

# Damage Functions for *Meloidogyne arenaria* on Peanut<sup>1</sup>

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**Abstract:** Microplot experiments were conducted in 1989 and 1990 to determine the relationship between yield of peanut (*Arachis hypogaea*) and inoculum density of *Meloidogyne arenaria* race 1. Nine inoculum densities were used, ranging from 0–200 eggs/100 cm<sup>3</sup> soil (1989) or from 0–100 eggs/100 cm<sup>3</sup> (1990), and each density was replicated 10 times. In 1989, higher final densities (mean of 1,171 juveniles [J2]/100 cm<sup>3</sup> soil) were obtained in plots inoculated with 0.5 to 50 eggs/100 cm<sup>3</sup> soil than in plots inoculated with 100 to 200 eggs/100 cm<sup>3</sup> (313 J2/100 cm<sup>3</sup> soil). In 1990, final densities of *M. arenaria* reached high levels ( $\geq 1,111$  J2/100 cm<sup>3</sup> soil) in all inoculated plots. Pod yield and dry weight of foliage at harvest were negatively correlated ( $P \leq 0.05$ ) with inoculum density in both seasons. In 1989, the relationship between pod weight ( $y$ ) and initial density ( $x$ ) was described by Seinhorst's equation, with  $y = 0.088 + 0.91(0.90)^{(x-1)}$  and  $r^2 = 0.826$ . In 1990, the relationship was  $y = 0.22 + 0.78(0.97)^{(x-1)}$  and  $r^2 = 0.794$ . These equations suggest tolerance limits of approximately 1 egg/100 cm<sup>3</sup> soil, which may require specialized methods, such as bioassay, for detection.

**Key words:** *Arachis hypogaea*, bioassay, damage function, *Meloidogyne arenaria*, nematode, peanut, root-knot nematode, Seinhorst's equation, threshold, tolerance limit.

The predominant nematode species damaging peanut (*Arachis hypogaea* L.) in the southeastern United States is *Meloidogyne arenaria* (Neal) Chitwood race 1 (9). Losses to this nematode can exceed 50% in severely infested fields (9). Management with nematicides is possible (11), although results have been inconsistent (3,4). Best results have been achieved by using both a preplant fumigant and a nonfumigant nematicide at peg initiation (3). Recent work has demonstrated the advantages of growing crops that are nonhosts or poor hosts to *M. arenaria* for one or more years preceding peanut production (4,10,12–14).

Because management of *M. arenaria* is expensive, involving growth of alternative crops and (or) nematicide usage, damage thresholds or tolerance limits (17) for *M. arenaria* would be useful, yet few studies have been conducted to establish these levels. In Texas, 8.8 to 16.6 eggs and juveniles (J2) of *M. arenaria* per 100 cm<sup>3</sup> soil resulted in 10% losses in pod yields in microplots (21). Data did not fit Seinhorst's (17) model, but the linear nature of the best-fit regression equations suggests that

threshold densities are low. In Alabama, peanut yields in 16 plots over three seasons were inversely related to *M. arenaria* J2 densities in soil 3 weeks before harvest (16). Yield losses occurred even where nematode densities at that time were low (<50 J2/100 cm<sup>3</sup> soil). Samples collected 3 weeks before harvest are too late to be used for management decisions in the existing crop, however. Yields could not be related to preplant densities because numbers were often very low and not detected by common laboratory extraction methods (16). Densities of *M. arenaria* J2 in soil increase exponentially from low or undetectable preplant densities during the peanut growing season (15). Preliminary data from Florida (analyzed by Duncan's multiple-range test) suggested threshold densities between 50–150 *M. arenaria* per 100 cm<sup>3</sup> in one season, but between 0–2 in another (2).

The objectives of this study were to obtain additional information on the damage threshold density and damage functions of *M. arenaria* race 1 on peanut in microplots (5,17). Additional damage functions were determined by reanalysis of existing data (2).

## MATERIALS AND METHODS

Two separate tests, one each in 1989 and 1990, were conducted in 76-cm-d mi-

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croplots encircled with 60-cm-wide fiberglass sheets inserted 50 cm into the soil (8). Microplots were arranged in rows 1.5 m apart in an Arredondo fine sand (93% sand, 4% silt, 3% clay; pH 5.8; 1% organic matter) treated with 977 kg methyl bromide/ha (98% a.i., 2% chloropicrin) applied broadcast under a 3-mil plastic covering 3–5 months before planting. The soil profile consisted of a sand ( $\geq 90\%$  sand) to a depth of  $>1.2$  m. In each test, the experimental design was a randomized complete block with nine inoculum levels replicated 10 times.

