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Host Suitability of *Ixora* spp. for the Root-knot Nematodes *Meloidogyne incognita* Race 1 and *M. javanica*[1]

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Abstract: Eight commonly cultivated *Ixora* species or cultivars were tested for their suitability as hosts and their level of tolerance to *Meloidogyne incognita* race 1 and *M. javanica* in a greenhouse study. Twenty weeks postinoculation with 5,000 eggs per pot, *M. incognita* race 1 and *M. javanica* produced galls and formed egg masses on roots of all eight *Ixora* species or cultivars tested. However, only *M. javanica*-infected 'Petite Yellow' and 'Maui' had decreases ($P \leq 0.05$) in root wet weights, suggesting that the other cultivars were more tolerant to these root-knot nematode species. Differential host suitability to each *Meloidogyne* species was based on the relative number of galls, galls per gram root weight, egg masses, and second-stage juveniles produced per plant. 'Bonnie Lynn,' 'Maui,' and 'Petite Red' were good to excellent hosts for both *Meloidogyne* spp. *Ixora coccinea* was a good host for *M. incognita* race 1 but less suitable for *M. javanica*. 'Singapore' and 'Petite Yellow' were poor hosts for *M. incognita* race 1 but excellent hosts for *M. javanica*. 'Nora Grant' and *I. casei* 'Super King' were poor hosts for both species of root-knot nematodes.

Key words: host-parasite relationship, *Ixora* spp., *Meloidogyne incognita* race 1, *M. javanica,* nematode, ornamental, root-knot nematode, woody ornamental.

The genes *Ixora* is a diverse group of perennial shrubs or trees with over 150 described species (6). It is mostly known from tropical regions of Asia, Africa, Australia, the Pacific Islands, and Central America, where it is often cultivated as a woody ornamental. In Florida and the Gulf States, several species and cultivars of *Ixora* have been introduced into landscapes for their attractive evergreen foliage and beautiful compact corymbs of small long-tubed red, orange, pink, yellow, or white flowers. This vegetatively propagated plant is easily grown in areas where night temperatures rarely go below 18 C. The most common forms of *Ixora* cultivated in Florida include *I. casei* Hance, *I. coccinea* L., and a plethora of crosses of *I. chinensis* Lam. × *I. coccinea.*

In southern Florida, mature specimens of certain cultivars of *Ixora* (e.g., *I. coccinea* cv. 'Maui') have been observed with severe chlorosis and formidable root galling, indicating susceptibility to local root-knot nematode populations (2). Knowledge of the differential host suitability and relative intolerance or resistance in available *Ixora* cultivars to root-knot nematodes would be valuable to landscapers and homeowners wishing to avoid postplant root-knot nematode problems. The two most common root-knot nematode species in southern Florida are race 1 of *Meloidogyne incognita* (Kofoid & White) Chitwood and *M. javanica* (Treub) Chitwood (R. A. Dunn, pers. comm.). The purpose of this study was to determine if differential host suitability and tolerance exists in eight commonly cultivated species and cultivars of *Ixora* to the root-knot nematodes *M. incognita* race 1 and *M. javanica.*

MATERIALS AND METHODS

Ixora propagation: Beginning 23 January 1991, eight accessions of *Ixora* were propagated for challenge with root-knot nematodes. Sixty cuttings were made of new wood from potted stock plants of each of the following named cultivars or species: 'Bonnie Lynn,' 'Maui,' 'Nora Grant,' 'Petite Red,' 'Singapore,' *I. coccinea,* and *I. casei* 'Super King' (= *I. duffii* 'Super King'). All but the newest four to six leaves were removed from the cuttings, and the stem (with three to five nodes) was trimmed to give a uniform starting weight for each

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cultivar. Each cutting was dipped into a 50% ethanol solution of indolebutyric acid (4×10^3 ppm) for 5 seconds and individually planted in 2.5 × 2.5 × 2.5-cm polyethylene cell packs filled with a 1:1 mixture of peat moss and perlite, which had been autoclaved for 1 hour at 122 C and 103 kPa. Cuttings were placed on a screened bench in a mist house for ca. 5 weeks and transferred to a 63% shadehouse with a daily 30-minute watering cycle for 2 weeks.

