RELATIONSHIP OF *MELOIDOGYNE KONAENSIS* POPULATION DENSITIES TO NUTRITIONAL STATUS OF COFFEE ROOTS AND LEAVES

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ABSTRACT

Hurchanik, D., D. P. Schmitt, N. V. Hue, and B. S. Sipes. 2003. Relationship of *Meloidogyne konaensis* population densities to nutritional status of coffee roots and leaves. Nematropica 33:55-64.

The relationship of population densities of *Meloidogyne konaensis* to nutritional status of roots and leaves of coffee in a commercial field in the Kona district on the island of Hawaii was assessed. The clustered spatial pattern of *M. konaensis* was inversely related to concentrations of K+Ca+Mg in the coffee roots. Other inverse relationships existed between the numbers of *M. konaensis* and concentrations of Mg (roots), Cu (roots and leaves), and Al (roots). From February (first flush of flowering) to May (early stage of fruit development), concentrations of P, K, Mg, K+Ca+Mg, Cu, B decreased in the coffee roots and Zn and Al concentrations decreased in the coffee leaves. The association between foliar nutrient deficiencies in trees associated with high population densities of the nematode is an indication that the nematode is inciting the deficiencies.

Key words: Coffea arabica, coffee, Kona coffee root-knot nematode, *Meloidogyne konaensis*, nematode, nematode distribution, plant nutrition, soil nutrients.

RESUMEN

Hurchanik, D., D. P. Schmitt, N. V. Hue, and B. S. Sipes. 2003. Relación entre densidades de populaciones de *Meloidogyne konaensis* y el estado nutricional de raices y hojas de café. Nematropica 33:55-64.

Se evaluó la relación entre densidades de populaciones de *Meloidogyne konaensis* y el estado nutricional de raices y hojas de café en un campo comercial en el distrito de Kona en la isla de Hawaii. La estructura espacial agrupada de *M. konaensis* era inversamente relacionada a las concentraciones de K+Ca+Mg en las raices del café. Otras relaciones inversas existian entre los números de *M. konaensis* y las concentraciones de Mg (raices), Cu (raices y hojas) y Al (raices). De febrero (primer brote de florecimiento) a mayo (estado temprano de desarrollo de la fruta), las concentraciones de P, K, Mg, K+Ca+Mg, Cu, y B disminuyeron en las raizes del café, y las concentraciones de Zn y Al disminuyeron en las hojas del café. La asociación entre deficiencias nutricionales de las hojas y las densidades altas del nemátodo indica que el nemátodo incita las deficiencias.

Palabras claves: Coffea arabica, café, nemátodo agallador de Kona, Meloidogyne konaensis, nemátodo, distribución de nemátodos, nutrición de plantas, nutrientes del suelo.

INTRODUCTION

Various factors contribute to sub-optimal growth of coffee in the Kona district on the island of Hawaii. These factors include poor soil fertility, low soil acidity, improper irrigation and fertilization regimes (Dean and Beaumont, 1939), stress from pruning, and/or pests and pathogens (Serracin *et al.*, 1999). Plantparasitic nematodes, especially *Meloidogyne konaensis* (Eisenback *et al.*, 1994), seriously limit coffee bean yield (Schmitt *et al.*, 2001; Zhang and Schmitt, 1995). This nematode, present in an estimated 34% of the coffee farms in the Kona districts, caused approximately 60% loss in coffee yield in the 2001 crop (Serracin and Schmitt, unpublished data).

Symptoms of "coffee decline", caused by Meloidogyne konaensis, are manifested in the foliage as nutrient deficiencies and wilting (Serracin et al., 1999). Leaves that emerge by March usually appear healthy; by May, however, symptoms of nutritional deficiencies begin to become evident and intensify during the following weeks. This reflects events that probably occurred in the roots during the first several months of the year or even in previous years. A disease of citrus caused by Tylenchulus semipenetrans also results in nutrient (N, Zn, and Mn) deficiencies in the leaves (Chandel et al., 1998). The concentration of P was lower on Pratylenchus penetrans infected Mahaleb cherry than on nematode-free control plants (Melakeberhan et al., 1997). The purpose of this research was to determine the relationship between the nutritional status of coffee and M. konaensis population densities when plants were flowering and new foliar growth was beginning, and the early phase of fruit development in a field in Kealakekua, Hawaii.

