HOST STATUS OF WOODY ORNAMENTAL PLANTS NATIVE TO SOUTHEASTERN U.S.A. TO THREE *MELOIDOGYNE* SPECIES¹

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ABSTRACT

Sharma, J., and J. R. Rich. Host status of woody ornamental plants native to Southeastern U.S.A. to three *Meloidogyne* species. Nematropica 35:23-30.

Use of indigenous (native) plants in landscapes is of interest because of their perceived low maintenance requirements, and possible resistance to pests including plant-parasitic nematodes. Meloidogyne spp. (root-knot nematodes) damage many ornamental landscape plants in the southeastern U.S.A. Infected plants may be stunted and can lose aesthetic value due to chlorosis, wilting, and leaf margin necrosis. We assessed reproduction of three root-knot nematode species, Meloidogyne arenaria, M. incognita, and M. javanica, on five plant taxa native to the southeastern U.S.A. and three non-native species commonly used in landscapes. The native taxa were: Hydrangea quercifolia 'Oakleaf,' Viburnum obovatum 'Densa,' Itea virginica 'Little Henry,' Illicium parviflorum, and Clethra alnifolia 'Ruby Spice.' The non-native shrubs were: Ligustrum japonicum 'Texanum,' Ilex crenata 'Compacta,' and Buxus microphylla 'Wintergem.' An interaction between plant taxa and Me*loidogyne* species was observed (P < 0.0001), and galling and nematode eggs were abundant on roots of the three non-native taxa. Among the plant species tested, highest galling (10) was observed on roots of I. crenata 'Compacta' infected with M. incognita, but largest number of eggs (6,397 eggs/g of roots) was observed in plants of this cultivar inoculated with M. javanica. Few or no galls were observed on roots of the five native plant taxa, and nematode eggs were recovered only from roots of I. virginica 'Little Henry' inoculated with M. arenaria and M. javanica (13 and 20 eggs/g of roots, respectively). Weight of shoots or roots of all species was not affected by nematode inoculation. Due to lack of root gall development and little or no reproduction on the native taxa, we conclude that these are poor or non-hosts to the three species of Meloidogyne. Landscape plantings of these plants, therefore, should be recommended as alternatives in sites with soil infested by Meloidogyne arenaria, M. incognita, or M. javanica.

Key words: host plant resistance, native plants, root-knot nematodes.

RESUMEN

Sharma, J., and J. R. Rich. Evaluación de la reproducción de tres especies de *Meloidogyne* en plantas leñosas nativas del Sureste de Estados Unidos. Nematropica 35:23-30.

La utilización de plantas nativas en paisajismo es de interés por la percepción de requerir menor mantenimiento, y por la posible resistencia a plagas como los nematodos fitoparásitos. *Meloidogyne* spp. (nematodo del nudo radical) afecta muchas especies de plantas ornamentales en el Sureste de Estados Unidos. Las plantas afectadas pueden ser de menor tamaño y pueden perder su valor estético debido a clorosis, marchitez, o necrosis de las márgenes foliares. Se evaluó la reproducción de tres especies del nematodo del nudo radical, *Meloidogyne arenaria, M. incognita* y *M. javanica*, en cinco especies de plantas nativas del Sureste de Estados Unidos (*Hydrangea quercifolia* 'Oakleaf,' *Viburnum obovatum* 'Densa,' *Itea virginica* 'Little Henry,' *Illicium parviflorum* y *Clethra alnifolia* 'Ruby Spice'), y tres especies no nativas comúnmente usadas como ornamentales (*Ligustrum japonicum* 'Texanum,' *Ilex crenata* 'Compacta' y *Buxus microphylla* 'Wintergem'). Se observó una interacción entre la planta y el nematodo (P < 0.0001), y se encontraron abundantes agallas y huevos en las raíces de las tres especies no nativas. La mayor formación de agallas (10) se encontró en raíces de *I. crenata* 'Compacta' con *M. incognita*, pero la mayor cantidad de huevos (6,397 huevos/g de raíces) se observó en plantas de esta variedad inoculadas con *M. javanica*. En las cinco especies de plantas nativas se observó escasa o ninguna formación de agallas, y se recuperaron huevos sólo de las raíces de *I. virginica* 'Little Henry' inoculada con *M. arenaria* y *M. javanica* (13 y 20 huevos/g de raíces, respectivamente). El peso de las raíces y de la parte aérea de la planta no se vio afectado por la presencia de los nematodos. Con base en la ausencia de agallas y en la reproducción limitada o inexistente en las plantas nativas estudiadas, concluimos que éstas no son buenos hospedantes de las tres especies de *Meloidogyne*, o son inmunes a ellas. Se debe recomendar, entonces, el uso de dichas plantas como alternativas en lugares en donde el suelo esté infestado con *Meloidogyne arenaria*, *M. incognita* o *M. javanica*.