Seven weeks after starting the cuttings, the roots of each plantlet were washed free of the planting mixture and transplanted into two nested 8.0 × 8.0 × 7.5-cm plastic pots filled with (±5%) 350 g dry weight of autoclaved 60-mesh silica sand. The sand was confined to the pots by covering the holes in the bottom of the inner pot with 35-mesh Lumite Saran® screen (Chicopee Manufacturing Co., Cornelia, GA) and by placing two pieces of fiberglass felt between the two nested pots. Cuttings of Nora Grant did not root; thus, rooted plantlets of this cultivar and Petite Yellow were obtained (Parrish Nursery, Cooper City, FL). No phytoparasitic nematodes were detected from samples of soil or roots of these plants. The plant roots were washed free of the peat moss–perlite planting mixture, and plants were sized and weighed and then transplanted into pots as above. Each pot received 4.0 g of Osmocote (18-6-12; N-P-K) slow release fertilizer (6-month release formulation) and 0.5 g of Micromax Plus (Grace Sierra Fertilizer Corp., Milpitas, CA) at transplanting. Squares of PAK ground cover cloth (A. H. Hummert Seed Co., St. Louis, MO) were placed over the soil surface to prevent light penetration and growth of algae on the soil surface. Nematode inoculations were made on 24 April 1991, approximately 13 weeks after cuttings were initiated. Two weeks after inoculation, *Ixora* were placed in a different 63% shadehouse and watered daily for 30 minutes at 0600 hours and for 10 minutes each at 1130 and 1300 hours. All plants were arranged in a randomized complete block design. There were 10 replications for each of three treatments (*M. incognita* race 1, *M. javanica,* and an uninoculated control) for each of the eight cultivars or species of *Ixora.* Soil temperature was monitored daily (except weekends) at a 2.5-cm depth at 1400–1500 hours and ranged from 26–33 C during the 20-week experiment.

Nematode inoculum: Pure cultures of *M. incognita* race 1 and *M. javanica* were obtained on 'Rutgers' tomato plants from Dr. M. Oostendorp at the University of Florida, Gainesville. Populations of *M. incognita* race 1 were increased on 'California Wonder' pepper, and populations of *M. javanica* were increased on 'Black Beauty' eggplant using methods described previously (8). These two hosts were used because they are mutually exclusive for these two *Meloidogyne* species.

Eggs were harvested using a sodium hypochlorite method (8), and a total of 5,000 eggs were transferred into four depressions 2.5 cm from the stem of each plant. As an inoculum check, 'Rutgers' tomato plants (12 weeks old) were transplanted to 8.0 × 8.0 × 7.5-cm pots with autoclaved sand as above and were also inoculated on 24 April 1991 with 5,000 eggs each (six replicates for each *Meloidogyne* species). After 7 weeks, tomato roots were washed free of soil and stained for quantification of root-knot nematodes (8). Species identification was confirmed at the conclusion of the experiment by examining perineal patterns of randomly collected females.

Estimation of plant performance and nematode densities: Ixora plants were harvested starting at 20 weeks after inoculation. Because of the large amount of harvest time involved for each plant, only one block was harvested daily. Each plant was cut at the soil line and a top wet weight determined. Roots were rinsed of sand, patted dry, and wet weighed. Roots were soaked individually in a Phloxine B solution (0.15 g/liter tapwater) for 15 minutes for egg mass staining (1). Each root system was then examined under a dissecting microscope for the number of galls and the number of egg

masses present. After counting, gall and egg mass numbers were categorized separately for each root system using a 0–5 system (7) (0 = no galls or no egg masses, 1 = 1–2, 2 = 3–10, 3 = 11–30, 4 = 31–100, and 5 = >100). All of the sand and the root rinsate was mixed in a stainless steel bowl in about 2 liters of water and passed twice through a 500-mesh screen (25-μm openings). Nematodes and debris were placed onto Baermann funnels in an intermittent mist (1 minute mist for every 10 minutes) for 24 hours. The samples were examined to determine the number of second-stage juveniles (J2) present and to verify that control plants were not infected.

Statistics: Nematode count data were log-transformed ($\log_{10}$ [N + 1]) prior to analysis using the general linear model procedure in SAS (5). All plant performance data and gall and egg mass indices were not transformed. Means were separated with a Waller-Duncan k-ratio *t*-test with k = 100 (5).

