Host Status of Thirteen Acacia Species to Meloidogyne javanica

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Abstract: Thirteen indigenous and exotic Acacia species grown in Saudi Arabia were evaluated for their host status for Meloidogyne javanica in pot tests both in the growth chamber and under outdoor conditions. In both experiments, 21-day-old seedlings were transplanted individually into 15-cm-diam. plastic pots containing a steam-sterilized mixture of equal parts loam and sandy loam. Seedlings were inoculated with 5,000 M. javanica eggs/plant 30 days later. After 120 days, fresh root weight, disease index (1-9 scale), the number of eggs/pot (Pf), eggs/g fresh root, and a reproductive factor (Rf) were determined. Results of both the growth chamber and the outdoor tests were similar. Species were grouped into host suitability categories according to Rf, and they were also grouped into resistance categories based on the sum of gall index, gall size, and percentage of the root system that was galled. Only A. salicina was a poor host and was resistant to M. javanica. Acacia farnisiana, A. gerrardii subsp. negevensis var. najdensis, and A. saligna were excellent hosts and highly susceptible. Both A. nilotica and A. stenophylla were classified as good hosts and highly susceptible, while A. ampliceps, A. ehrenbergiana, A. gerrardii subsp. negevensis var negevensis, A. sclerosperma, A. seyal, A. tortilis, and A. tortilis subsp. spirocarpa were also good hosts but were classified as susceptible rather than highly susceptible. This is the first report on the susceptibility of Acacia species to M. javanica in Saudi Arabia, including some new hosts worldwide.

Key words: Acacia species, forest trees, host suitability, Meloidogyne javanica, reproduction, resistance, root-knot nematode, screening, susceptibility.

The genus Acacia Miller comprises about 1,100 to 1,200 tree and shrub species worldwide (Kennenni, 1991). Most of these species grow in arid and semiarid regions, where the average temperature is 40 to 45 °C in summer and less than 5 °C in winter (Singh et al., 1990). Acacia species, which possess most of the features required to withstand severe climatic conditions, have been called the most successful survivors in arid regions. Acacia tortilis and A. seval are the most often recommended species to be planted in arid and semiarid regions (El-Amin, 1976). Most Acacia grown in Saudi Arabia are trees and shrubs, mostly belonging to the species A. tortilis, A. ehrenbergiana, and A. seyal. However, A. gerrardii subsp. negevensis var. najdensis and A. gerrardii subsp. negevensis var. negevensis are distributed in central and northern Saudi Arabia only. During the past two decades, forestation activities in Saudi Arabia have increased tremendously, and many indigenous and exotic *Acacia* species have been planted (Aref, 1996).

Meloidogyne javanica (Treub) Chit., the most widespread and prominent nematode in Saudi Arabia, has been shown to infect many vegetables, field crops, ornamentals, trees, shrubs, and grasses (Al-Hazmi et al., 1995). No information is available on the attack of Acacia species by root-knot nematodes in Saudi Arabia, but several reports have indicated the susceptibility of some Acacia species to M. javanica in different parts of the world. These include A. nilotica and A. farnesiana in India (Dahiya et al., 1988; Haseeb et al., 1981), A. holosericea and A. mangium in Sengal (Duponnois et al., 1998), and Acacia spp. in Egypt (Ibrahim et al., 1986).

Recently, we have found some *Acacia* trees with relative poor growth that suffered from yellowing, especially during the hot summer months, on the Agricultural Experiment Station at Dierab, Riyadh. Roots of these trees were heavily galled, with root-knot nematode egg masses visible. Females were extracted from the galled roots and identified as *M. javanica* based on the morphological features of the perineal pattern (Hartman and Sasser, 1985). The objective of this

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study was to determine the susceptibility and host status to *M. javanica* of the most commonly grown *Acacia* species in Saudi Arabia.