Nematode inoculum for the microplots consisted of eggs of *M. arenaria* race 1 extracted (6) from roots of tomato (*Lycopersicon esculentum* Mill. 'Rutgers') maintained in a greenhouse. In 1989, treatments consisted of 0, 0.5, 1, 5, 10, 30, 50, 100, or 200 eggs/100 cm<sup>3</sup> soil; 1.0 egg/100 cm<sup>3</sup> soil is equivalent to 1,040 eggs/microplot. In 1990, treatment levels were 0, 0.25, 0.5, 1, 5, 10, 30, 50, and 100 eggs/100 cm<sup>3</sup> soil. Eggs were added to each microplot in 1 liter water and mixed into the top 22.5 cm of soil. Overhead irrigation was applied to all plots immediately after addition of inoculum. Dates of nematode inoculation were 25 April 1989 and 7 May 1990.

Four days before nematode inoculation, each microplot was fertilized with 29 g of 0-10-20 (N-P-K). On 25 April 1989 and 9 May 1990, each plot was planted with three seedlings of 'Florunner' peanut. Seeds were germinated in petri dishes on moist filter paper for 6 days before planting, and the roots were sprinkled with a *Rhizobium* spp. inoculant (Nitragin, Lipha Tech, Milwaukee, WI) at planting. Gypsum (30 g per plot) was applied around each plant at early flowering. Chlorothalonil (2 ml/liter) was applied weekly beginning 35 to 45 days after planting for control of early and late leafspots, *Cercospora arachidicola* Hori or *Cercosporidium personatum* (Berk. and Curt.). Weeds were removed manually from the plots, and overhead irrigation was applied as needed.

Soil samples consisting of five cores (2.5-cm-d to a 20-cm depth) were collected from each microplot to determine nema-

tode densities in soil early in the season (Pi), at midseason (Pm), and at harvest (Pf). Sampling dates were 5 May 1989 and 9 May 1990 for Pi, 18 July 1989 and 11 July 1990 for Pm, and 6 September 1989 and 2 October 1990 for Pf. On every sampling date except 9 May 1990, the five soil cores from each plot were mixed, and a 100-cm<sup>3</sup> subsample was processed by a centrifugal-flotation technique (7). On 9 May 1990, a 140-cm<sup>3</sup> subsample was transferred to a plastic cup and planted with a 2-week-old 'Rutgers' tomato seedling. These bioassay plants were maintained in a greenhouse until 30 May, at which time the number of galls per root system were counted and galling was rated on a 0–5 scale (20). Numbers of root-knot nematodes in root systems stained with acid fuchsin (1) were determined by examination with a stereomicroscope.

As an indicator of plant size, diameters of individual plants were measured on 26 June 1989 and 19 June 1990 by measuring the horizontal spread of foliage of each plant in a north-south direction. At harvest (8 September 1989 and 2 October 1990), plants were removed and pods were dried to 7% moisture before weighing. Plants were dried to constant weight in an oven at 60 C.

Correlation coefficients (19) were calculated between yield parameters and log-transformed (base e) nematode densities [ $\log_e(x + 1)$ ]. Unless stated otherwise, all correlations referred to in the text were significant at  $P \leq 0.05$ . A computer algorithm (5) was used to determine the parameters of the Seinhorst equation (17) best fitting each data set, and from this equation, the tolerance limit and the relationship between yield and nematode density were determined. Best-fit Seinhorst equations were also used to relate pod weights to inoculum density for data collected in 1983 and 1984 (2) from the same microplots under experimental conditions similar to those used in 1989 and 1990.

## RESULTS

*Meloidogyne arenaria* J2 densities increased rapidly, reaching high densities in

