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Effect of Host Plant Age on Population Development and Pathogenicity of *Ditylenchus destructor* on Peanut[1]

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Abstract: The effect of inoculating peanut, *Arachis hypogaea* cv. Sellie, with *Ditylenchus destructor* at timed intervals after planting and with different initial nematode population densities (Pi) was tested in greenhouse experiments. Final nematode population densities (Pf) in hulls and seeds were greater (Pf < 0.001) in plants inoculated at or before 9 weeks after planting. Pod disease symptoms correlated positively with the Pf in the pods. The seedgrade of peanuts inoculated at or before 9 weeks after planting was reduced, whereas grade of peanuts from plants inoculated at 15 weeks or later was not reduced. Peanut plants inoculated 12 weeks after planting with a Pi of 10–100 had a lower Pf ($P < 0.05$) than plants with a Pi of 250 to 8,000. Seed of plants with a Pi of 250 or less could be marketed as choice edible seed, whereas those with a Pi of 500 or more were of reduced seedgrade. These results suggest that as few as 500 nematodes per plant at 12 weeks after planting can build up to injurious levels before harvest. A nematicide should therefore be active for longer than 12 weeks after planting to sufficiently suppress the population.

Key words: Arachis hypogaea, Ditylenchus destructor, host age, inoculation, nematode, population dynamics.

Traditionally, nematodes have not been regarded as a serious pest of peanut (*Arachis hypogaea* L.) in South Africa. Since the discovery of *Ditylenchus destructor* Thorne on peanut (5) and the assessment of its damage potential (10) and range of distribution (3), extensive efforts have been made to control this nematode. *Ditylenchus destructor* invades the testa and embryo of peanut seeds, but not the cotyledons (6). The first disease symptoms appear at the pod base where the peg joins the pod. The necrotic tissue is dark brown and has a corky appearance. In a more advanced stage, the dark discoloration extends longitudinally along the veins, just beneath the pod surface (3). In greenhouse experiments, as few as 50 *D. destructor* per seedling caused crop failure due to downgrading of seed of the peanut cultivar Sellie (1). *Ditylenchus destructor* has been reported mainly from temperate regions (3) and is important as a pest of potato tubers and bulbs of flowers (4). It has not been reported in hulls and seeds of peanut outside South Africa.

Peanuts are grown on ca. 200,000 ha annually in South Africa. The most widely grown cultivar, Sellie, is susceptible to *D. destructor.* Although populations of *D. destructor* can be suppressed by nematicides (A. H. McDonald, unpubl. data), the activity of most nematicides lasts for about only 8 weeks (11). Nematodes that survive this period can infect the plants again. The objective of this study was to determine the effect of host plant age and initial population density on the population development of *D. destructor* and the pathogenicity of this nematode.

MATERIALS AND METHODS

Two greenhouse experiments were conducted using the following procedures. Nematode-free peanut seeds, cv. Sellie, were planted in plastic pots filled with 3,000 cm^3 of steam-pasteurized soil (85% sand, 8% silt, 7% clay) supplemented with *Rhizobium* nitrogen-fixing bacteria. Seedlings were thinned to one per pot after emergence. Inoculum of *D. destructor* was obtained from monoxenic cultures of callus tissue initiated from peanut leaves (9) and consisted of nematodes of various life

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stages. Nematodes were pipetted in 10-ml aqueous suspensions into a 10-cm-deep hole in the soil near the roots of the seedling. Holes were filled with sand after inoculation. Water, instead of a suspension of nematodes, was used in control pots. Plants were maintained at 20–25C with a 13-hour photoperiod and fertilized weekly with a hydroponic nutrient solution (6.5% N, 2.7% P, 13% K). Both experiments were randomized complete block designs.

Experiment 1: Peanut plants were inoculated with 4,000 ± 100 *D. destructor* at 0, 3, 6, 9, 12, 15, and 18 weeks after planting to imitate the active period of a nematicide and the new infestation of the nematodes. Five plants from each treatment already inoculated were harvested every 3 weeks from 3 to 24 weeks after planting. At each sampling, fresh root, peg, hull, and seed weights were determined. Nematodes from 5 g fresh roots and 1 g pegs were extracted by the centrifugal-flotation method (2) and from 5 g fresh hulls and seeds by soaking the tissues in shallow water in Petri dishes for 24 hours at room temperature (1). The hulls and seeds of the plants harvested at 24 weeks were indexed for disease severity. Mature pods were rated for disease severity on a 0–10 scale: 0 = clean pod; 2 = black discoloration at either peg or beak end of pod; 4 = black discoloration at both ends of pod; 6 = black discoloration extending along one longitudinal vein joining peg and beak ends of pod; 8 = black discoloration extending along both longitudinal veins joining peg and beak ends of pod; and 10 = black discoloration extending over more than 80% of pod surface. Disease severity of seeds was determined by (no. blemished seeds/total no. seeds) ×10. Blemished seeds were characterized by dark veins and discolored testa (10). Peanut seed were graded according to established regulations (7). Peanut seed with less than 10% blemished seed is graded as choice edible seed, seed with 10% to 20% is graded as standard edible seed, and seed with more than 20% blemished seed is graded as crushing seed.