MATERIALS AND METHODS

The relationship between *Meloidogyne konaensis* population density and the nutritional status of *Coffea arabica* cv. Typica land race 'Guatemalan' was evaluated in a commercial coffee field infested with *M. konaensis* (soil type: Kealakekua silty clay loam; order: Andisol; series: Kealakekua; soil classification: Typic hydrantep, thixoptropic, isothermic). This field was located at Greenwell Farms in Kealakekua, Hawaii (elevation 1300-1400 m). Trees were approximately 10-years-old. They were pruned according to the 3-vertical "Kona style" pruning system (Bittenbender and Smith, 1999) and fertilized according to soil test recommendations. Fertilizer was applied once during the experiment in early May just prior to the final sampling.

A 1.600 m² area was selected for sampling. Five transects, spaced six trees apart, were established with 5 sampling points (also spaced six trees apart) per transect. Soil, roots, and leaves were collected twice from each selected tree: 23 February 2000 (first flush of flowering for the season) and 6 May 2000 (early stage of fruit development). Approximately 1000 cm³ soil, including roots, was collected from the top 30 cm of the soil profile from the drip line of each tree. Half of the sample was collected from one side of each tree and the other half from the opposite side of the tree and composited. The fourth pair of leaves from the apex of 3 arbitrarily selected lateral branches was collected for nutrient analysis (Jones et al., 1991). All samples were placed in plastic bags and transported in insulated containers to the laboratory.

In preparation for analysis, each sample was sieved through a screen with 6-mm square openings. The soil passing through the sieve was divided into a 250-cm³ subsample for nematode assay, a 2-g subsample for nutrient analysis and a variable volume of excess soil. The coffee roots removed from the soil were cut into 2.5-cm length pieces, mixed, and divided: one half for nutrient analysis and the other half for nematode assay.

Nematodes were separated from soil by a combination of elutriation (Byrd et al., 1976) and centrifugal flotation (Jenkins, 1964). Roots were placed in a misting system (Seinhorst, 1950) for 5 days to extract nematodes. Tylenchida and Apelenchida were classified to genus, except root-knot nematodes were identified to species. Microbivorous species, consisting primarily of Rhabditida, were grouped together. Mononchidae were also catalogued.

Concentrations of P, K, Ca, Mg, Mn, and Zn were measured in soil and plant tissues. Cu, B, and Al were determined only in plant tissues. Plant tissues and soil samples were analyzed for nutrients by the Agricultural Diagnostic Service Center at the University of Hawaii at Manoa. Leaves and roots were washed with a 2% mild detergent solution (Sonneveld and Van Dijik, 1982), dried at 80°C for 48 hr, and ground into fine particles. From each batch of ground tissues, 0.25 g was ashed in a muffle furnace at 500°C for 4 hours. The ash was dissolved in 1 M HNO₃ and evaporated under a flame-hood at 200°C. Samples were diluted by a factor of 80 with 0.1 M HCl for nutrient analysis (Hue et al., 2000). The soil was air dried at 25°C for 4 days and sieved through a screen with 4 mm square openings. Nutrients were extracted from 2.0 g of soil using Mehlich 3 solution (Mehlich, 1984) for 5 minutes and filtered through a 6S Whatmann filter paper. The Mehlich 3 extractant diluted samples 12.5-fold. The nutrient concentrations were assayed with an inductively coupled plasma spectrometer.

Nematode communities in the soil in February and May 2000 were assessed using absolute population densities, averages of clusters, relative density, relative frequency, and prominence values (Norton and Schmitt, 1978). Population densities of M. konaensis in coffee roots from the two sampling dates were tested for equal variances, averaged, and used as an independent variable for statistical analyses of nutritional status of coffee. Analysis of variance procedures (SAS, SAS Institute, Cary, NC, USA) were performed on the arbitrarily grouped and pooled nematode population densities (log₁₀-transformed) in the roots: low (8 J2/g root, range of 2-18), low-medium (95 J2/g root, range of 26-258), medium-high (429 J2/g root, range of 304-563), and high (1,253 J2/g root, range of 665-2,853). Isopleths were constructed based on these groupings of M. *konaensis* population densities to delineate the spatial pattern of nematodes and nutrients in the field.

