Relationship of *Meloidogyne konaensis* Population Densities to Coffee Growth

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ABSTRACT

Zhang, F., and Schmitt, D. P. 1995. Relationship of *Meloidogyne konaensis* population densities to coffee growth. Plant Dis. 79:446-449.

Coffee (*Coffea arabica* L.) shoot and root growth as affected by *Meloidogyne konaensis* was determined under greenhouse and field conditions. The greenhouse test involved five coffee cultivars: Guatemalan, S. L. 28, Guadalupe, Mundo Novo, and Red Bourbon. *Meloidogyne konaensis* damaged all coffee cultivars under greenhouse conditions. Shoot growth was suppressed at all inoculum densities (150–18,750 eggs per plant). Dry shoot and root weights were negatively correlated with the $\log_{10}(Pi + 1)$ of *M. konaensis*. Minimum predicted shoot height and dry shoot and root weights were 35.2, 7.6, and 10.1% of the maximum predicted levels, respectively. Growth of cultivar Guatemalan and Guatemalan as a scion on Deweveri rootstock was characterized in a field naturally infested with *M. konaensis*. A negative linear regression relationship ($r = 0.95$) existed between the population density of this nematode and percent increase in coffee height in the field.

Coffee (*Coffea arabica* L.) is an economically important crop grown on 9 million ha with an annual world production of 5.6 billion kg (9). Brazil, Colombia, Mexico, some countries of Africa, India, El Salvador, Guatemala, and Costa Rica are the major coffee producing countries (9). Many of these countries grow this crop primarily for export. The major consumers are from developed countries such as the United States, the European Economic Community, and Japan. The United States is the largest importer of coffee (25–30% of world imports) (9).

Coffee is important to the agricultural economy of Hawaii. It was grown on approximately 2,100 ha in 1991; hectarage is predicted to increase substantially within the next 5 yr since the subtropical climate of Hawaii will result in a high quality product (2).

Many plant-parasitic nematodes, especially *Meloidogyne* spp., are associated with coffee (4). Some of them cause substantial yield reductions (4). Annual worldwide loss due to nematodes on coffee is estimated at about $2.5 billion (10). Direct and indirect losses due to nematodes, and added costs of protection have made coffee production uneconomical for some growers (4).

Meloidogyne konaensis Eisenback, Bernard, & Schmitt was described from an isolate obtained from coffee at the Kona Experiment Station on the island of Hawaii in 1991 (5). The original planting of coffee on the experimental farm became so damaged by this nematode that all trees were destroyed and the field replanted with a coffee rootstock trial. The objectives of this study were to determine the susceptibility of selected coffee cultivars to *M. konaensis* under greenhouse conditions and the relationship between coffee growth and population density of this nematode in the field.

MATERIALS AND METHODS

Greenhouse experiment. This experiment was conducted twice in a greenhouse (average temperature of 30 C) at Whitmore, Oahu, Hawaii. Three- (run 1) and five-month-old (run 2) nematode-free seedlings of coffee cultivars Guatemalan, Mundo Novo, S.L.28, Red Bourbon, and Guadalupe were used. Seedlings were transplanted into 15-cm-diameter clay pots (1,500 cm^3 soil) filled with a mixture of steam-sterilized 0.04-mm-diameter sand and soil (clayey, kaolintic, isothermic, Tropeptic Eutrustox, Oxisol) (1:1, v/v) 2 wk before inoculation. Seedlings were selected for uniform size and planted one per pot.

Egg inoculum for the experiments was obtained from a culture of *M. konaensis* from 2-mo-old tomato (*Lycopersicon esculentum* (L.) Mill. 'Rutgers') in a greenhouse. Eggs were extracted from the gelatinous matrix 1 day before inoculation of test plants using a NaOCl-blender extraction method (6). Eggs in 5-ml aqueous suspensions were pipetted into depressions in the soil around the root system of each seedling. The five inoculum densities were 0, 150, 750, 3,750, and 18,750 eggs per plant. Control plants were watered with egg-free filtrate. Each inoculum density was replicated five times. Pots were arranged in randomized complete blocks on three greenhouse benches. All pots were irrigated immediately after inoculation. Plants were watered daily with tap water and fertilized biweekly with water-soluble fertilizer (23% N, 19% P, 17% K) at a rate of 1.15 mg/cm^3 soil. Ambient greenhouse air temperatures ranged from 25 to 35 C. Plants were grown for 4 mo after inoculation in both runs. Run 1 was conducted from May to September 1991 and run 2 from October 1991 to February 1992.