Experiment 2: Eight plants were each inoculated with suspensions of 0, 10, 100, 250, 500, 1,000, 2,000, 4,000, or 8,000 nematodes per seedling at 12 weeks after planting to determine the maximum inoculum level that could be allowed at this period without suppressing yield loss or downgrading the peanuts. Fresh root, peg, hull, and seed weights were determined at 24 weeks after planting. Nematodes were extracted from the different plant parts and pods were indexed for disease severity as discussed previously.

Data analysis: Nematode data were transformed to log $(x + 1)$ and analysed using analysis of variance for experiment 1 and factorial analysis in experiment 2 with plant part and Pi as main effects. Comparisons of means or groups of means were conducted by Scheffé's method (8). The seed and hull symptoms were correlated with time of inoculation (experiment 1) and inoculum level (experiment 2) by Spearman's correlation analysis (8).

Results

Experiment 1: The average numbers of *D. destructor* in the roots and pegs at the different sampling dates were 73/g and 1,600/g, respectively, and are not discussed. Population densities in hulls and seeds of plants sampled 15 weeks after planting were very low because the pods had just begun to develop. There were no differences among the treatments.

Mean nematode population densities at 18 weeks after planting in hulls of plants inoculated between 0 and 9 weeks after planting were 1,600/g tissue and exhibited little or no change between 21 and 24 weeks after planting (±7,500/g) (Fig. 1A). At all three sampling times, plants inoculated 12 to 18 weeks after planting had lower ($P < 0.001$) population densities than plants inoculated 0 to 9 weeks after planting.

Nematode population development in seeds was similar to those in hulls but with lower population densities. At the 18-, 21-, and 24-week sampling dates, nematode

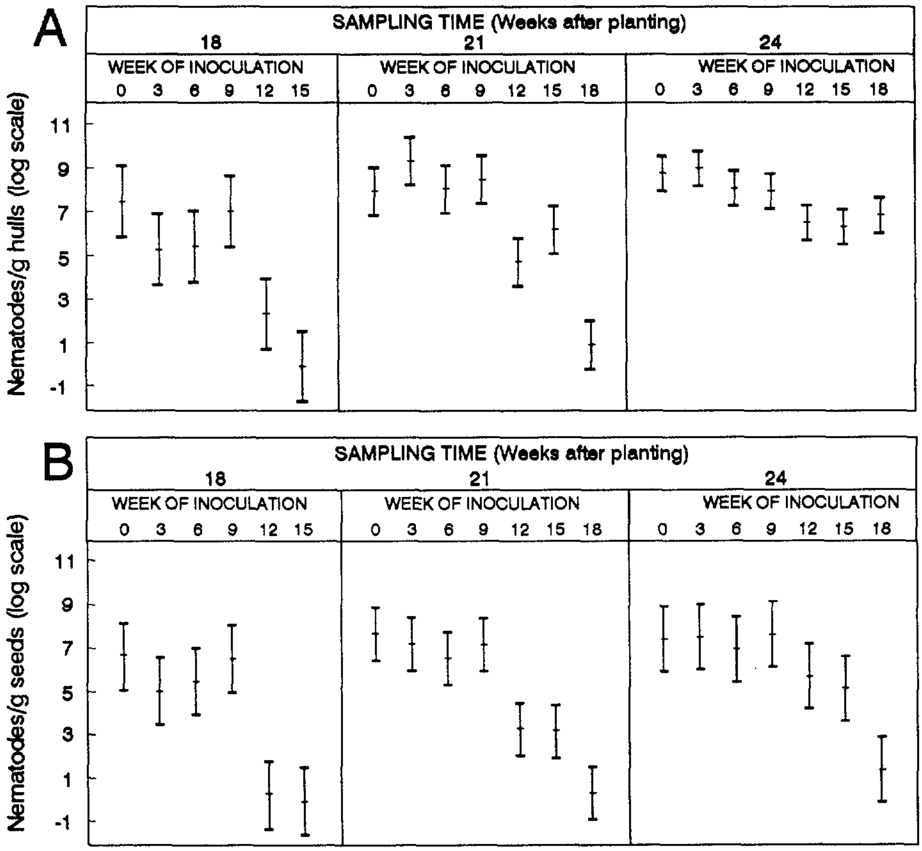

FIG. 1. Effect of time of inoculation on development of populations of *Ditylenchus destructor* in hulls (A) and seeds (B) of peanut in greenhouse tests. Bars indicate 95% confidence intervals.

population densities in seed of plants inoculated at 12 to 18 weeks after planting were again lower ($P < 0.001$) than population densities in seed of plants inoculated at 0 to 9 weeks after planting. At 24 weeks after planting, nematode population densities in seed of plants inoculated at 18 weeks after planting were also lower than those for all inoculation times.