RESULTS

Microbivorous nematodes had the highest relative density, relative frequency, and prominence values compared to the other taxonomic groups (Table 1). This group included several species. Of the single nematode species comparisons, values of the three measures for community analysis were highest for an Aphelenchus sp. in February and highest for M. konaensis in May compared to the other six species recovered. The values increased from February to May for M. konaensis and the microbivorous nematodes. They decreased, especially for relative density and prominence value, for most of the other nematodes. Absolute population densities of M. konaensis were similar in February when trees were producing their first flush of flowers for the season and May when fruit development was in the early stages (Fig. 1), except for the mean number of J2 in the high population density cluster in the soil.

The concentration of several soil and plant nutrients changed between February and May. The soil macronutrients P, K, Mg and K+Ca+Mg were 75, 29, 29, and 14% higher, respectively, in May than in February (Fig. 2). The micronutrients Mn and Zn content of the soil increased by 36 and 62%, respectively (Fig. 2).

Even though soil nutrient concentrations increased in the soil, the concentration of three of four macronutrients and Table 1. Relative density, relative frequency, and prominence values (Norton and Schmitt, 1978) of plant-parasitic, microbivorous and mononchidae/250 cm³ soil in a coffee field in Kealakekua, Hawaii during February and May 2000.

Taxon	February 2000			May 2000		
	RD ^x	$\mathbf{RF}^{\mathbf{y}}$	PV^{z}	RD	RF	PV
Meloidogyne konaensis	16.6	19.6	54.1	22.8	18.1	64.1
Tylenchus sp.	9.5	7.7	19.4	2.6	15.5	6.7
Paratylenchus sp.	5.2	16.2	15.3	1.7	14.2	4.2
Helicotylenchus sp.	0.6	2.6	0.7	0	0	0
Radopholus sp.	0	0	0	0.2	2.6	0.2
Microbivorous	40.5	21.3	137.6	68.9	32.3	258.4
Aphelenchus sp.	23.4	20.4	78.0	2.5	7.7	4.5
Aphelenchoides sp.	2.2	8.5	4.8	1.3	9.0	2.5
Mononchidae	2.0	3.4	2.7	0	0	0

^xRelative density (RD) = <u>Mean number of individuals of a taxon in a sample</u> \times 100 Total of mean numbers of all individuals in a sample

 $\frac{1}{3}$ Relative frequency (RF) = $\frac{\text{frequency of taxon}}{\text{sum of frequency of all taxon}} \times 100$

²Prominence value (PV) = absolute density $\sqrt{absolute frequency}$

three of five micronutrients decreased in roots from February to May (Fig. 3). P, K, Mg and K+Ca+Mg decreased 25, 53, 20 and 38%, respectively. The concentration of Cu and B in the roots decreased 17% and 52%. Zn followed a similar trend. Mn and Al concentrations in the roots were similar at the two sampling times (Fig. 3). In the leaves, concentrations of Ca, Zn, and Al increased and Cu decreased from February to May (Fig. 4). These changes were +18% for Ca, +25% for Zn, +33% for Al and -53% for Cu.

The spatial pattern of *Meloidogyne konaensis* was clustered and inversely related to the concentration of K+Ca+Mg in coffee roots (Fig. 5). The clusters were in definable elongated groupings in the down slope direction of the field. Concentration of K+Ca+Mg in the roots (Fig. 5B) was relatively high (5.4%) in areas of the field with low M. konaensis population densities (8 J2/g root) (Fig. 5A) and was relatively low (3.6-4.8%) in coffee roots where numbers of the nematode ranged from 95 to 3,000 J2/g root. The sum of these nutrients decreased linearly in the roots and leaves as nematode numbers increased (Fig. 6A-B). Regression coefficients for numbers of M. konaensis versus the concentration of K+Ca+Mg were -0.64 (P < 0.01) for the roots (Fig. 6A) and -0.33 (P < 0.01) for the leaves (Fig. 6B). The concentration of Mg was also negatively related to M. konaensis population densities and this relationship was best described by a quadratic equation (Fig. 6C).