Results and Discussion

No leaf chlorosis or above-ground disease symptoms were observed 20 weeks after inoculation for any of the nematode treatments or untreated controls. However, several plants died during the course of the experiment and were discarded. Root-knot nematode inoculation significantly affected plant performance in very few cases (Table 1). *Meloidogyne javanica* caused a reduction in root and total plant wet weight in 'Petite Yellow,' and a similar trend was observed in 'Maui' (Table 1). Overall, these results suggest that either 20 weeks under these artificial conditions is not enough time for above-ground manifestation of disease symptoms or that most of these cultivars are relatively tolerant of the two root-knot nematodes tested. In Florida, *Ixora* often exhibits severe chlorosis of the leaves during the winter and early spring. This experiment was conducted during the summer, and fertilizer and water were not limiting as they often are in the sandy soils of the southern Florida landscape. Thus, we suspect that during this experiment, the plants with heavy galling from *Meloidogyne* spp. inoculations (Tables 2,3) were spared from exhibiting symptoms. Long-term field experiments are needed to confirm whether most of these *Ixora* cultivars are indeed tolerant to the *Meloidogyne* species used in this study.

At 7 weeks, each of the tomato plants inoculated with *M. incognita* race 1 or *M. javanica* was severely galled (gall index = 5.0), indicating viable inocula. 'Maui' was the most suitable host for *M. incognita* race 1 showing the highest number of galls, galls per gram of root, and egg masses of all the *Ixora* cultivars tested (Table 2). This cultivar was also a suitable host for *M. javanica* (Table 3). 'Bonnie Lynn' and 'Petite Red' were very suitable hosts for both *M. incognita* race 1 and *M. javanica,* with gall and egg mass indices above 3.0 and mean numbers of J2 above 100 per pot (Tables 2,3). 'Singapore' and 'Petite Yellow' appeared to be marginal hosts for *M. incognita* race 1, whereas they were excellent hosts for *M. javanica* (Tables 2,3). In fact, the reduction in the root wet weight (Table 1) with concurrent galling suggests that 'Petite Yellow' is relatively intolerant to *M. javanica. Ixora coccinea* was a good host for *M. incognita* race 1 but not as good a host for *M. javanica.* 'Nora Grant' and 'Super King' were very poor hosts of *M. incognita* race 1, with means of only two egg masses per plant (Table 2). 'Super King' was also a very poor host for *M. javanica,* whereas 'Nora Grant' was only a marginal host (Table 3).

To our knowledge, there have been no evaluations of host resistance in *Ixora* to any phytoparasitic nematodes. This study demonstrates that there is differential host suitability for *M. incognita* race 1 and *M. javanica* among the eight *Ixora* species or cultivars we tested. 'Petite Yellow' appears to be intolerant to *M. javanica.* 'Super King' and 'Nora Grant' appear to be fairly resistant to both species of root-knot nematodes, with only marginal reproduction occurring. The rest of the *Ixora* culti-

TABLE 1. Plant performance measurements (mean ± standard deviation) 20 weeks after inoculation for eight *Ixora* cultivars inoculated with 5,000 eggs of *Meloidogyne* spp. per plant.

