ESTIMATING CROP LOSS IN ORCHARDS WITH PATCHES OF MATURE CITRUS TREES INFECTED BY *TYLENCHULUS SEMIPENETRANS*†

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ABSTRACT

Duncan, L. W., P. Mashela, J. Ferguson, J. Graham, M. M. Abou-Setta, and M. M. El-Morshedy. 1995. Estimating crop loss in orchards with patches of mature citrus trees infected by *Tylenchulus semipenetrans.* Nematropica 25:43-51.

Two citrus orchards, identified in 1982 as having patches of trees infected by *Tylenchulus semipenetrans,* were used to study the long-term effect of the nematode on tree quality. In 1992-93, the distribution of infected trees in both orchards continued to be aggregated. Trees were selected in one of the orchards, which had only trace amounts (0-17 juveniles and males/100 cm^3 soil), or high levels (500-9 875 juveniles and males/100 cm^3 soil) of the nematode. Six edaphic and biotic variables measured in soil from the two groups of trees were not different ($P \leq 0.05$). Percent silt was slightly higher (3.0 vs. 1.4%) in soil with trace numbers of nematodes. There were no differences between the two groups of trees in variables that reflect long-term damage (tree height, canopy diam, and canopy density). However, fibrous root mass density, leaf area, and fruit yield were 33, 8, and 23% lower, respectively, on trees heavily infected by the nematode.

Key words: citrus, citrus nematode, crop-loss assessment, spatial patterns, *Tylenchulus semipenetrans.*

RESUMEN

Duncan, L. W., P. Mashela, J. Ferguson, J. Graham, M. M. Abou-Setta y M. El-Morshedy. 1995. Estimación de pérdidas en huertos con parches de árboles de cítricos en producción infestados con *Tylenchulus semipenetrans.* Nematropica 25:43-51.

Dos huertos de cítricos, identificados en 1982 con infestaciones en parche de arboles atacados por *Tylenchulus semipenetrans,* se utilizaron para estudiar el efecto a largo plazo de los nematodos en los concerniente, la calidad del árbol. En 1992-1993 la distribución de los arboles infestados en ambos huertos continuó siendo agregado. Algunos árboles fueron seleccionados en uno de los huertos que poseían bajos niveles de infestación (0-17 juveniles y machos/100 cm^3 de suelo) o altos niveles (500-9 875 juveniles y machos/100 cm^3) de nematodos. Seis variable edáficas y bióticas que se evaluaron del suelo de ambos grupos de árboles no fueron diferentes ($P \leq 0.05$). El porcentaje de arcilla fue ligeramente superior (3.0 vs. 1.4%) en los suelos con bajos niveles de infestación de nematodos. No hubo diferencias entre los dos grupos de árboles en las variables que reflejaban el daño a largo plazo (altura, diámetro de dosel, y densidad de dosel). Sin embargo, la densidad fibrilar radical, área foliar y producción de frutos fueron 3, 8 y 23% más bajos respectivamente, en los árboles que estaban mas severamente infestados por los nematodos.

Palabras clave: cítricos, evaluación de pérdidas de cosecha, nematodos de los cítricos, patrón espacial, *Tylenchulus semipenetrans.*

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INTRODUCTION

The citrus nematode, *Tylenchulus semipenetrans* Cobb, is commonly encountered in citrus-growing regions worldwide. The nematode is well adapted to parasitism on citrus; it has a relatively narrow host-range encompassing only several species of woody plants; it establishes specialized feeding cells in the host; and trees can support very high (hundreds of thousands of nematodes per liter of soil) population densities of citrus nematodes while exhibiting only mild to moderate symptoms of injury (4,14).

Estimates of crop loss due to citrus nematodes vary, depending mainly on how well local conditions favor population development (5,14). In Florida, the nematode is thought to be most damaging to citrus growing in coastal and some inland areas where soils contain higher levels of clay (27), organic matter (22), and salinity (21,23). There is little experimental documentation, however, of the relative response to nematode management of trees growing in these areas compared to trees growing in the light sandy soils of the central ridge region of the state.

Control of citrus nematodes in mature orchards in Florida using fumigant and non-fumigant nematicides has produced highly variable responses in fruit yield and tree quality. Published data indicate that control of the nematode increases fruit yield in the second year following treatment by 15-20% on average (6,26,29). Similar loss estimates have been obtained in South Africa from survey data relating yield to naturally occurring nematode population densities in citrus (30). Both methods of loss assessment have recognized limitations. Even with the addition of certain experimental control treatments (29), it is unknown whether non-target effects of nematicides (on the tree directly or on other organisms that interact with the citrus tree) may have influenced the outcomes of trials (1). In the case of survey data, it is not possible to know whether unmeasured variables that influence nematode densities also affect yield (9).