Palabras clave: nematodo del nudo radical, plantas nativas, resistencia de planta hospedante.

INTRODUCTION

Production and planting of ornamentals in landscapes is a multi-billion dollar industry in southeastern U.S.A. In this region, nematodes are important pests of ornamental plants, and while several genera of nematodes cause disease, Meloidogyne spp. (root-knot nematodes) are particularly damaging (Nemec and Struble, 1968; Benson and Barker, 1985; Williams-Woodward and Davis, 2001). Three species of Meloidogyne. Meloidogyne arenaria (Neal) Chitwood, Meloidogyne incognita (Kofoid and White) Chitwood, and Meloidogyne javanica (Treub) Chitwood are widely distributed in the region and damage a wide variety of plants, including ornamentals (Koenning et al., 1999). Susceptible plants are often stunted and may exhibit chlorosis, leaf margin necrosis, and wilting due to inhibited root functions. A number of studies have been conducted to verify host status of woody ornamentals to Meloidogyne species (Barker et al., 1979; Bernard and Witte, 1987; Santo and Lear, 1976; Giblin-Davis et al., 1992; William-Woodward and Davis, 2001). However, the research has included a relatively small number of commonly grown woody ornamentals compared to the wide diversity of plants used in southeastern U.S.A. landscapes. Furthermore, native taxa which have recently been introduced or have recently become common in landscapes largely remain untested for susceptibility to root-knot nematodes.

Sanitation, plant resistance, chemical control, and exclusion of infected materials are common methods for preventing or reducing infection of plants by nematodes (Dunn and Crow, 2001). While use of nematicides to reduce infestation might be a suitable method for minimizing damage and is an option for commercial producers, use of plants that are naturally resistant or tolerant to nematode infection is an environmentally sustainable alternative for both commercial plant producers and consumers. It also may be the only economically viable option for consumers interested in preventing soil infestation or introduction of pests through infected planting stock. Selecting and planting non-hosts or tolerant species, therefore, will benefit the ornamental plant industry and consumers alike.

Use of nematode resistant woody ornamental plants is an alternative to planting commonly grown, susceptible species. Many native plants have desirable ornamental attributes and are adapted to regional environmental conditions that may make them suitable for use in landscapes. Because they have evolved in soils harboring species of *Meloidogyne*, several plant taxa may have developed inherent tolerance or resistance to these pests. Before widespread plantings are encouraged, however, plants should be tested for their resistance to *Meloidogyne* species prevalent in the region.

The objective of this study was to assess the reproduction of Meloidogyne arenaria (Neal) Chitwood Race 1, Meloidogyne incognita (Kofoid and White) Chitwood Race 2, and Meloidogyne javanica (Treub) Chitwood on cultivars of five native taxa and three commonly grown non-native shrubs: Hydrangea quercifolia Bartr. 'Oakleaf,' Viburnum obovatum Walt. 'Densa,' Itea virginica Illicium parviflorum 'Little Henry,' L. Michx. ex Vent., and Clethra alnifolia L. 'Ruby Spice,' Ligustrum japonicum Thunb. 'Texanum,' Ilex crenata Thunb. 'Compacta,' and Buxus microphylla Sieb. and Zucc. 'Wintergem.'