Of the micronutrients, Cu and Al concentrations in roots and leaves were affected by *M. konaensis*. Cu concentrations

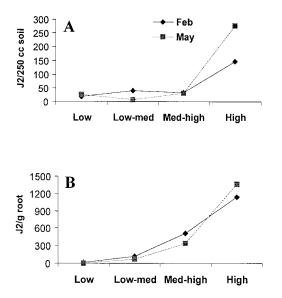


Fig. 1. Mean numbers of *Meloidogyne konaensis* secondstage juveniles grouped into arbitrarily selected population clusters (low, low-med, med-high and high) for February and May 2000: A) soil, and B) roots. Nematodes were collected from Greenwell Farms in Kealakekua, Hawaii.

in the roots averaged 7 ppm higher at the two highest nematode population density clusters than at the two lower clusters over the two sampling periods. Concentration of this nutrient in the leaves had a relationship that was opposite that in the roots. In February, Cu concentration was 20 ppm in leaves of trees at the lowest population clusters and averaged 3 ppm less at the higher population clusters. In May, the concentration was 12 ppm with low numbers of M. konaensis. Cu concentration was 6 ppm at the high population density. The mean concentration of Al was higher in the roots (1900 ppm) than in the leaves (70 ppm). The concentration of Al in roots in the two high population levels was approximately 700 ppm greater than in the two low population density clusters. Al concentration was not affected in the leaves by M. konaensis.

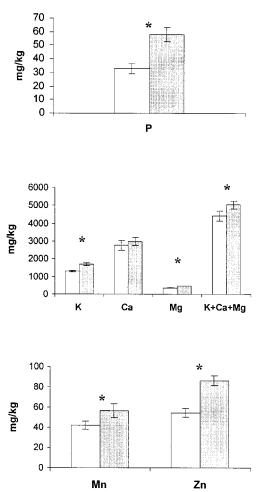


Fig. 2. Soil macronutrient and micronutrient content in a *Meloidogyne konaensis* infested coffee field in Kealakekua, Hawaii. February (white bars) and May (gray bars) 2000. Error bars = standard error. *P < 0.05.

DISCUSSION

Concentration of some nutrients in roots and leaves was altered by *Meloidogyne konaensis*. Deficiency of nutrients in plant tissues, even though there was an adequate and available supply in the soil can be partially explained by the inability of the roots to take up nutrients because of root dam-

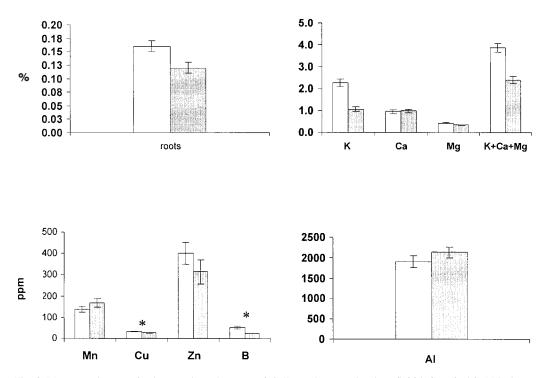


Fig. 3. Macronutrients and micronutrients in roots of *Coffea arabica* growing in a field infested with *Meloidogyne konaensis*. Samples were collected in February (white bars) and May (gray bars) 2000. Error bars = standard error. *P < 0.05.

age caused by M. konaensis. This was the case with okra in which high levels of residual nutrients remained in the soil where root-knot nematode levels were high and root damage severe (Ritzinger and McSorley, 1998). On cherry, concentrations of Mg and Ca were higher in leaves from one of three cultivars infected with Pratylenchus penetrans (Melakeberhan et al., 1997). The opposite result occurred in a greenhouse experiment in which Ca and inoculation of M. konaensis were controlled treatments. Leaves of coffee seedlings (~7-months-old) grown in soil with excessive Ca became Ca deficient in M. konaensis infected plants (Hurchanik, unpublished data). If nutrient behavior is similar in the field to this greenhouse experiment, then the low Ca

concentration in the coffee leaves in our study was probably due to nematode effects since Ca was excessive in the soil.

Zn concentrations in coffee leaves were too low for optimal coffee growth. In an experiment currently in progress to test interactive effects of Zn and M. konaensis, Zn concentrations were also low in coffee leaves even though Zn concentrations in roots were high (Schmitt, unpubl.). The nematode may be altering the translocation and/or partitioning of Zn. Another possible mechanism used by the plant to minimize detrimental effects could be the storage of Zn and other trace elements in the roots with minimal translocation to leaves (Hagemeyer and Breckle, 1996).