The short-term response of fruit yield to nematode management may reflect only a portion of yield loss due to the pest. Citrus orchards are long-lived, and we are unaware of reports measuring the long-term effects of nematode parasitism on the size and quality of mature trees. Thus, yield responses to nematode management in mature trees may under-represent yield losses if trees are significantly smaller or otherwise debilitated by nematodes at the beginning of research trials. The difficulty in measuring the effects of nematodes on the size and quality of mature citrus trees is due to the length of time required to establish and conduct such experiments. From the time they are planted, citrus trees may not attain full size for 15-20 years.

A survey conducted in 1982 to estimate the occurrence of citrus nematodes in Florida's citrus industry reported a large number of orchards that contained patchy distributions of infected trees (10,13). These orchards may be potentially useful to measure long-term effects of nematode parasitism on tree size and quality, depending on the validity of several assumptions. First, it is likely that parasitism of infected trees in these orchards was of long duration because populations of *T. semipenetrans* require nearly a decade after introduction to reach damaging levels (5). Second, patches of infected trees likely result from random inoculation events and not from perturbations that reduce populations of nematodes on some trees in the orchard. Finally, if the latter assumption is correct, edaphic conditions between patches of trees with and without nematodes should on average be equivalent. This condition would preclude the existence of unmeasured variables that may affect nematode population density as well as tree quality.

This paper reports the results of two surveys to investigate the distribution of nematodes in orchards that contain infected and apparently non-infected trees. Also reported are comparisons of several measures of tree quality between the two groups of trees.

MATERIALS AND METHODS

Four orchards were selected from those found to have patchy distributions of nematode-infected trees in the 1982 survey (13). All orchards were within 3 km of one another in Indian River County on Florida's east coast and were owned and managed by the same company. In a preliminary survey, 20 trees in a 3-ha area of each orchard were sampled either with shovels or with 2.0-cm-diam × 30-cm-long soil probes, depending on soil texture. Sampling and sample processing methods were described previously (8). Two of the four orchards were selected for the study based on the results of these preliminary samples.

The first orchard contained 27-yr-old 'Hamlin' orange trees (*Citrus sinensis* (L.) Osbeck) on sour orange rootstocks (*C. aurantium* L.). Trees grew in double rows 8.1 m apart (6 m between trees) on beds of soil to increase rooting volume due to high water tables. Trees were irrigated by flooding the furrows created by the beds. No nematicide had ever been used in the orchard. Trees were hedged and topped at 2- to 3-year intervals to facilitate harvesting and grove maintenance operations. Sixty-three trees were sampled in April 1992 and again in April 1993 to confirm their status as infected or apparently non-infected. Trees were selected by choosing every third tree (excluding replanted trees) up to the 25th tree space in 13 consecutive rows. Based on those samples, two groups of trees were selected: those with population densities greater than 500 juvenile and male *T. semipenetrans*/100 cm^3 soil (n = 18) and those with "trace" infections—no detectable nematodes (n = 11) or only one (n = 8) or two (n = 4) juveniles or males detected in the samples from either date.

Soil samples from each tree also were used to measure soil texture (2), soil moisture, soil solution pH, and electrical conductivity (28), fibrous root mass density (12), and propagule density of *Phytophthora nicotianae* Breda de Haan (syn. *P. parasitica* Dastur.) (12). Trees were subjectively rated on a scale of 0-4, depending on canopy density, to estimate tree quality. Tree heights, average canopy radius in two directions, and trunk diameters were measured. Five new, fully expanded leaves were picked from each quarter of the canopy of each tree and areas determined on an area meter (model LI3000, Lambda Inst. Corp., Lincoln, NE). Fruit from each tree were harvested in December 1992 and measured as numbers of 41 kg boxes.

The second orchard was also planted in double rows on beds and flood-irrigated. It consisted of 'Valencia' orange trees (*C. sinensis* (L.) Osbeck), on 'rough lemon' rootstock (*C. limon* (L.) Burm.f.). Ninety trees were sampled in February 1993 and an additional 90 trees one month later. Tree spacing, age, and orchard cultural practices were identical to those of the other orchard. Trees were selected in essentially the same manner, although the distance within rows at which trees were selected varied in different portions of the survey area.