MATERIALS AND METHODS

Plant materials: Seeds of 13 indigenous and exotic Acacia species were treated with concentrated sulfuric acid for 10 minutes, washed in running tap water for 30 minutes, and sown in clean plastic trays containing a steam-sterilized mixture of equal parts peat moss and sandy loam soil in the growth chamber (30/25 °C; 12-hour night). The indigenous species included A. ehrenbergiana Hayne, A. gerrardii subsp. negevensis var. najdensis Chaudhary, A. gerrardii subsp. negevensis var. negevensis Zoh., A. nilotica (L.) Willd. ex Del., A. tortilis (Forssk.) Hayne, and A. tortilis subsp. spirocarpa (A. Rich.) Brenan. The exotic species included A. ampliceps Maslin, A. farnesiana (L.) Willd., A. salicina Lindley, A. saligna (Labill) Wendl, A. sclerosperma Benth., A. seyal Del., and A. stenophylla A. Cunn. ex Benth. Three weeks after germination, five uniform seedlings of each Acacia species were transplanted individually into 15-cm-diam. plastic pots partially filled with a steam-sterilized mixture of equal parts sand and sandy loam soil. Two-week-old tomato seedlings (Lycopersicon esculentum Mill.) cv. Rutgers were also individually transplanted into three pots containing the same soil mixture as a susceptible check. All seedlings were left for 4 weeks to allow rooting before inoculation.

Inoculum source and preparation: A single egg mass of *M. javanica* was originally isolated from an *Acacia* tree growing on our Agricultural Experiment Station at Dierab, Riyadh, and the population expanded on eggplant (*Solanum melongena* L.) cv. Black Beauty in the greenhouse. Whenever needed, inoculum was prepared by extracting nematode eggs from 8-week-old eggplant roots with sodium hypochlorite 0.5% for 3 minutes (Hussey and Barker, 1973).

Screening tests: All seedlings were inoculated with an aqueous suspension of 5,000 eggs/plant placed in three holes in the soil, 3 cm from the base of the plant (Sasser et al., 1984). Seedlings were returned to the growth chamber, arranged in a completely randomized design, and watered and fertilized as needed. One hundred twenty days after inoculation, pots were gently tapped to loosen the soil from around the roots. Roots were carefully washed with a gentle stream of tap water and weighed. Roots were rated on a 1-to-9 scale for gall index (GI): 0 = no galls, 2 = 1 to 5 galls, 3 = 6 to 10 galls, 4 = 11to 20 galls, 5 = 21 to 30 galls, 6 = 31 to 50 galls, 7 = 51 to 70 galls, 8 = 71 to 100 galls, and 9 = >100 galls per plant (Sharma et al., 1993). Gall size (GS) and percent galled area (GA) were also rated on a 1-to-9 scale. For GS: 1 = no galls, 3 = very small galls (about 10% increase in root area at the galled region over non-galled normal root area), 5 = small galls (about 30% increase), 7 =medium galls (about 31 to 50% increase), and 9 = big galls (about 51 to 100%) increase). For GA: 1 = no galls, 3 = 1 to 10%root area galled, 5 = 11 to 30% root area galled, 7 = 31 to 50% root area galled, and 9 = >50% root area galled (Sharma et al., 1993). A damage index (DI) was calculated by dividing the sum of GI, GS, and GA by 3 for each replicate (Sharma et al., 1993). Based on DI, the host susceptibility (designation of resistance) of each Acacia plant species was determined according to the following scheme: plants with DI = 1 were designated as highly resistant; DI = 2 to 3, resistant; DI = 4 to 5, moderately resistant; DI = 6to 7, susceptible; and DI = 8 to 9, highly susceptible (Sharma et al., 1993).

Roots were then cut into 1-cm segments and macerated in a stronger solution of sodium hypochlorite (1%) on a shaker for 10 minutes to extract eggs (Pf) (Hussey and Barker, 1973). Nematode reproductive factor (Rf) was calculated according to the formula (Rf = Pf/Pi), where Pf = the number of eggs at the termination of the test and Pi = the initial inoculum level (Oostenbrink, 1966). Average number of nematode eggs per g fresh root was also calculated to determine the host suitability of the tested plants. Based on Rf values, plants were grouped in five categories of host suitability: nonhost (Rf = 0), poor host (Rf = 0.1 to 0.9), maintenance host (Rf = 1.0 to 2.9), good host (Rf = 3.0 to 9.9), and excellent host (Rf = 10>) (Ferris et al., 1993).

The experiment was repeated with the same materials and procedures 6 months later, under outdoor conditions. The average temperature during this study period was $38 \pm 6/20 \pm 5$ °C day/night, and the average relative humidity was 31.5%. Data from both experiments were combined to determine the host susceptibility and suitability. Data of final nematode population (Pf), number of eggs per g fresh root, and nematode reproductive factor (Rf) were subjected to ANOVA (SAS Institute Inc., Cary, NC). When F values were significant, means were separated by the Fisher's protected LSD ($P \le 0.05$).