Soil samples were collected from each pot 2 mo after inoculation. Before sampling, plant shoot height was measured. The entire soil and root mass was removed from each pot. The plants were removed carefully from the soil to minimize damage to the root system. The soil was mixed and a 250-cm^3 subsample was taken from the 1,500 cm^3 soil mass. Each plant was replanted into the original pot with the soil remaining after sampling. Second stage juveniles (J2) were extracted from each soil sample by elutriation (3) and centrifugal flotation (7). The counts were converted to numbers of J2/1,500 cm^3 soil.

At the end of each run, plant shoot height was determined and a 250-cm^3 soil sample was collected following the methods used at 2 mo. Galling index (0–10) (1) and root necrosis (%) were rated after carefully washing each root system with tap water. Shoots and roots were separated and oven dried at 70 C for 5 days.

Field experiment. The relationship of *M. konaensis* population density changes and coffee growth was determined in a coffee field naturally infested with *M. konaensis*. This field was located at the Kona Experiment Station, Kealakekua, Hawaii (elevation 450 m). The annual soil temperature ranges from 15 to 32 C (daily mean of 25 C).

Guatemalan and Guatemalan scion on Deweveri rootstock were selected from a field planted with eight combinations of 2 coffee scions (502 and Guatemalan)

The research reported in this publication was funded in part by Governor's Agricultural Coordinating Committee and the College of Tropical Agriculture and Human Resources.

Journal series No. 3983 of the Hawaii Institute of Tropical Agriculture and Human Resources.

Accepted for publication 12 January 1995.

on four rootstocks (Purpuree, Congensis, Deweveri, and Kaffe), and cultivars 502 and Guatemalan. The trees are planted in rows with 20 trees in most rows. The spacing is 3.5 m between rows and 2.0 m in the row. The tree roots were 3.5 yr old and the shoots were about 6 mo old when the study was initiated because the shoots were removed when the plants were about 3 yr old.

Experimental design was a randomized complete block. Twelve trees from three replications (four per replicate, if possible) were selected for sampling and monitoring of coffee growth parameters. Soil cores (5 cm diameter) were collected with a soil bucket auger at 0–15, 15–30, and 30–45 cm depths at 60 cm from the trunk on opposing sides of each tree. Tree height, canopy width, and trunk diameter (at 10 cm above the soil surface) were measured four times at approximately 3-mo intervals from November 1992 to August 1993.

Soil samples were composited for assays. The soil was sieved through a screen with 1-mm openings. Coffee roots were carefully removed from the sieves and oven dried at 70 C for 5 days. A 250-cm^3 soil subsample was placed in a plastic beaker and immersed in tap water for 24 h, then nematodes were extracted by a combination of elutriation (3) and centrifugal flotation (7).

Data analysis. Linear regressions (8) were performed on shoot lengths, dry shoot weight, and dry root weight versus $\log_{10}$-transformed nematode numbers. A tolerance limit of coffee cultivar to nematode population density was estimated using Seinhorst's model (11). Data were analyzed by analysis of variance by cultivar and time. Correlation coefficients (12) were calculated among plant growth parameters and $\log_{10}$-transformed nematode numbers over time. Least-squares analysis was used to fit simple linear and quadratic models to data on plant growth and nematode numbers.

RESULTS

Greenhouse experiment. The coffee cultivars had different growth rates in the absence of nematodes. In run 1, the shoot height of Guatemalan was 9.5 cm. The growth of S. L. 28 and Mundo Novo was 2.5 times that of Guatemalan. Red Bourbon and Guadalupe grew 1.7 times taller than Guatemalan. Shoot height of S. L. 28, Red Bourbon, Mundo Novo, and Guadalupe was 3.0, 2.3, 2.0, and 1.4 times greater than Guatemalan (16 cm), respectively, in run 2.

Coffee growth in the presence of nematodes also differed greatly among cultivars (Table 1). In run 1, slopes of the linear regressions between height and inoculum densities did not differ ($P = 0.05$) among Guatemalan, Guadalupe, S. L. 28, and Red Bourbon, but the slope for Mundo Novo was steeper than those

Table 1. Correlation coefficients (r) and linear regression models relating shoot height (SH) (cm), dry shoot (DS), and root weights (DR) (g) of five coffee cultivars to $\log_{10}(Pi + 1)$ of *Meloidogyne konaensis* in a greenhouse[a]