Average values of the edaphic and biotic variables measured in each of the two categories of trees were compared using Student's t-tests. A Fortran program (15) was used to determine the fit (x^2) of the negative binomial model to the frequency distributions of nematode counts in each orchard.

RESULTS

Nematode spatial patterns in orchards: Two of the four orchards evaluated in the preliminary survey were not considered for further observation because nematodes were detected on nearly all of the trees in one orchard and only low levels of nematodes were detected on two trees in the other.

The frequency distributions of nematode counts on trees in the 'Hamlin' and 'Valencia' orchards chosen for further study were very different (Fig. 1). The count distribution in the 'Valencia' block was reasonably well-fit by a negative binomial model (P > 0.90), with 73% of trees supporting no or non-detectable levels of nematodes. Only two trees supported greater than 500 nematodes/100 cm^3 soil. The average population density in infected 'Valencia' trees was 165 nematodes/100 cm^3 soil. No further study was conducted

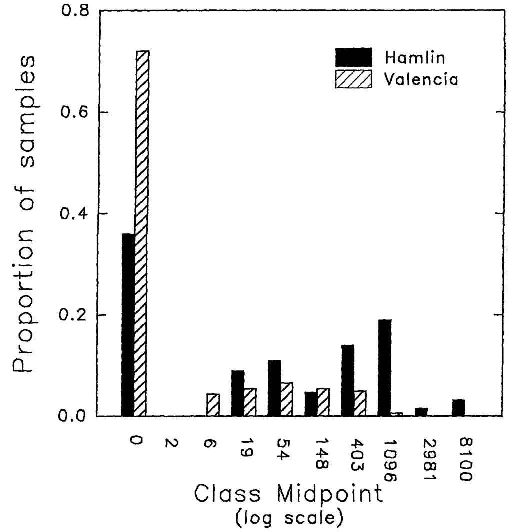

Fig. 1. Frequency distributions of *Tylenchulus semipenetrans*/100 cm^3 soil beneath individual citrus trees in two orchards.

in this block due to the small differences in nematode population densities on individual trees.

The frequencies of counts in the 'Hamlin' block had two peaks at approximately 0 and 1000 nematodes/100 cm^3 soil (Fig. 1). The negative binomial model fit (P ≤ 0.50) these data poorly. No nematodes were ever detected on 17% of the trees.

Table 1. Comparison of edaphic and biotic characteristics of soils from trees infested with high or trace population levels of *Tylenchulus semipenetrans*.

Variable	High	Trace
Moisture (%)	6.0	6.0
pH	6.0	6.0
Ec	494.0	486.0
% Sand	95.7	94.8
% Silt	1.4	3.0*
% Clay	3.0	2.2
Phytophthora nicotianae (propagules/cm^3)	7.8	6.6

*Horizontal means different (P ≤ 0.05) according to Student's *t*-test.

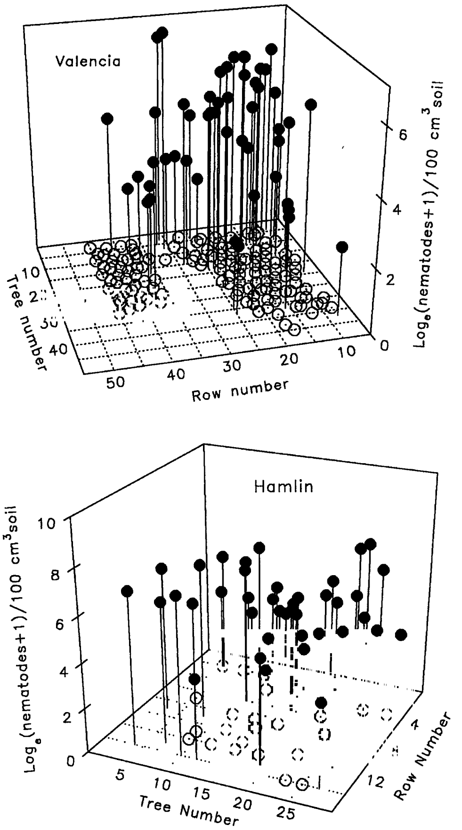

Fig. 2. Spatial patterns of *Tylenchulus semipenetrans*/100 cm^3 in two citrus orchards. Open circles represent trees beneath which no nematodes were detected.