RESULTS

Results of both tests were similar. Tomato roots (the susceptible control) were heavily galled and had numerous egg masses, confirming the viability and aggressiveness of the inoculum (data not shown). The tested *Acacia* species differed in their susceptibility (Table 1) and ability to support nematode reproduction (Table 2). Only A. salicina was resistant (DI = 2.3) and a poor host (Rf = 0.3) to M. javanica. Acacia farnesiana, A. gerrardii subsp. negevensis var. najdensis, and A. saligna were highly susceptible (DI > 8) and excellent hosts (Rf > 10). Acacia nilotica and A. stenophylla were highly susceptible but good hosts (Rf = 6.1 and 5.0, respectively). Acacia ampliceps, A. ehrenbergiana, A. gerrardii subsp. negevensis var. negevensis, A. sclerosperma, A. seyal, A. tortilis, and A. tortilis subsp. *spirocarpa* were susceptible (DI = 6 to 7) and good hosts (Rf = 3.4 to 8.6) (Tables 1,2). Number of eggs/g fresh root was highest ($P \le 0.05$) for A. nilotica, A. ehrenbergiana, and A. ampliceps and was lowest on A. salicina (Table 2).

DISCUSSION

Based on DI values, all tested Acacia species were susceptible or highly susceptible to *M. javanica* except *A. salicina*, which was resistant. The susceptibility of *A. farnesiana* and *A. nilotica* to *M. javanica* has been previously reported in India (Dahiya et al., 1988; Hasseb et al., 1981). Other Acacia species (not included in this study) also have

TABLE 1. Host susceptibility of 13 Acacia species to Meloidogyne javanica.

| Acacia species | GI ^a | GS^{b} | GAc | DI^d | Host susceptibility ^e |
|--|-----------------|----------|-----|--------------------------|-------------------------------------|
| A. ampliceps | 7.8 | 7.0 | 7.0 | 7.3 | S |
| A. ehrenbergiana | 5.3 | 8.0 | 5.0 | 6.1 | S |
| A. farnesiana | 7.2 | 8.4 | 9.0 | 8.2 | HS |
| A. gerrardii subsp. negevensis var. najdensis | 7.9 | 9.0 | 9.0 | 8.6 | HS |
| A. gerrardii subsp. negevensis var. negevensis | 7.0 | 7.6 | 7.4 | 7.3 | S |
| M. nilotica | 8.0 | 8.0 | 8.0 | 8.0 | HS |
| A. salicina | 2.3 | 2.4 | 2.2 | 2.3 | R |
| A. saligna | 8.0 | 7.8 | 8.4 | 8.1 | HS |
| A. sclerosperma | 5.8 | 7.6 | 5.4 | 6.3 | S |
| A. seyal | 7.1 | 6.6 | 7.2 | 7.0 | S |
| A. stenophylla | 8.4 | 9.0 | 8.6 | 8.7 | HS |
| A. tortilis | 7.9 | 6.6 | 7.5 | 7.3 | S |
| A. tortilis subsp. spirocarpa | 6.6 | 6.6 | 8.0 | 7.1 | S |

Data are an average of 10 replications from two experiments executed 120 days after inoculation.

^a GI = gall index (1–9) where: 1 = no galls, 2 = 1–5, 3 = 6–10, 4 = 11–20, 5 = 21–30, 6 = 31–50, 7 = 51–70, 8 = 71–100, and 9 = >100 galls per plant.

^b GS = gall size index (1–9) where: 1 = no galls, 3 = very small galls (about 10% increase in root area at the galled region over non-galled normal root area), 5 = small galls (about 30% increase), 7 = medium galls (about 31–50% increase), and 9 = big galls (about 51–100% increase).

 $^{\circ}$ GA = percent galled area index (1–9) where: 1 = no galls, 3 = 1–10% root area galled, 5 = 11–30% root area galled, 7 = 31–50% root area galled, and 9 = >50% root area galled.

^d DI = damage index (1-9) = [gall index (1-9 scale) + gall size (1-9 scale) + %root area galled (1-9 scale)]/3.