	Run 1		Run 2	
Cultivar	**Model**	r	**Model**	r
Guadalupe	SH = 16.64 − 1.33x[b]	−0.90**	SH = 21.72 − 1.44x	−0.78**
	DS = 1.86 − 0.3x	−0.81**	DS = 17.14 − 2.18x	−0.74**
	DR = 2.13 − 0.32x	−0.88**	DR = 5.41 − 0.36x	−0.69**
Guatemalan	SH = 9.46 − 0.88x	−0.53**	SH = 15.97 − 1.54x	−0.85**
	DS = 1.18 − 0.07x	−0.16NS	DS = 1.93 − 0.29x	−0.41**
	DR = 0.44 − 0.04x	−0.21NS	DR = 0.49 − 0.05x	−0.2NS
Mundo Novo	SH = 23.21 − 2.25x	−0.74**	SH = 32.73 − 4.39x	−0.91**
	DS = 5.24 − 1.03x	−0.91**	DS = 20.14 − 3.13x	−0.71**
	DR = 2.01 − 0.36x	−0.91**	DR = 6.48 − 0.9x	−0.72**
Red Bourbon	SH = 16.52 − 0.99x	−0.40*	SH = 36.71 − 4.69x	−0.90**
	DS = 6.5 − 1.2x	−0.93**	DS = 8.87 − 0.99x	−0.34*
	DR = 2.58 − 0.48x	−0.92**	DR = 4.06 − 0.55x	−0.57**
S. L. 28	SH = 23.17 − 1.38x	−0.70**	SH = 47.18 − 6.3x	−0.92**
	DS = 10.22 − 1.84x	−0.89**	DS = 25.75 − 3.09x	−0.60**
	DR = 4.18 − 0.72x	−0.85**	DR = 8.77 − 0.89x	−0.65**

[a] Data are means of five replications.
[b] $x = \log_{10}(Pi + 1)$, where Pi = initial population. * = significant at $0.05 < P \leq 0.1$. ** = significant at $P \leq 0.05$. NS = linear regression not significant ($P \leq 0.05$).

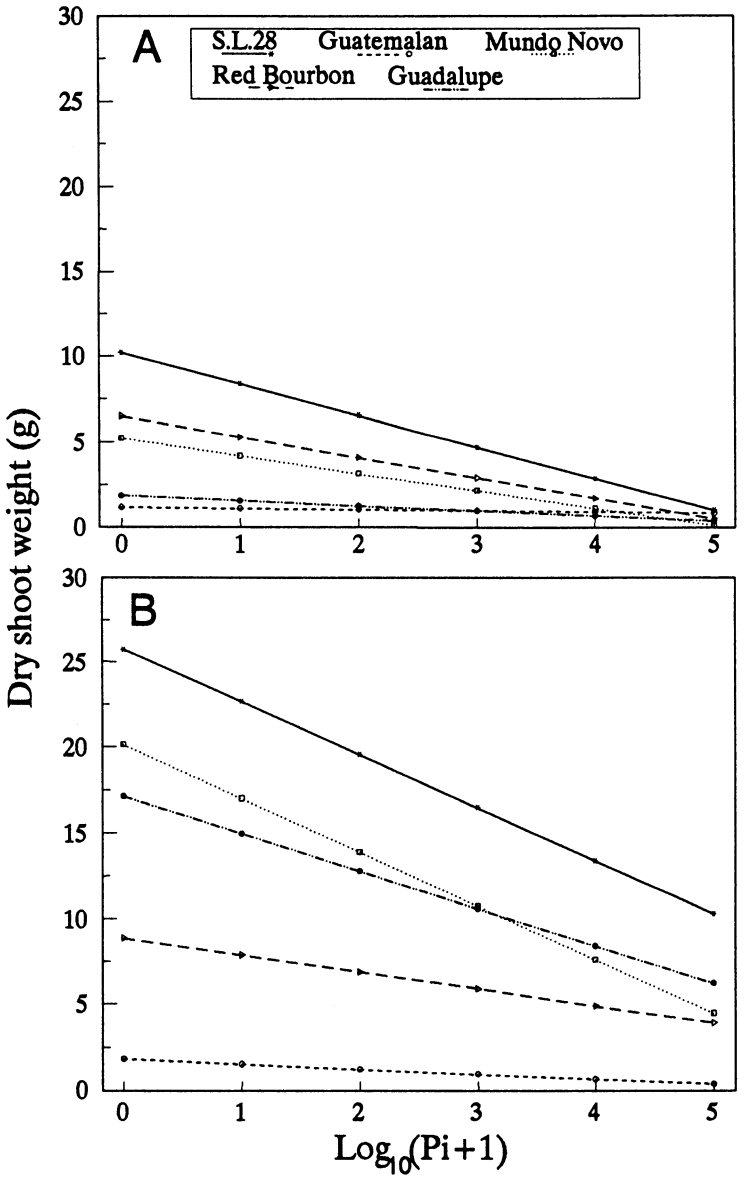

Fig. 1. Relationship between initial population density of *Meloidogyne konaensis* and dry shoot weight (g) of coffee 4 mo after inoculation in a greenhouse. **(A)** Run 1; and **(B)** Run 2. Data are means of five replications.