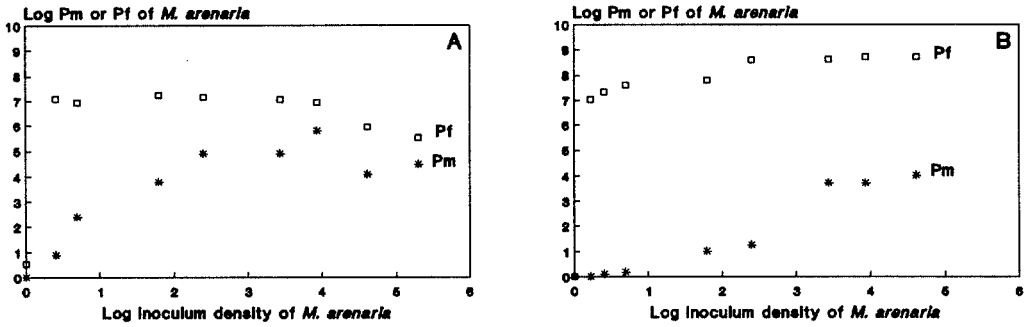


FIG. 1. Relationships between inoculum density of *Meloidogyne arenaria* eggs per 100 cm<sup>3</sup> soil and midseason (Pm) and final (Pf) densities of *M. arenaria* juveniles per 100 cm<sup>3</sup> soil in microplots. Each point is the mean of 10 replications, and both axes are log<sub>e</sub> scales. A) 1989. B) 1990.

all inoculated plots by harvest (Fig. 1A,B). Ten days after planting in 1989, densities of 1.0 or more J2/100 cm<sup>3</sup> soil were found in plots inoculated with 30 or more eggs/100 cm<sup>3</sup>, but densities of only 0.1 J2/100 cm<sup>3</sup> were found in plots receiving 1–10 eggs/100 cm<sup>3</sup> (data not shown). Pm and Pf resulting from inoculum levels of 100–200 eggs/100 cm<sup>3</sup> were lower ( $P \leq 0.05$ ) than those resulting from intermediate (10–30 eggs/100 cm<sup>3</sup>) inoculum levels (Fig. 1A). Plots inoculated with 0.5 to 50 eggs/100 cm<sup>3</sup> soil had a mean Pf of 1,171 J2/100 cm<sup>3</sup> soil, whereas those inoculated with 100–200 eggs/100 cm<sup>3</sup> averaged 313 J2/100 cm<sup>3</sup> soil. In 1990, Pf was much greater than in 1989 (e.g., 5,415 in 1990 vs. 1,152 in 1989 for the 30/100 cm<sup>3</sup> inoculum level). In 1990, Pf was  $\geq 1,111$  in all inoculated plots and increased as inoculum density increased. In 1989, log-transformed Pf in the 80 microplots inoculated with  $\geq 0.5$  eggs/100 cm<sup>3</sup> soil was negatively correlated with log-transformed inoculum density ( $r$

= -0.549); whereas in 1990 the correlation was positive ( $r = 0.649$ ).

In 1989, inoculum density was inversely correlated with plant diameter, foliage dry weight, and pod weight (Table 1). Negative effects of nematodes on yield were less evident in 1990, when pod weight and foliage weight were inversely correlated with inoculum density but plant diameter was not (Table 1). In 1989, relationships between plant growth and inoculum density conformed ( $r^2 > 0.90$ ) to Seinhorst's equations (Fig. 2). In 1989, plant diameters were reduced when inoculum densities were above a tolerance limit of 23 eggs/100 cm<sup>3</sup> soil. Dry weight of foliage declined when inoculum densities were above a tolerance limit of only 1.0 egg/100 cm<sup>3</sup> soil; minimum foliage weight was only 12% of that at maximum predicted levels.

For the relationship between pod weight and inoculum density, the best-fit Seinhorst equation indicated a tolerance limit of only 1.0 egg/100 cm<sup>3</sup> soil in both 1989

TABLE 1. Correlation coefficients of plant growth measurements with log-transformed inoculum densities of *Meloidogyne arenaria* or with bioassay results.

	Plant diameter	Foliage dry weight	Pod weight
Inoculum density, 1989	-0.550**	-0.822**	-0.846**
Inoculum density, 1990	n.s.	-0.264*	-0.553**
Bioassay results, 1990:			
Galls per root system	n.s.	-0.327**	-0.489**
Nematodes per root system	-0.213*	-0.277**	-0.423**
Gall index	n.s.	-0.310**	-0.540**

Correlation coefficients ( $r$ ) based on 88 degrees of freedom. Asterisks (\*, \*\*) indicate significant  $r$  values at  $P \leq 0.05$  and  $P \leq 0.01$ , respectively; n.s. =  $r$  not significant at  $P \leq 0.05$ .