^e Host susceptibility: S = susceptible (DI = 6–7), HS = highly susceptible (DI = 8–9), R resistant (DI = 2–3).

| Acacia species | No. eggs/g fresh root | Final population (Pf) | Reproductive factor (Rf) ^a | Host suitability ^b |
|--|-----------------------|--------------------------|--|----------------------------------|
| A. ampliceps | 35,900 b | 43,100 bcd | 8.6 ab | G |
| A. ehrenbergiana | 54,100 a | 27,600 cde | 5.5 bc | G |
| A. farnesiana | 22,500 bcd | 54,600 a | 10.9 a | Е |
| A. gerrardii subsp. negevensis var. najdensis | 26,600 bcd | 52,600 a | 10.5 a | E |
| A. gerrardii subsp. negevensis var. negevensis | 10,300 de | 16,800 ef | 3.4 cd | G |
| N. nilotica | 64,600 a | 30,300 bcde | 6.1 abc | G |
| A. salicina | 500 e | 1,400 f | 0.3 d | Р |
| A. saligna | 28,700 bc | 50,700 bc | 10.1 a | Е |
| A. sclerosperma | 18,100 cd | 23,600 def | 4.7 bcd | G |
| A. seyal | 12,700 cde | 18,900 def | 3.8 bcd | G |
| A. stenophylla | 13,200 cde | 24,900 def | 5.0 bcd | G |
| A. tortilis | 25,100 bcd | 32,600 bcde | 6.5 abc | G |
| A. tortilis subsp. spirocarpa | 28,600 bc | 18,900 def | 3.8 bcd | G |

TABLE 2. Host suitability of 13 Acacia species to Meloidogyne javanica.

Values are means of 10 replications from two experiments executed at 120 days after inoculation; means followed by the same letter(s) within a column are not significantly ($P \le 0.05$) different according to FLSD.

^a RF (reproductive factor) = final population/initial population (5,000 eggs/pot).

^b Host suitability: G = good host (RF = 3.0-9.9), E = excellent host (RF = 10>), P = poor host (RF = 0.1-0.9).

been reported to be susceptible to M. javanica in Egypt (Ibrahim et al., 1986) and Sengal (Dupponnois et al., 1998) and to Meloidogyne spp. in Sarawak (Chin, 1986). Although plant-parasitic nematodes rarely kill Acacia trees, they suppress growth and predispose the trees to other pathogens (Nasr et al., 1980). Nematodes may also limit the absorption of water and utilization of nutrients and, in certain cases, serve as vectors of plant viruses (Mai, 1985; Sasser, 1989). However, young seedlings could be severely damaged or killed by root-knot nematodes in certain cases. Carpenter (1964) reported that root-knot nematodes, principally M. javanica, killed more than 90% of the seedlings of 50 date palm varieties prior to emergence when seeds were sown in heavily infested soil.

The current results indicated that *M. javanica* has high potential to infect and reproduce on all tested *Acacia* species except *A. salicina*. Numbers of eggs per g fresh root were used to examine the relative efficiency of the tested *Acacia* species for the nematode reproduction. *Acacia nilotica, A. ehrenbergiana,* and *A. ampliceps* had Rf values ranging from 5.5 to 8.6 and were categorized as good hosts. However, they gave the highest numbers of eggs/g fresh root. This may be attributed to the fact that these species had the

lowest root fresh weights among the tested Acacias (data not shown). Because Acacia trees and shrubs are of great value in our agro-forestry system (Milton and Hall, 1981), it is important to protect this national resource from attack by *M. javanica* or any other nematodes or pathogens. Root-knot nematodes may increase the water stress of trees, especially in the hot summer months where temperatures sometimes exceed 50 °C. *Meloidogyne javanica* infection can also adversely affect nodule production and nitrogen fixation in *Rhizobium*-legume interactions (Huang, 1987).

Fortunately, A. salicina, which was the only species in our study that was both resistant and a poor host for M. javanica, is well adapted to the Saudi environment. This species exhibits rapid early growth and produces many branches. It is relatively drought tolerant and also tolerant of high temperature and blowing dust and sand, and can be successfully used in reforestation and as wind breaks and green belts (Al-Zoghet and Tag El-Din, 1995). This species may also be important as a source of resistance to M. *javanica* in breeding programs or as a resistant rootstock for grafting of other more desirable Acacia species. Additional Acacia species should be tested to determine their resistance to M. javanica and other plantparasitic nematodes that are common in the region, to continue to improve our agroforestry system.

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