of the other four cultivars ($P = 0.05$). In run 2, there were three groups of slopes ($P = 0.05$). The steepest slope (−6.3) was for S. L. 28; the slopes for Red Bourbon and Mundo Novo were intermediate (−4.69 and −4.39, respectively); and the slopes of Guadalupe and Guatemalan were −1.54 and −1.44, respectively.

Dry shoot and root weights reflected the trends of shoot height. All of these parameters were negatively correlated with $\log_{10}(Pi + 1)$ of *M. konaensis* levels in both runs (Fig. 1 and Table 1). The correlations between nematode Pi and plant growth parameters of coffee cultivars Mundo Novo, Guadalupe, and S. L. 28 ranged from 0.60 to 0.92 and were significant ($P \leq 0.05$). Dry root weight of Red Bourbon had the strongest correlation of any parameter with Pi from the two runs. Dry shoot and root weight in run 1 and shoot height in run 2 were highly correlated with Pi. For Guatemalan, only shoot height in both runs and dry shoot weight in the second run gave significant ($P < 0.05$) correlations with Pi.

Table 2. Galling index and percentage of root necrosis on coffee cultivars 4 mo after inoculation with *Meloidogyne konaensis*[a]

	Inoculum level[b]									
	Run 1					Run 2				
Cultivar	0	1	2	3	4	0	1	2	3	4
Galling index (0–10)[c]										
Guadalupe	0	4	5	6	8	0	2	3	5	6
Guatemalan	0	4	6	8	10	0	2	5	6	7
Mundo Novo	0	3	6	7	10	0	3	5	5	7
Red Bourbon	0	2	6	8	8	0	3	4	6	6
S. L. 28	0	3	3	7	10	0	2	3	5	6
Root necrosis (%)										
Guadalupe	0	30	54	64	58	0	14	26	34	38
Guatemalan	10	44	46	92	100	16	42	54	50	54
Mundo Novo	3	28	45	94	100	2	22	26	46	52
Red Bourbon	2	20	40	79	95	14	36	50	66	66
S. L. 28	2	17	23	77	95	0	10	33	43	54

[a] Data are means of five replications.
[b] Number of eggs per plant. 0 = none; 1 = 150; 2 = 750; 3 = 3,750; and 4 = 18,750.
[c] Percentage of galling. 0 = none; 1 = 1–10%; 2 = 11–20%; 3 = 21–30%; 4 = 31–40%; 5 = 41–50%; 6 = 51–60%; 7 = 61–70%; 8 = 71–80%; 9 = 81–90%; and 10 = 91–100%.