Species	Cultivar	Nematode treatment	Wet weight (g) of top	Wet weight (g) of bottom	Total wet weight (g)	N‡
(*I. coccinea*	'Bonnie Lynn'	*M. incognita*	40.4 ± 14.7 a†	22.8 ± 9.4 a	63.1 ± 22.9 a	10
× *I. chinensis*)		*M. javanica*	34.9 ± 19.8 a	17.1 ± 11.1 a	52.0 ± 29.5 a	10
		Uninoculated	47.4 ± 11.3 a	23.4 ± 8.8 a	70.8 ± 19.5 a	10
I. coccinea	'Maui'	*M. incognita*	42.7 ± 18.6 a	16.7 ± 7.1 a	59.5 ± 24.8 a	10
		M. javanica	40.7 ± 18.2 a	11.3 ± 6.1 b	52.0 ± 23.9 a	10
		Uninoculated	44.8 ± 15.3 a	15.8 ± 7.2 ab	60.6 ± 21.9 a	10
I. coccinea	'Nora Grant'	*M. incognita*	40.1 ± 18.8 a	16.9 ± 11.1 a	57.0 ± 29.1 a	10
		M. javanica	31.3 ± 16.1 a	11.3 ± 8.9 a	42.6 ± 24.4 a	10
		Uninoculated	37.6 ± 20.5 a	14.9 ± 11.7 a	52.4 ± 31.5 a	10
I. coccinea	'Petite Red'	*M. incognita*	17.1 ± 7.9 a	11.3 ± 9.5 a	28.4 ± 16.7 a	10
		M. javanica	13.9 ± 6.8 a	6.9 ± 3.9 a	20.8 ± 10.6 a	10
		Uninoculated	12.0 ± 9.3 a	8.3 ± 7.9 a	20.3 ± 16.7 a	8
I. coccinea	'Petite Yellow'	*M. incognita*	12.8 ± 4.7 a	5.1 ± 2.2 a	17.9 ± 6.9 a	10
		M. javanica	7.9 ± 3.2 b	2.9 ± 1.1 b	10.7 ± 4.1 b	10
		Uninoculated	10.1 ± 3.0 ab	4.8 ± 3.3 a	14.8 ± 5.7 a	10
I. coccinea	'Singapore'	*M. incognita*	31.1 ± 12.7 a	10.5 ± 4.8 a	41.6 ± 17.2 a	9
		M. javanica	42.1 ± 35.6 a	12.3 ± 6.1 a	54.6 ± 40.4 a	10
		Uninoculated	22.7 ± 6.3 a	5.9 ± 1.7 b	28.6 ± 7.9 a	10
I. coccinea	—	*M. incognita*	40.8 ± 9.6 a	18.4 ± 7.5 a	59.2 ± 16.1 a	7
		M. javanica	30.8 ± 21.9 a	12.9 ± 11.3 a	43.7 ± 33.0 a	7
		Uninoculated	43.2 ± 13.7 a	16.4 ± 6.8 a	59.5 ± 20.0 a	7
I. casei	'Super King'	*M. incognita*	46.9 ± 24.4 a	16.1 ± 8.6 a	63.1 ± 32.0 a	10
		M. javanica	47.7 ± 19.2 a	20.5 ± 10.9 a	68.2 ± 29.2 a	9
		Uninoculated	57.4 ± 27.1 a	20.1 ± 12.0 a	77.5 ± 38.3 a	9

† Values in a column for a specific *Ixora* cultivar with a letter in common are not different by the Waller-Duncan k-ratio *t*-test (k = 100) at $P < 0.05$.
‡ N = number of plants on which data were collected.

TABLE 2. Population estimates (mean ± standard deviation) of *Meloidogyne incognita* race 1 on eight *Ixora* cultivars at 20 weeks postinoculation with 5,000 eggs/plant.

Species	Cultivar	Galls/plant	Gall index†	Galls/g wet root weight	Egg masses /plant	Egg mass index†	J2/pot
(*I. coccinea* × *I. chinensis*)	'Bonnie Lynn'	109 ± 61 b‡	4.5 ± 0.7 a	4.9 ± 2.4 c	78 ± 54 b	3.9 ± 0.7 ab	131 ± 91 a
I. coccinea	'Maui'	329 ± 205 a	5.0 ± 0.0 a	20.0 ± 8.2 a	262 ± 190 a	4.7 ± 0.5 a	614 ± 882 a
I. coccinea	'Nora Grant'	7 ± 5 ef	2.0 ± 0.8 de	0.5 ± 0.4 d	2 ± 2 d	0.9 ± 0.9 e	4 ± 6 b
I. coccinea	'Petite Red'	77 ± 35 bc	4.3 ± 0.7 ab	9.7 ± 5.8 b	63 ± 38 b	3.9 ± 0.7 ab	110 ± 120 a
I. coccinea	'Petite Yellow'	16 ± 19 ef	2.2 ± 1.6 de	3.0 ± 3.5 cd	14 ± 16 c	1.9 ± 1.7 d	17 ± 26 b
I. coccinea	'Singapore'	24 ± 24 de	2.6 ± 1.3 cd	2.0 ± 1.6 cd	18 ± 20 c	2.3 ± 1.3 cd	7 ± 9 b
I. coccinea	—	58 ± 47 cd	3.4 ± 1.7 bc	3.4 ± 3.1 cd	43 ± 34 b	3.0 ± 1.4 bc	152 ± 184 a
I. casei	'Super King'	5 ± 5 f	1.4 ± 1.1 e	0.3 ± 0.3 d	2 ± 4 d	0.7 ± 0.9 e	2 ± 4 b

There were 10 replicates for each cultivar, except for missing values due to plant deaths in two cultivars ($N = 9$ for 'Singapore' and $N = 7$ for *I. coccinea*). Also, $N = 7$ for the J2/pot for all cultivars, except *I. coccinea*, where $N = 4$.