MATERIALS AND METHODS

Six-month-old, clonally propagated plants were obtained from commercial nurseries. Roots were shaken to remove excess growing medium and were inspected to insure absence of root galling or other signs of deterioration. Individual plants were placed in the growing substrate [4 parts builders sand and 1 part growing medium (Growing Mix 2-Conrad Fafard, Inc., Agawam, Maine, U.S.A.)] contained in 20-cm-diam plastic pots (volume = 3.2 L). Plants were grown for four weeks in the greenhouse before inoculation with nematode eggs. A factorial arrangement of eight plant taxa and four nematode treatments (three species of nematodes and one without nematode inoculation) was established in a completely randomized design. A pot containing one plant was an experimental unit, and each of the 32 treatments was replicated six times. Plants were fertilized as recommended with 19-6-12 N-P₂O₅-K₂O by using Osmocote® (Scotts-Sierra Horticultural Products Company, Marysville, OH) and were watered daily. Ambient temperature in the greenhouse averaged 27°C. No supplemental irradiance was provided in the greenhouse.

Plant roots were inoculated by injecting (via a syringe) 5 ml of deionized water containing approximately 10,000 nematode eggs into each of the three, 6-cmdeep holes created approximately 3-cm from the base of plants. Inoculum was extracted from infected roots of 'Rutgers' tomato (Lycopersicon esculentum Mill.) by using a sodium hypochlorite (NaOCl) extraction method (Hussey and Barker, 1973). Ten weeks after inoculation, plants were removed from pots, and roots were washed to remove the substrate. Fresh weights of shoots and roots were determined. Galling on roots was rated visually on a linear scale of 0 to 10, where 0 = nogalls and 10 = 100% of roots with galls. Eggs were extracted from 20 g roots by using a 1% NaOCl solution (Hussey and 1973). Rinseate was poured Barker, through stacked sieves (150-µm-pore and 25-µm-pore size), eggs were collected, and suspensions were diluted as needed to count eggs using a stereomicroscope.

An analysis of variance (ANOVA) model was tested by using the GLM procedure in SAS/STAT (Version 8.02, SAS Institute, Inc., Cary, North Carolina, U.S.A.) with galling and number of eggs as dependent variables. Gall rating scale reflected the percentage of roots with galls and was a continuous variable; however, Chi-square (Fisher's Exact Test) also was used to evaluate galling data. When appropriate, means were separated by using Fisher's LSD ($\alpha = 0.05$). The REG procedure was used to estimate the relationship ($\alpha = 0.05$) between galling and number of eggs per gram of roots.

RESULTS AND DISCUSSION

Both root galling and egg production were influenced by an interaction between plant taxon and species of nematode (P < 0.0001 and P < 0.0001 for galling and eggs, respectively). Galling counts were also significantly different (Fisher's Exact Test; P < 0.001; Table 1). Fresh weight of roots and shoots were plant-specific and were not influenced by inoculation with nematodes (Table 1).

Among the 32 plant-nematode treatments, eggs were sometimes present on roots that did not form galls. Egg production on plants without galls occurred on I. virginica 'Little Henry' plants inoculated with M. arenaria and M. javanica, and L. japonicum 'Texanum' inoculated with M. incognita or M. javanica (Table 1). Nematode infection and reproduction was greatest on I. crenata 'Compacta' inoculated with M. incognita and M. javanica; galling on roots ranged from 0 (all control treatments) to 10 on I. crenata 'Compacta' inoculated with M. incognita (eggs/g roots = 3,455). More eggs (6,397/g roots; galling = 8) were extracted from I. crenata 'Compacta' plants inoculated by M. javanica, however. All three non-native shrubs were moderate to good hosts for all three Meloidogyne species; for example, egg production (5,034/g roots) and galling (mean = 6) was prolific in B. microphylla 'Wintergem' inoculated with M. javanica (Table 1). No galling occurred on three of the five native shrubs: C. alnifolia 'Ruby Spice,' I. parviflorum, and V. obovatum 'Densa.' While these three hosts were completely resistant to the three Meloidogyne species in this study, some galling (mean rating = 1)

was observed on roots of *I. virginica* 'Little Henry' inoculated with *M. incognita*. A linear and positive relationship between galling and number of eggs/g of roots was observed for the three non-native taxa ($r^2 = 0.5$; P < 0.0001; Fig. 1).