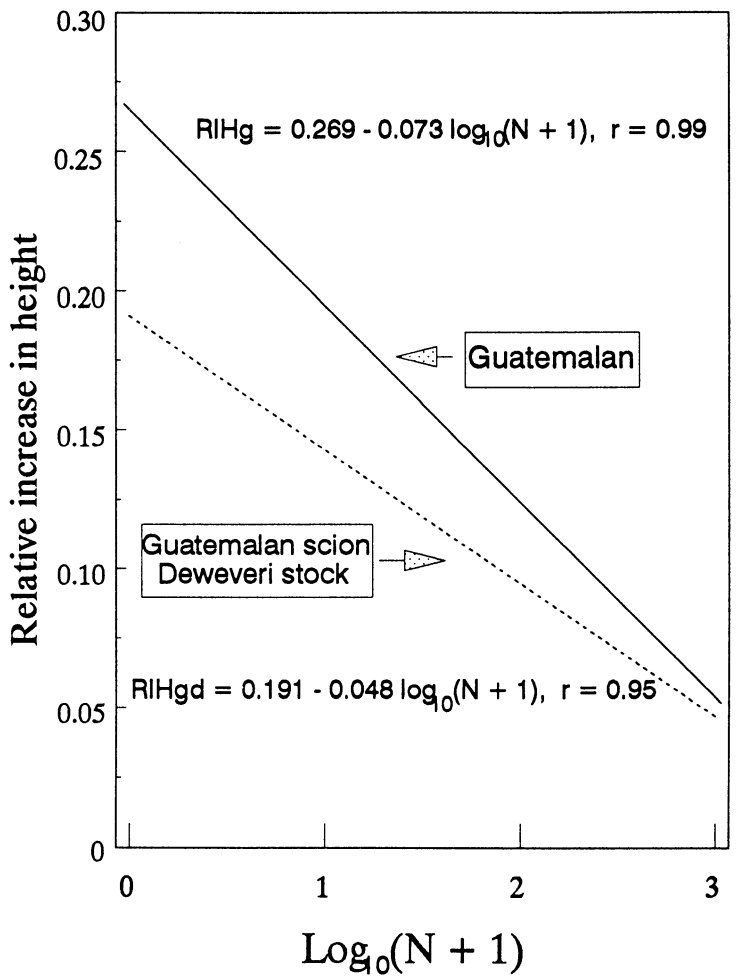

Fig. 2. Relative increase of coffee height in relation to number of *Meloidogyne konaensis* in 1992 and 1993 in a field (average of three sampling times). RIHg = predicted increase in percent of coffee height for cv. Guatemalan; RIHgd = predicted increase in percent of coffee height for Guatemalan scion on Deweveri stock.

Meloidogyne konaensis damaged all coffee cultivars (Tables 1 and 2, Fig. 1). Plant shoot growth was suppressed by all inoculum densities. Minimum predicted shoot height and dry shoot and root weights (average over cultivars) were 35.2, 7.6, and 10.1% of the predicted levels, respectively.

Few J2 were recovered from soil samples at both 2 and 4 mo (0–10/250 cm^3 soil). Galling index and root necrosis were positively related with the inoculum densities (Table 2). More galling and root necrosis occurred in run 1 than in run 2. No galling and very little root necrosis were observed in the noninoculated controls in both runs.

Field experiment. The number of *M. konaensis* J2 in the field was low at the beginning of the experiment on both Guatemalan and Guatemalan scion–Deweveri rootstock. Population densities of J2 increased from November 1992 to June 1993, then decreased from June to September 1993. The mean numbers of J2 were different ($P \leq 0.01$) between the two treatments, with population densities being 1.4–2.6 times greater on Guatemalan than on Guatemalan-Deweveri.

The height, canopy width, and trunk diameter of the coffee trees increased throughout the year; the greatest increase occurred between the first two sampling periods. The tree height of Guatemalan increased 14.5% and the height of Guatemalan-Deweveri increased 13.1%. The relative increases of tree height were 6.9% and 4.9% from March to June 1993, and 13.8% and 6.0% from June to September 1993 for Guatemalan and Guatemalan-Deweveri, respectively.

Meloidogyne konaensis population densities (average of the four sampling periods) were inversely related to coffee growth (Fig. 2). The decrease was greater on Guatemalan than on Guatemalan-Deweveri. The slope values were −0.073 for Guatemalan and −0.048 for Guatemalan-Deweveri. A negative linear relationship was also found between nematode population density and dry root weight of Guatemalan (data not shown). All other measured parameters did not give a significant relationship. Some trees of Guatemalan died because of the high population density of *M. konaensis* whereas none of the Guatemalan-Deweveri died.

DISCUSSION

Meloidogyne konaensis is damaging to coffee. Even the lowest inoculum density of 150 eggs per plant caused substantial galling (20–40%) and root-necrosis (17–44%) on coffee grown in the greenhouse. Since hatch of this nematode is 26% at 30 C (14), approximately 40 J2 are sufficient to damage the root significantly. The impact of reduced coffee growth rate in the field adds strength to the argument that *M. konaensis* is a pathogen of coffee. Using the Seinhorst model, we calculated the threshold to be slightly less than 10 eggs. With 26% as the hatching rate, 2.5 infections per plant seedling is adequate to cause damage. The situation becomes more complex if the tree becomes infected much later. In time, the nematode will probably cause the tree to die. In Kona, trees as old as 100 yr were observed that were dead and heavily galled. Thus, even though coffee is not a good host (13) for *M. konaensis*, it is sensitive.

Meloidogyne konaensis is a potential threat to the coffee industry, especially in plantations at elevations above 400 m where temperatures are favorable (14) for the growth and development of this nematode. There are many hectares of coffee at lower elevations in Hawaii. More studies and surveys should be conducted to identify if this nematode has been distributed to these lower elevation areas.

ACKNOWLEDGMENTS

We wish to thank Philip J. Ito for allowing us to use his rootstock trial for this research, and M. Meisner and staff at the Kona Experiment Station, D. Meyer, and M. Young for their technical assistance in this research.

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