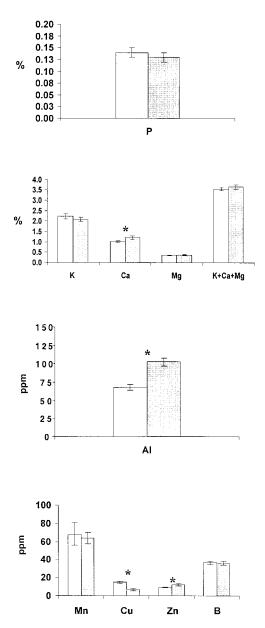


Fig. 4. Macronutrients and micronutrients in leaves of *Coffea arabica* growing in a field infested with *Meloido-gyne konaensis*. Samples were collected in February (white bars) and May (gray bars) 2000. Error bars = standard error. * P < 0.05.

The horizontal spatial distribution pattern of *M. konaensis* was clustered. This

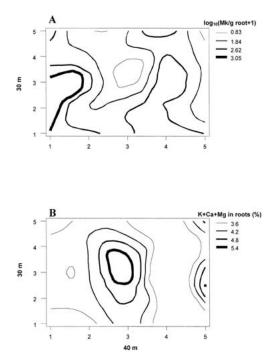


Fig. 5. Isopleths of *Meloidogyne konaensis* population densities and concentration of K+Ca+Mg concentration in the roots of coffee. Nematode population densities from the February and May 2000 samplings were averaged and \log_{10} -transformed; nutrient concentrations were from the February samples. A) *Meloidogyne konaensis*, B) concentrations of K+Ca+Mg in the soil.

type of field distribution is typical for plant-parasitic nematodes (Barker and Nusbaum, 1971; Barker et al., 1985; Noe and Barker, 1985). Clustering was associated with some nutrients (e.g., K+Ca+Mg) and with topography of the field. These types of nematode field distributions have been attributed to the influence of various physical and chemical properties of the soil. Clay content, Na and Cu concentrations were associated with horizontal spatial distribution of М. incognita, Tylenchorhynchus claytoni, and Helicotylenchus dihystera in loamy sands (Noe and Barker, 1985). Soil pH and concentrations of Mg and Cu were correlated with Het-

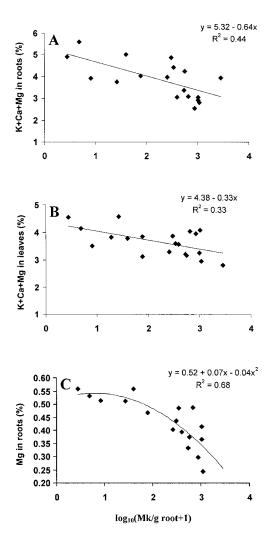


Fig. 6. Relationships between *Meloidogyne konaensis* population densities in the roots and the nutrient levels of A) K+Ca+Mg in the roots (February 2000), B) K+Ca+Mg in the leaves (May 2000), and C) Mg in the roots (February 2000). Nematode numbers from samples collected in February and May 2000 were averaged and log₁₀-transformed.

erodera glycines egg population densities in a Norfolk loamy sand planted with soybean (Francl, 1992). The orientation of clusters of nematodes with the slope of the field site in the direction of water flow during heavy rainfall events is an indication of the impact of soil erosion on moving nematodes within the farm.

The high population density of M. konaensis on coffee at this test site, considering its high damage potential, would account for the severe damage observed on the coffee roots (Zhang and Schmitt, 1995). The high population density of microbivorous nematodes indicates an ample food supply, which most likely were bacteria associated with the damaged and decomposing roots of the coffee. In addition to dving coffee roots, decomposing leaf matter below coffee trees may also be a source of energy for microbivorous Such organic nematodes. materials impact nematode population dynamics (Abawi and Chen, 1998; Conroy et al., 1972; Evans and Haydock, 1993; Powell, 1971).

M. konaensis and coffee nutrition are associated sufficiently to assume that the nematode is inducing nutritional problems for the plant. The grower practice of applying fertilizer to soil associated with "nutrient deficient" plants has not solved the foliar deficiency problem on infested farms. This adds credibility to the hypothesis that the deficiency is nematode induced. Thus, an important step for correcting the nutritional problem in coffee in the Kona area of Hawaii is to control the nematode.

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