While many plants were severely galled, inoculation with the three Meloidogyne species did not affect growth of plants (estimated by fresh weight of shoots and roots) in this study. The greenhouse environment was optimal for plant growth and possibly reduced adverse effects of nematode infection as noted also by Benson and Barker (1982). Number of eggs per gram of roots and galling data were related, but use of both indices is recommended for species that remain untested because different taxa may respond differently to infection by nematodes, and reproduction can occur in the absence of galling. The non-native plant taxa were good to excellent hosts to the three Meloidogyne species and recommendations for their use should be tempered by considering their host status to the three species of root-knot nematodes commonly found in the southeastern U.S.A. Ilex crenata 'Compacta' has been reported to be susceptible to Race 1 of M. arenaria and Race 3 of M. incognita among other species of root-knot nematodes not included in this study, and our results confirm the susceptibility of this taxon to Race 1 of M. arenaria (Benson and Barker, 1982; Bernard et al., 1994; William-Woodward and Davis, 2001). Our results also show that this cultivar is highly susceptible to M. javanica-a finding not previously reported. Many cultivars of several holly species are susceptible to *M. arenaria*, M. incognita, and M. hapla, and it appears that as a whole this genus is a common host to root-knot nematodes (Barker et al., 1979; Bernard and Witte, 1987; Bernard et al., 1994; Nemec and Struble, 1968). Buxus *microphylla* was susceptible to *M. incognita* Table 1. Means of root galling, galling count, and number of eggs on roots of eight woody ornamental plants inoculated with three species of *Meloidogyne*. Mean shoot and root weights of each plant taxon also were determined.

| Plant taxon | Meloidogyne species | Mean galling ^x | Galling count | Eggs/g roots ^y | Weight (g) ^z | |
|--------------------------------|------------------------|------------------------------|------------------|------------------------------|-------------------------|-------|
| | | | | | Shoots | Roots |
| Ligustrum japonicum 'Texanum' | | | | | 34 | 24 |
| | Control (uninoculated) | $0 \mathrm{g}$ | 0 | 0 f | | |
| | Meloidogyne arenaria | 3 e | 17 | 1,067 d | | |
| | Meloidogyne incognita | $0 \mathrm{g}$ | 2 | 999 d | | |
| | Meloidogyne javanica | $0 \mathrm{g}$ | 0 | 413 e | | |
| Ilex crenata 'Compacta' | | | | | 5 | 3 |
| - | Control | $0 	ext{ g}$ | 0 | 0 f | | |
| | Meloidogyne arenaria | 4 d | 22 | 3,760 с | | |
| | Meloidogyne incognita | 10 a | 58 | 3,455 с | | |
| | Meloidogyne javanica | 8 b | 45 | 6,397 a | | |
| Buxus microphylla 'Wintergem' | | | | | 4 | 4 |
| 17 8 | Control | 0 g | 0 | 0 f | | |
| | Meloidogyne arenaria | 4 d | 26 | 1,318 d | | |
| | Meloidogyne incognita | 3 e | 15 | 251 e | | |
| | Meloidogyne javanica | 6 c | 37 | 5,034 b | | |
| Hydrangea quercifolia 'Oaklea | f | | | | 123 | 40 |
| | Control | $0 	ext{ g}$ | 0 | 0 f | | |
| | Meloidogyne arenaria | 0 g | 0 | 0 f | | |
| | Meloidogyne incognita | 0 g | 0 | 0 f | | |
| | Meloidogyne javanica | $0 \mathrm{g}$ | 0 | 0 f | | |
| Illicium parviflorum | | | | | 19 | 10 |
| 1 5 | Control | $0 	ext{ g}$ | 0 | 0 f | | |
| | Meloidogyne arenaria | 0 g | 0 | 0 f | | |
| | Meloidogyne incognita | 0 g | 0 | 0 f | | |
| | Meloidogyne javanica | 0 g | 0 | 0 f | | |
| Clethra alnifolia 'Ruby Spice' | | | | | 10 | 8 |
| 5 7 1 | Control | 0 g | 0 | 0 f | | |
| | Meloidogyne arenaria | 0 g | 0 | 0 f | | |
| | Meloidogyne incognita | 0 g | 0 | 0 f | | |
| | Meloidogyne javanica | 0 g | 0 | 0 f | | |
| Viburnum obovatum 'Densa' | | | | | 91 | 18 |
| | Control | $0~{ m g}$ | 0 | 0 f | 41 | 10 |

Table cont.