† 0 = no galls or egg masses, 1 = 1–2, 2 = 3–10, 3 = 11–30, 4 = 31–100, 5 = >100 galls or egg masses per plant.

‡ Values in a column with a letter in common are not different by the Waller-Duncan k-ratio *t*-test (k = 100) at $P < 0.0002$.

TABLE 3. Population estimates (mean ± standard deviation) of *Meloidogyne javanica* on eight *Ixora* cultivars at 20 weeks postinoculation with 5,000 eggs/plant.

Species	Cultivar	Galls/plant	Gall index†	Galls/g wet root weight	Egg masses /plant	Egg mass index†	J2/pot
(*I. coccinea* × *I. chinensis*)	'Bonnie Lynn'	59 ± 43 ab‡	3.6 ± 1.2 ab	3.5 ± 2.8 cd	41 ± 33 cd	3.2 ± 1.5 b	188 ± 220 a
I. coccinea	'Maui'	30 ± 20 bc	3.2 ± 0.8 bc	2.7 ± 1.5 cd	28 ± 18 c	3.3 ± 0.7 b	112 ± 131 a
I. coccinea	'Nora Grant'	21 ± 18 cd	2.5 ± 1.5 cd	3.0 ± 2.9 cd	9 ± 8 e	1.8 ± 1.1 c	9 ± 17 b
I. coccinea	'Petite Red'	60 ± 16 a	4.0 ± 0.0 ab	10.6 ± 5.8 b	30 ± 15 bc	3.2 ± 0.4 b	77 ± 112 a
I. coccinea	'Petite Yellow'	88 ± 45 a	4.4 ± 0.5 a	31.7 ± 14.1 a	73 ± 39 a	4.1 ± 0.6 a	100 ± 146 a
I. coccinea	'Singapore'	87 ± 44 a	4.4 ± 0.7 a	7.1 ± 3.5 bc	62 ± 37 ab	3.8 ± 0.6 ab	259 ± 284 a
I. coccinea	—	26 ± 23 cd	2.6 ± 1.4 cd	3.7 ± 5.0 cd	15 ± 11 de	2.3 ± 1.1 c	19 ± 33 b
I. casei	'Super King'	10 ± 10 d	1.9 ± 0.9 d	0.8 ± 1.1 d	3 ± 5 f	1.0 ± 1.1 d	6 ± 6 b

There were 10 replicates for each cultivar, except for missing values due to plant deaths in two cultivars ($N = 9$ for 'Super King' and $N = 7$ for *I. coccinea*). Also, $N = 7$ for the J2/pot for all cultivars, except *I. coccinea* where $N = 4$.

† 0 = no galls or egg masses, 1 = 1–2, 2 = 3–10, 3 = 11–30, 4 = 31–100, 5 = >100 galls or egg masses per plant.

‡ Values in a column with a letter in common are not different by the Waller-Duncan k-ratio *t*-test (k = 100) at $P < 0.0002$.

vars or species in this study seem to be tolerant to *M. incognita* race 1 and *M. javanica,* but this needs to be confirmed in the field, especially during the winter and early spring when *Ixora* spp. are prone to exhibit leaf chlorosis in southern Florida. It is quite likely that 'Maui' and the other *Ixora* cultivars that exhibited extensive galling when challenged with *Meloidogyne* spp. could be at a higher risk for negative interactions with plant pathogens, such as *Fusarium oxysporum* Schlechtend, which has recently been isolated from *I. coccinea* in Florida (4).

As noted by McSorley and Dunn (3), perennials that are tolerant to a particular species of nematode should be avoided when intolerant perennial or annual ornamentals will be planted nearby. In essence, the tolerant plant becomes a reservoir for nematodes, which can attack the intolerant plant. It appears that some of the *Ixora* cultivars we tested may potentially function this way in long-term plantings. For example, 'Bonnie Lynn,' 'Maui,' 'Petite Red,' and *I. coccinea* produced large numbers of egg masses and yielded relatively large numbers of J2 of *M. incognita* race 1 (Table 2), whereas 'Bonnie Lynn,' 'Maui,' 'Petite Red,' 'Petite Yellow,' and 'Singapore' were very productive hosts for *M. javanica* juveniles.

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