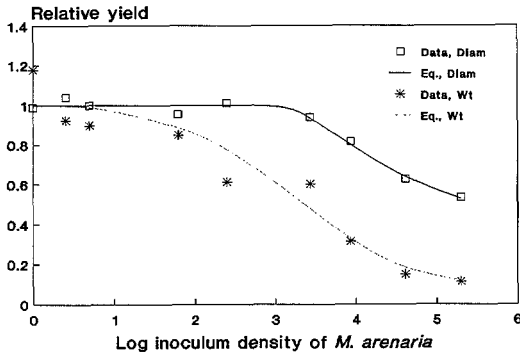


FIG. 2. Relationships between inoculum density of *Meloidogyne arenaria* per 100 cm<sup>3</sup> soil ( $x$ ) and relative yield ( $y$ ) in microplots, 1989. Plant diameter:  $y = 0.51 + 0.49(0.98)^{(x-23)}$  for  $x > 23$  and  $y = 1.00$  for  $x \leq 23$ ; maximum yield 30.4 cm;  $r^2 = 0.985$ . Foliage dry weight:  $y = 0.12 + 0.88(0.97)^{(x-1)}$  for  $x > 1$  and  $y = 1.00$  for  $x \leq 1$ ; maximum yield = 237.6 g;  $r^2 = 0.913$ . Each point (Data) is the mean of 10 replications, and lines (Eq.) indicate Seinhorst equations. The x-axis is log<sub>e</sub> scale.

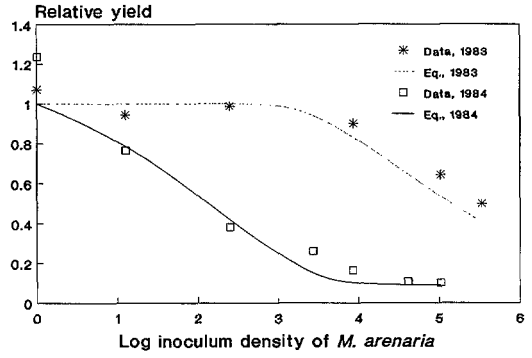


FIG. 4. Relationships between inoculum density of *Meloidogyne arenaria* per 100 cm<sup>3</sup> soil ( $x$ ) and relative yield ( $y$ ) in peanut pods (dry weight) in microplots. 1983:  $y = 0.33 + 0.67(0.99)^{(x-23)}$  for  $x > 23$  and  $y = 1.00$  for  $x \leq 23$ ; maximum yield = 233.8 g;  $r^2 = 0.966$ . 1984:  $y = 0.090 + 0.91(0.90)^x$  for  $x \geq 0$ ; maximum yield = 493.6 g;  $r^2 = 0.912$ . Each point (Data) is the mean of 10 replications, and lines (Eq.) indicate Seinhorst equations. The x-axis is log<sub>e</sub> scale.

and 1990 (Fig. 3). Predicted minimum yield was 42 g in 1990 but only 16 g in 1989, when the slope of the damage function was steeper. When the Seinhorst model was fit to data from 1983 and 1984 (Fig. 4), a tolerance limit of zero eggs/100 cm<sup>3</sup> and a minimum yield of 44 g was predicted for the 1984 data. The least yield reduction occurred in 1983, when a toler-

ance limit of 23 eggs/100 cm<sup>3</sup> and a minimum yield of 77 g was predicted.

In 1990, a bioassay of soil samples collected at the time of planting was used to obtain a better estimate of Pi. Bioassay results correlated directly with inoculum density (data not shown) and inversely with pod weights (Table 1). Seinhorst's equation could not be fit ( $P \leq 0.05$ ) to the relationship between pod weight and root-gall index. However, the relationship between pod weight ( $y$ ) and number of galls on the root system of a tomato bioassay plant ( $x$ ) is given by:  $y = 0.97^{(x-5.5)}$  with  $r^2 = 0.775$ . This suggests that reduction in peanut pod weight would be anticipated when more than 5.5 *M. arenaria* were observed on the root system of a bioassay plant.

### DISCUSSION

For all experiments, the relationship between pod weight and inoculum density conformed to Seinhorst's (17) equation. Thus it was possible to estimate a tolerance limit or threshold density for losses in pod weight. These densities were 1, 1, 0, and 23 nematodes per 100 cm<sup>3</sup> soil for 1990, 1989, 1984, and 1983, respectively. Estimates from three of the four years were in