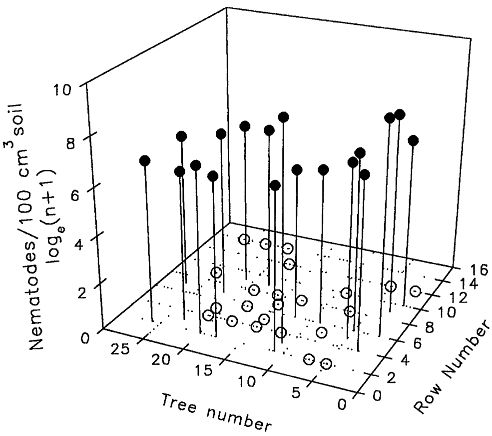

Fig. 3. Spatial patterns of *Tylenchulus semipenetrans* in two classes of trees in an orchard of 'Hamlin' trees growing on sour orange rootstock. Open circles represent trees beneath which only trace levels of nematodes ($\bar{x}$ = 6 nematodes/100 cm^3 soil) were detected. Closed circles represent trees beneath which high nematode population densities were measured ($\bar{x}$ = 1975 nematodes/100 cm^3 soil).

Counts of one or two nematodes/sample (equivalent to 8-17 nematodes/100 cm^3 soil) were detected on 19% of the trees and more than 500 nematodes/100 cm^3 soil on 29% of the trees. Trees with trace infestation levels were nearly always adjacent to sampled trees with non-detectable population densities (Fig. 2). The mean number of nematodes on trees assigned to the high and trace infestation groups was 1975 and 6/100 cm^3 soil, respectively (Fig. 3).

Nematode-infected trees in the 'Valencia' block were aggregated primarily near one end of the rows and on one side of the block (Fig. 2). In the 'Hamlin' block, patches of nematode infected and non-infected trees were dispersed relatively evenly within the block.

Edaphic and biotic differences: No differences ($P \leq 0.05$) were detected between soil from high and trace infestation trees, with respect to soil moisture, content of clay or sand, soil solution pH, electrical conductivity, or propagule density of *P. parasitica* (Table 1). A two-fold difference ($P \leq 0.05$) was found in the percent silt (1.4 vs. 3.0%). There were no differences in the average height (range 2.6-3.4 m), canopy radius (range 3.9-5.3 m), or vigor ratings (range 1.5-3.75) between the two groups of trees, but the average trunk diam of heavily infected trees was 7% greater (36.8 vs. 33.8 cm, $P \leq 0.05$) than of trees with trace infections (Fig. 4A).

Root mass density was 32% lower ($P \leq 0.05$) on heavily infected trees (Fig. 4B). Leaf area was 8% smaller ($P \leq 0.05$) on these trees. Twenty-two percent fewer fruit by volume ($P \leq 0.05$) were harvested from

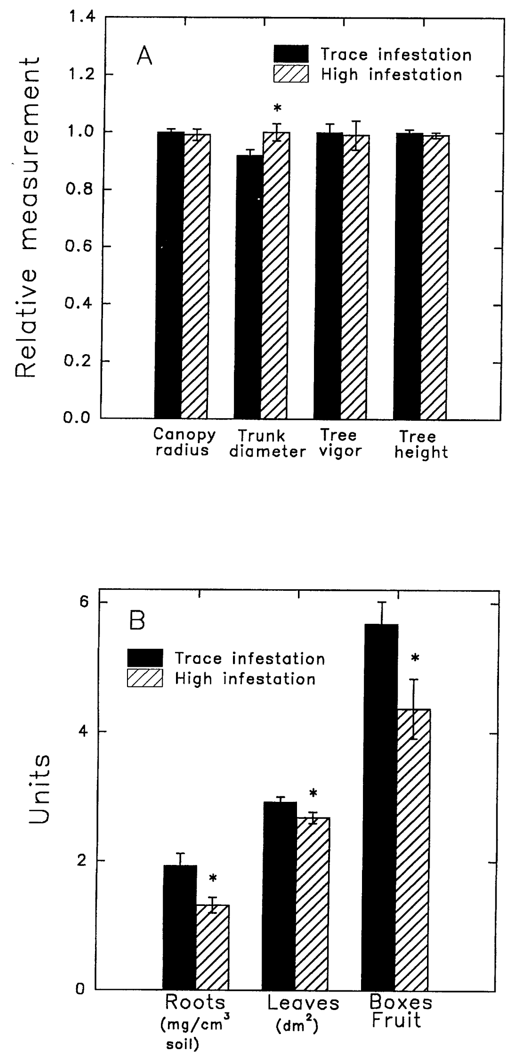

Fig. 4. Differences in (A) canopy radius, trunk diameter, tree height, and tree vigor and (B) root density, leaf area, and fruit yield, between trees with heavy ($\bar{x}$ = 1975 nematodes/100 cm^3 soil) and trace ($\bar{x}$ = 6 nematodes/100 cm^3 soil) infestations of *Tylenchulus semipenetrans.* Relative measurements in (A) obtained by dividing data by the highest mean in each category. Mean differences ($P \leq 0.05$) according to Student's *t*-test, denoted by an asterick.

trees growing in soil heavily infested with the nematode.

