Reaction of Citrus Rootstocks to Meloidogyne javanica

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Abstract: The response of *Citrus* spp. and related rootstocks to a population of *Meloidogyne javanica* was evaluated in a screenhouse experiment. Palestine and Rangpur lime, rough lemon, sour orange, Sexton and Thentriton tangelo, and Volkamer lemon were not infected by *M. javanica*. Galls and tip swellings were observed on the roots of *Poncirus trifoliata* and Troyer citrange. There was no evidence of nematode development. Symptoms induced by the nematode were stelar division, syncytia formation in the vascular tissues, and necrotic cells. *Key Words:* root-knot nematode, histopathology, *Poncirus trifoliata*, Troyer citrange.

Root-knot nematode infections on *Citrus* spp. are rare and of limited economic importance. Five *Meloidogyne* species—the Asiatic pyroid citrus nema, *M. exigua* Goeldi, *M. incognita* (Kofoid & White) Chitwood, *M. indica* Whitehead, and *M. javanica* (Treub) Chitwood—have been reported to infect citrus roots worldwide (1, 2, 3, 5, 6, 8, 10, 11). Nematode reproduction has been observed in citrus roots infected with the Asiatic pyroid citrus nema in India and Taiwan (1), *M. exigua* in Surinam (3) and Guadeloupe (8), *M. incognita* in Queensland, Australia (2), and *M. indica* in India (11). *Citrus* spp. are probably invaded by populations of these species that usually reproduce in other hosts.

Meloidogyne javanica has been reported on citrus more frequently than the other species of *Meloidogyne*. The parasite usually failed to complete its life cycle, which was attributed to a lack of syncytial formation following nematode penetration (7). This paper reports the host range of an *M. javanica* population on *Citrus* spp. and related genera, and histological changes caused by nematode invasion.

MATERIALS AND METHODS

The following *Citrus* species and related genera were used: sour orange (*Citrus aurantium* L.), rough lemon (*C. limon* [L.] Burm. f.), Palestine and Rangpur lime (*C. reticulata* var. *austera* Swing.), Volkamer lemon (*C. volkameriana* Pask.), *Poncirus trifoliata* (L.) Raf., Sexton and Thentriton tangelo (*C. paradisi* Macf. X *C. reticulata* Blanco), and Troyer citrange (*C. sinensis* [L.] Osb. X *P. trifoliata*).

In a screenhouse, 40 seeds of each species were sown in a bin, 11 x 2 x 0.5 m deep, filled with a volcanic sandy soil containing 80.7% sand, 11.3% silt, and 8.0% clay, naturally infested with about three eggs and juveniles of *M. javanica* per ml of soil. Seeds of each rootstock were randomly planted in four rows, each row alternating with rows of tomato (*Lycopersicon esculentum* Mill.) cv. Roma, the latter planted to maintain a high population density of root-knot nematodes. Tomato and citrus seeds respectively germinated in 10-15 and 25-30 days. Plants were grown under a Mediterranean climate in Sicily, from April to September 1976, and were given normal screenhouse maintenance. The tomato plants were pruned frequently to prevent excessive growth. The citrus and tomato roots were rated 5 months after the seed was sown. Galling indices for *M. javanica*-infected roots were: 0, none; 1, light; 2, moderate; 3, moderate to heavy; and 4, heavy (9). Root segments with galls were washed free of soil, fixed in FAA (formalin, acetic acid, alcohol) for 48 h, dehydrated in tertiary butyl alcohol, and embedded in paraffin. The 10-to-15-μm sections were stained in safranin-fast green, mounted in Permount, and observed with a compound microscope.

RESULTS AND DISCUSSION

No galls or swellings were found on the roots of any *Citrus* sp. Tip swelling and galls were present on the feeder roots of *P. trifoliata* and Troyer citrange. Gall indices ranged from 1 to 3 (Fig. 1). All tomato plants were severely galled (rating of 4) and had about 800 egg masses/g of fresh feeder root protruding from the root surface. Only juvenile stages were observed in galls of infected *P. trifoliata* and Troyer citrange seedlings.