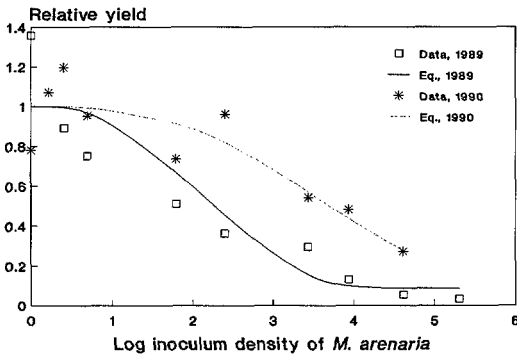


FIG. 3. Relationships between inoculum density of *Meloidogyne arenaria* per 100 cm<sup>3</sup> soil ( $x$ ) and relative yield ( $y$ ) in peanut pods (dry weight) in microplots. 1989:  $y = 0.088 + 0.91(0.90)^{(x-1)}$  for  $x > 1$  and  $y = 1.00$  for  $x \leq 1$ ; maximum yield = 181.5 g;  $r^2 = 0.826$ . 1990:  $y = 0.22 + 0.78(0.97)^{(x-1)}$  for  $x > 1$  and  $y = 1.00$  for  $x \leq 1$ ; maximum yield = 193.0 g;  $r^2 = 0.794$ . Each point (Data) is the mean of 10 replications, and lines (Eq.) indicate Seinhorst equations. The x-axis is log<sub>e</sub> scale.

close agreement; the reason(s) for divergence of the 1983 results are unknown.

Although damage thresholds were not calculated in a related study (21), comparisons of the *M. arenaria* densities needed to cause a 10% reduction in pod weight can be made. In the two microplot experiments in Texas (21), 10% losses were caused by 8.8 and 16.6 nematodes per 100 cm<sup>3</sup>. From the Seinhorst equations presented here (Figs. 3–4), nematode densities per 100 cm<sup>3</sup> soil corresponding to a 10% loss are 5.5, 2.1, 1.1, and 39.1 for 1990, 1989, 1984, and 1983, respectively. Thus, in three of four cases, the levels needed to obtain a 10% yield loss in Florida were lower than those in Texas.

The magnitude of these threshold densities in Florida is of particular concern. In three tests, 1–5 *M. arenaria* per 100 cm<sup>3</sup> soil resulted in 10% pod losses, and thresholds for plant damage were 0–1 per 100 cm<sup>3</sup>, which may be at or near the limits of detection. It is well established that large numbers of *M. arenaria* J2 can build up on peanut from preplant numbers at or below the limits of detection (15,16). Therefore, recognizing (and perhaps lowering) the limits of nematode detection appears to be critical for managing *M. arenaria* on peanut. In our 1989 and 1990 experiments, we used mainly eggs (with a trace of freshly hatched J2) as inoculum, but as Rodríguez-Kábana et al. (15) pointed out, eggs are not detected by commonly used soil extraction methods. Soil sampling of microplots 10 days after inoculation with eggs give some indication of Pi; however, this estimate is not accurate because it assesses only J2 in soil and not eggs or J2 that have penetrated roots. Bioassay provided a more sensitive method for measuring Pi, although the tolerance limit of 5.5 *M. arenaria* on the root system of a bioassay plant is still quite low.

Regardless of the detection methods used, it is apparent that tolerance limits of *M. arenaria* on peanut may, in most seasons, be quite close to the limits of detection. When present, this nematode causes severe damage on deep sandy soils in Flor-

ida (3,4). Of additional concern are observations that large J2 densities may build exponentially from undetected preplant levels (15). Pf data presented here are also based on J2 densities, and not on total population densities. Nevertheless, there appears to be a carrying capacity of about 5,000–6,000 J2/100 cm<sup>3</sup> soil reached in 1990 (Fig. 1B). In 1989, J2 densities reached an upper limit of about 1,200 J2/100 cm<sup>2</sup> for most of the range in Pi (Fig. 1A), with declines at the highest Pi perhaps even suggesting overpopulation (18). Clearly, the carrying capacity appears very different in each season. It is possible that J2 made up a lower proportion of the population density in 1989 than in 1990, if many unhatched (and therefore undetected) eggs were present. Or possibly there is just great seasonal variation in the carrying capacity. In field plots in Alabama, a carrying capacity of 600 J2/100 cm<sup>3</sup> soil was reached after several years of continuous peanut (10). It is not surprising that a higher carrying capacity was observed in microplots than in the field, where nematode parasites and other antagonists would be more abundant. Additional research is needed to better predict the population density buildup and carrying capacity of *M. arenaria* on peanut. Data on the survival of *M. arenaria* over the winter are also essential, because decision making based on counts taken shortly before planting is not practical with *M. arenaria* on peanut growth in deep sandy soils. Based on the present study and on previous work (15), Pf of the previous crop provides the best opportunity to detect *M. arenaria* by typical sampling and extraction methods.

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