Disease severity of the mature hulls and seeds was determined 24 weeks after planting (Fig. 2). There was a negative correlation ($r = -0.6$; $P = 0.001$) between the hull symptoms and the time of inoculation and between the seed symptoms and time of inoculation ($r = -0.8$; $P < 0.001$). The seeds of plants inoculated at 15 and 18 weeks had a disease severity of less than 1 and were graded as choice edible seed. The seeds of plants inoculated 12 weeks after planting were graded as standard edible seed, because the disease severity was between 1 and 2. The seeds of plants inoculated at 9 weeks or earlier had a disease severity of greater than 2 and were graded as crushing seed.

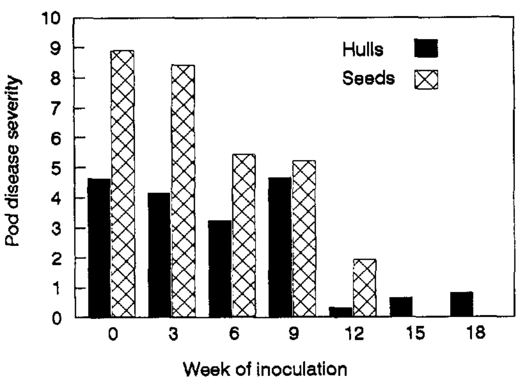

FIG. 2. Effect of time of inoculation of *Ditylenchus destructor* on pod disease severity of peanut at 24 weeks after planting.

Experiment 2: The number of nematodes recovered from the roots (ca. 100/5 g) and pegs (ca. 2,000/5 g) were lower than those in the hulls (ca. 8,000/5 g) and seeds (ca. 5,000/5 g). A similar pattern of nematode population development occurred in both the hulls and seeds, with greater ($P < 0.001$) numbers of nematodes in the hulls than in the seeds (Fig. 3). The hulls and seeds of plants inoculated with 10 or 100 nematodes had fewer ($P < 0.05$) nematodes than those inoculated with 250 or more.

A low correlation ($r = 0.54$; $P < 0.01$) between seed symptoms and inoculum level of 1,000 nematodes or less was observed, and no correlation ($r = 0.03$; $P = 0.88$) was observed at inoculum level of 1,000 nematodes or more (Fig. 4). No correlation was found between hull symptoms and the number of nematodes inoculated. Seeds of the plants inoculated with 250 or fewer nematodes had a disease severity of less than 1 and would be graded as choice edible seed, whereas seeds of the plants inoculated with 500 or more had a disease severity of 1 to 4 and would be graded standard edible or crushing seed.

Discussion

The South African Oil Seeds Board has certain requirements whereby peanuts are classed into choice, standard, crushing and undergrade seed, for which decreasing prices are paid. The percentage of unsound, blemished, soiled, and shrinked nuts, inter alia per sample, are taken into consideration in this grading system to fix a price for the specific delivery of peanuts. Venter et al. (10) describe the effect that *D. destructor* has on yield reduction, including downgrading of the seed.

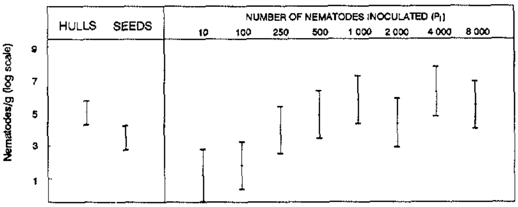

FIG. 3. Effect of initial level on population densities of *Ditylenchus destructor* in hulls and seeds of peanut at 24 weeks after planting when inoculated at 12 weeks after planting. Bars indicate 95% confidence intervals.

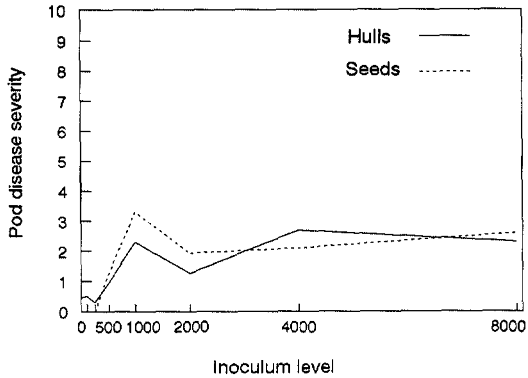

FIG. 4. Effect of initial inoculum levels of *Ditylenchus destructor* on pod disease severity of peanut at 24 weeks after planting, when inoculated at 12 weeks after planting.

Although no pods were found on plants before 9 weeks after planting, these trials showed that inoculation of 4,000 *D. destructor* at this stage will result in a reduction in grade of seed from choice edible to crushing seed. Indeed, inoculum of only 500 *D. destructor* at 12 weeks after planting would also cause sufficient damage to reduce the grade of the seed. These findings indicate that if a nematicide is active for only 12 weeks after planting and as few as 500 nematodes per rhizosphere were found in the soil at this time, the population would be capable of building up to injurious levels before harvest. Most nematicides currently under investigation have active periods of approximately 8 weeks after application. Only two nematicides, fenamiphos and terbufos, are currently registered on peanuts in South Africa. They are applied at planting as preventive systemic nematicides and remain active for approximately 6 and 13 weeks, respectively (11).

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