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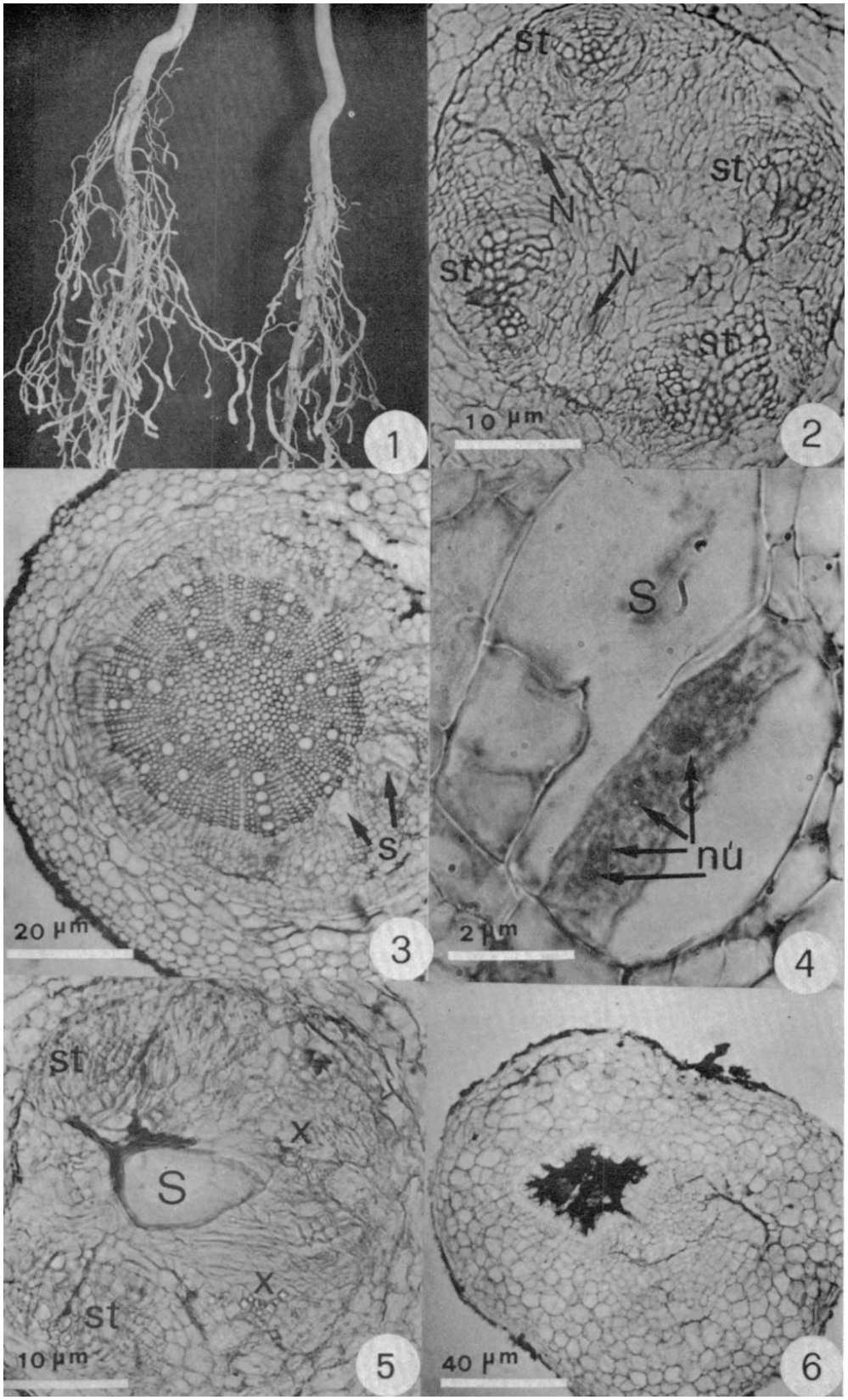
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Cross sections of infected feeder roots of *P. trifoliata* showed that juveniles penetrated the epidermis and cortex and fed on xylem parenchyma cells. The nematode feeding activity disorganized and divided the stele into several bundles, inducing syncytia with thickened walls in the primary tracheal elements (Fig. 5). Stelar division increased wherever several nematodes invaded the root at the same level, as observed in Troyer citrange (Fig. 2). In infected roots, the separated stelar portions were usually well delimited from each other by ground tissue, but sometimes groups of xylem cells were scattered in the ground tissue (Fig. 5). Observed in the secondary vascular tissue in all sections were several multinucleate syncytia, with granular cytoplasm and no evidence of thickening of secondary walls (Fig. 3, 4). The stelar area appeared eccentric because of hyperplasia in the vascular parenchyma (Fig. 3). A necrotic reaction in the cells surrounding the nematode head often occurred, preventing syncytial formation and larval development and causing eventual death of the nematode. When this necrotic reaction was accentuated, the entire stelar area dissolved and the original vascular tissue was very difficult to distinguish. A large necrotic spot remained in the root center (Fig. 6).

Similar root damage was observed in infected Troyer citrange feeder roots. The simultaneous invasion of several juveniles induced multiple stelar division. In root cross-sections, the nematodes appeared to be localized in the hyperplastic ground tissue among the divided stele (Fig. 2). No syncytia formation was observed in cross-sections of the root.

Stelar division with delayed nematode development has been reported in galls of *M. incognita*-infected *Lycopersicon pimpinellifolium* Mill. (4). *M. javanica* infection in *P. trifoliata* and Troyer citrange induced stelar division, but the nematode did not complete its life cycle. *Poncirus trifoliata* and Troyer citrange exhibited less galling than tomato, and nematode invasion induced syncytia in some instances. Stelar disorganization, which causes localized root swelling, always occurred before necrosis of the cells surrounding the nematode.

Because the juveniles failed to complete development in the roots of *P. trifoliata* and Troyer citrange, removing the plants from the source of infection should enable them to recover from nematode infection. That was shown when *P. trifoliata* and Troyer citrange seedlings infected with *M. javanica* were transplanted to clay pots containing clean soil. Eleven months later they evidenced no galling in the root system. Damage caused by nematode penetration into the feeder roots can be avoided by keeping *Citrus* and related genera separate from hosts susceptible to *Meloidogyne* sp.

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FIG. 1-6. *1)* Root-galls on Troyer citrange seedlings induced by *Meloidogyne javanica*. *2)* Cross-section of Troyer citrange root with divided stele (St) caused by *M. javanica* (N) infection. *3)* Cross section of *Poncirus trifoliata* feeder root showing syncytia (S) in the secondary vascular tissue. *4)* Multinucleate syncytium (S) in a cross section of *Poncirus trifoliata* feeder root; nu = nuclei. *5)* Cross-section of *Poncirus trifoliata* feeder root showing a syncytium (S) in primary tracheal elements surrounded by divided stele (St) and xylem cells (X). *6)* Cross-section of *Poncirus trifoliata* feeder root showing necrotic reaction of cells in the disorganized stelar area due to *M. javanica* infection.

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