveum* and *M. incognita*, wilt symptoms were evident 14 days after transplanting. Plants in soil infested with *F. o. niveum* alone did not show wilt symptoms until 19 days after transplanting. Total wilt incidence was greater ($P = 0.05$) in the *M. incognita*-plus-*F. o. niveum* treatment (85%) than in the *F. o. niveum*-alone treatment (35% wilt) (Fig. 3). The highly resistant Goldneck did not show any wilt symptoms when exposed to *F. o. niveum* alone and 5% wilt when exposed to both *F. o. niveum* and *M. incognita* (Fig. 4). Both Early Prolific Straightneck and Goldneck were slightly stunted by *M. incognita*, and roots of both cultivars were heavily galled (root-gall index

>3.5). No wilt symptoms were evident on plants infected by *M. incognita* alone.

DISCUSSION

The susceptibility of selected summer squash cultivars to *F. o. niveum*, incitant of Fusarium wilt of watermelon, was reported recently by Martyn and McLaughlin (10). Although they evaluated 34 squash cultivars, they only used one inoculum level. In our study, we observed that the percent wilt of selected summer squash cultivars was affected by *F. o. niveum* inoculum concentration. Similar results were reported for resistance to *F. o. niveum* in watermelon cultivars (11,13). In general, squash cultivars with moderate to slight resistance to *F. o. niveum*, as defined for watermelons by Martyn and McLaughlin (11), showed increased susceptibility as the concentration of *F. o. niveum* increased from 10^3 to 10^6 microconidia per milliliter. Cultivars with high levels of resistance were unaffected by increased inoculum concentrations. In contrast to the differential resistance shown by the six cultivars to *F. o. niveum*, each of the four squash cultivars tested for their reactions to *M. incognita* were equally susceptible. The high root-gall and egg-mass indices obtained indicated that summer squash was susceptible to damage caused by *M. incognita* and supported high levels of nematode reproduction. These data are similar to a previous report (15) suggesting that summer squash is generally susceptible to *M. incognita*.

Our data suggest that the effect of *M. incognita* on resistance to *F. o. niveum* in squash is cultivar-specific. Incidence of wilt in Early Prolific Straightneck, which is moderately resistant to *F. o. niveum*, was increased by concomitant infection with *M. incognita*. Nematode infection both shortened the time required for expression of wilt symptoms and increased the incidence of wilt. However, Goldneck, which is highly resistant to *F. o. niveum*, was unaffected by concomitant *M. incognita* infection. These data are similar to those reported by Abawi and Barker (1) and Jones et al (9) for *F. oxysporum* f. sp. *lycopersici* (*F. o. lycopersici*) on tomato. Both groups reported that tomato cultivars with high levels of resistance to *F. o. lycopersici* were unaffected by concomitant infection by *M. incognita*. Abawi and Barker (1) also reported that wilt development was increased by *M. incognita* infection in tomato cultivars possessing only moderate levels of resistance to *F. o. lycopersici*. Bergeson (4) reported similar results with concomitant infection of *M. incognita* and *F. oxysporum* f. sp. *melonis* on muskmelon (*Cucumis melo*). His data indicated that the ability of *M. incognita* to break Fusarium wilt resistance varied among the different cultivars and resistance was most easily broken in a cultivar with incomplete resistance (4). These observations, coupled with the results in squash, suggest that although root-knot nematode infection can increase susceptibility of certain cultivars to Fusarium wilt, this ability is not uniformly consistent throughout a given host species.

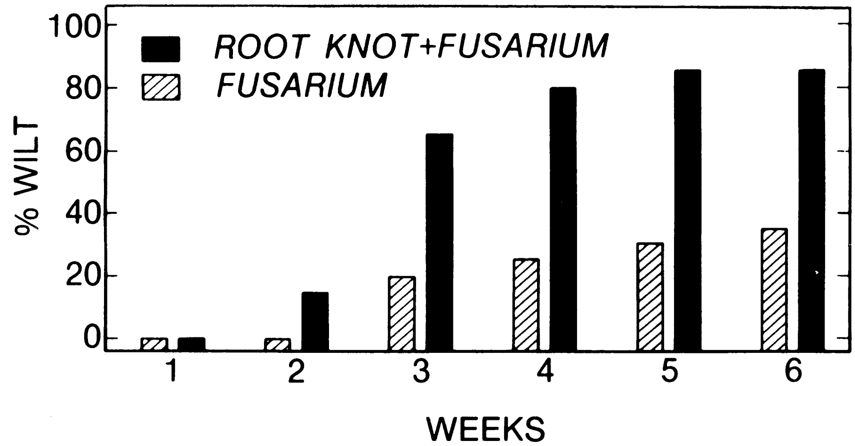

Fig. 3. Effect of *Meloidogyne incognita* on the resistance of Early Prolific Straightneck summer squash to Fusarium wilt (*F. oxysporum* f. sp. *niveum*).

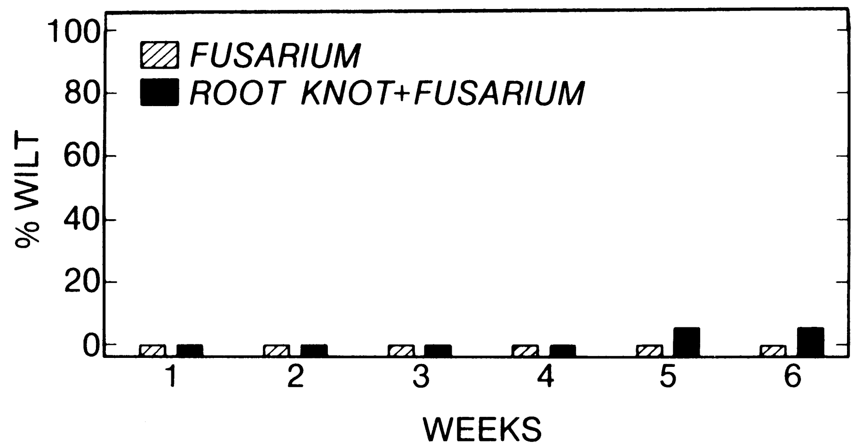

Fig. 4. Effect of *Meloidogyne incognita* on the resistance of Goldneck summer squash to Fusarium wilt (*F. oxysporum* f. sp. *niveum*).

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