Table 1. (Continued) Means of root galling, galling count, and number of eggs on roots of eight woody ornamental plants inoculated with three species of *Meloidogyne*. Mean shoot and root weights of each plant taxon also were determined.

| Plant taxon | Meloidogyne species | Mean galling [*] | Galling count | Eggs/g roots ^y | Weight (g) ^z | |
|-------------------------------|-----------------------|------------------------------|------------------|------------------------------|-------------------------|-------|
| | | | | | Shoots | Roots |
| | Meloidogyne arenaria | 0 g | 0 | 0 f | | |
| | Meloidogyne incognita | $0 \mathrm{g}$ | 0 | 0 f | | |
| | Meloidogyne javanica | $0 \mathrm{g}$ | 0 | 0 f | | |
| Itea virginica 'Little Henry' | | | | | 20 | 15 |
| | Control | $0 \mathrm{g}$ | 0 | 0 f | | |
| | Meloidogyne arenaria | $0 \mathrm{g}$ | 0 | 13 e | | |
| | Meloidogyne incognita | 1 f | 7 | 9 e | | |
| | Meloidogyne javanica | $0 \mathrm{g}$ | 1 | 20 e | | |

*Plants were grown for 10 weeks, after which galling was rated on a scale of 1 to 10, where 0 = no galling and 10 = 100% galling. Means within a column followed by the same letter are not significantly different [Fisher's lsd ($\alpha = 0.05$)]. Galling counts were evaluated by using Fisher's Exact Test (P < 0.001).

'Eggs were extracted from roots to estimate the number of eggs/gram of roots. Means within a column followed by the same letter are not significantly different [Fisher's LSD ($\alpha = 0.05$)].

'Mean fresh weights of shoots and roots were determined before eggs were extracted from the roots. Only main effect of plant taxon was significant and is reported.

and M. arenaria in other studies (Benson and Barker, 1982), but the cultivar 'Wintergem' was not included (Benson and Barker, 1982; Williams-Woodward and Davis, 2001). This cultivar was affected most severely by M. javanica in our tests. To our knowledge, the host status of Ligustrum japonicum 'Texanum' to these three nematode species has not been reported. Data on other Ligustrum species also are sparse; while Ligustrum sinensis 'Variegatum' is susceptible to M. hapla (Bernard and Witte, 1987), susceptibility of other cultivars is not known, nor is its susceptibility to other nematode species is known.

Although the native taxa tested in this study were poor or non-hosts to the three common species of *Meloidogyne*, the host status mostly remains unknown for many other ornamental taxa native to southeastern U.S.A. *Myrica cerifera* L. (wax myrtle) is one of the very few southeastern woody

plants known to be a host to Meloidogyne species (Crow and Dunn, 2002). Generalizations, however, cannot be realistically made based on tests conducted on a few native species, and therefore, host status of individual species and perhaps their cultivars should be evaluated before recommendations are made for their widespread use in landscapes. For example, limited reproduction of nematodes on roots of taxa such as I. virginica 'Little Henry' could affect adjacent, more susceptible plants in the landscape. Further, simply because some plants grow in similar habitats in nature would not indicate that their host status to root-knot nematodes would be similar. Resistance or tolerance to nematode infection appears to be a plant response, and two species growing in the same location may respond differently. Use of poor or non-host plants and cultivars is likely the most acceptable and cost-effec-



Fig. 1. Relationship between root galling and number of eggs/g of roots of three commonly used ornamental taxa (*Ligustrum japonicum* 'Texanum,' *Ilex crenata* 'Compacta,' and *Buxus microphylla* 'Wintergem') infected with *Meloidogyne arenaria*, *Meloidogyne incognita*, or *Meloidogyne javanica*. Galling was rated on a scale of 0 to 10, where 0 = no galling and 10 = 100% galling.

tive alternative for managing nematode infections of ornamental plants.

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