DISCUSSION

Differences between trees heavily infected with *T. semipenetrans* and those with trace infection levels appeared primarily in tree organs subject to continuous renewal — fibrous roots, leaves, and fruit. With the exception of trunk diam (which was slightly greater on heavily infected trees), no differences were detected in the size or overall appearance of these mature trees. These observations suggest either that: 1) nematode-infected trees allocated sufficient nutrients to attain full size at the expense of growing roots, leaves, and fruit, or 2) trees were infected after reaching maturity, or 3) periodic hedging and topping of the trees reduced size differences between infected and non-infected trees. The first possibility is not supported by reports that trees planted in soil heavily infested with *T. semipenetrans* grow more slowly than trees in non-infested soil (24) or that growth of young trees with marked potassium deficiency is reduced severely (19). The citrus nematode is known to reduce levels of leaf K in citrus (31, P. Mashela, unpublished). Conversely, variable rates of K fertilization were shown to influence citrus fruit yield, but not canopy surface area during a 9-year period, beginning when trees were 7 years old (18). Thus, depending on the extent of nematode-induced K deficiency in the trees, priority of vegetative over reproducing growth may have occurred. Although there were approximately 30-36% differences in the heights and canopy radii of the trees, respectively, hedging and topping would have at least partially masked potential size effects due to the nematode. Regardless of the phenology of tree infection and effects of cultural practices on tree size, canopy density was not affected by at least a relatively long association with the nematode. Different results might occur in orchards with better over-all tree condition. The 'Hamlin' trees in this orchard were typical of those growing on Florida's east coast; canopies were generally thin due to high water table, salinity, and infection by *P. parasitica.*

The apparent effects of *T. semipenetrans* on roots, leaves, and fruit in the 'Hamlin' block were in reasonable agreement with previous studies. *Tylenchulus semipenetrans* damaged the fibrous root cortex (3) and reduced fibrous root density in mature trees (16). Leaf size has been reported to be reduced by this nematode (17). The magnitude of the yield difference between these groups of trees is consistent with field trials in Florida in which nematodes were controlled with nematicides (6,26).

The fact that the orchards were found to have patchy distributions of trees in 1982 and again a decade later supports the assumption that infected trees were in that condition for a long duration. Other than severe water deficit, we know of no condition that would cause high population levels of citrus nematodes to decline to non-detectable levels. Repeated sampling of the trees in this study showed no indication of population resurgence on trees with non-detectable levels of the nematode or population decline on heavily-infected trees. The similarity of soil conditions for both groups of trees provides no evidence that nematode populations on individual trees were affected differently. The small difference in silt between the two groups of trees could not suppress population densities to non-detectable levels; moreover, silt has been shown to favor population development of *T. semipenetrans* (7). Therefore, it is likely that the patchy distribution of infected trees in these orchards was the result of planting infected trees or contamination from equipment. Although the use of flood-irrigation in these orchards should increase the movement of nematodes compared to sprinkler types of irrigation, movement in irrigation water does not appear to have been great.

The assumption that edaphic and biotic variables in soil would not differ between infected and apparently non-infected groups of trees (due to random inoculation events) was well supported by these data. The lack of correlation between randomly-distributed discrete classes (high vs. trace levels) of nematodes and other soil variables in this study is in strong contrast with other surveys measuring a continuous distribution of naturally occurring *T. semipenetrans* population densities (20).

The methods reported herein appear to be useful to study the long-term effects of some soil pathogens on perennial crops. *Tylenchulus semipenetrans* is a good organism for this approach because it is detectable at low (non-damaging) as well as high population densities (11), populations grow slowly and are relatively stable over long periods of time (5,12), and unaided, this nematode migrates very slowly in the soil (7,25). We found site selection to be costly in terms of both time and labor. However, these costs are offset by gaining better control of extraneous variables in the field without establishing and maintaining long-term experimental treatments.

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