

Reproductive and Damage Potential of *Ditylenchus destructor* on Peanut¹

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Abstract: The reproductive and damage potential of *Ditylenchus destructor* on peanut, *Arachis hypogaea* cv. Sellie, was determined in greenhouse tests. Final nematode population densities (Pf) in roots, hulls, and seeds increased ($P = 0.01$) as a function of increasing initial population (Pi). Final population densities were higher in hulls than in seeds and roots. Final densities in hulls and seeds were positively ($P = 0.01$) correlated. Fresh root and hull weight and number of pods and seeds per plant were not affected by *D. destructor*. Second generation germination and pod and seed disease severity increased ($P = 0.01$), whereas fresh seed weight decreased ($P = 0.01$) as a function of increasing Pi, and Pf in seeds and Pf in hulls. At Pi 250 and higher, 10–25% of seeds germinated into second generation seedlings before harvest. At Pi 250 and higher, fresh weight of harvested seed was suppressed 20–50%. At Pi 50 or Pf greater than 20 per seed, pod disease severity was 3–7 (on a scale of 1 to 10) and 15–80% of seeds were blemished or unsound.

Key words: *Arachis hypogaea*, *Ditylenchus destructor*, pathogenicity, peanut, population dynamics, yield loss.

Nematodes are important pests of peanut (*Arachis hypogaea* L.) in many production areas of the world (13). *Meloidogyne arenaria* (Neal) Chitwood, *M. hapla* Chitwood, *Pratylenchus brachyurus* (Godfrey) Filipjev & Schuurmans-Stekhoven, *Belonolaimus longicaudatus* Rau, and *Criconebella ornata* (Raski) Luc & Raski are considered the most important nematodes limiting peanut yield worldwide. These nematode species attack the roots, pegs, and hulls, but not the seeds of peanut. However, formation, appearance, quality, and yield of seeds are affected through the suppression of root growth, disruption of peg and pod development, and increased infection by fungi. *Aphelenchoides arachidis* Bos, also known as the peanut-testa nematode, caused no injury to roots of peanut in Nigeria, but it affected seed appearance and weight (4). Symptoms caused by *D. destructor* on peanut seeds are similar to those caused by *A. arachidis* (12). Annual yield

losses of peanut due to nematodes are ca. 12% worldwide (14).

In South Africa, peanut is grown on about 200,000 ha annually. Nematodes were not considered serious pests of peanut in South Africa until *Ditylenchus destructor* Thorne was isolated from roots, pegs, hulls, and seeds from the Transvaal Province (11). A subsequent survey showed *D. destructor* to be present in all major peanut-producing areas (2). *Ditylenchus destructor* caused no visible lesions on roots, but it affected the appearance of pods and seeds. Infected hulls showed brown necrotic tissue at the point of connection with the peg and dark brown to black discoloration along the longitudinal veins. Infected seeds were usually shrunken with dark brown to black micropyles and flaccid testae with dark vascular strands (7). In heavily infested fields, approximately 40–60% of pods and seeds showed symptoms (7). *Ditylenchus destructor* has not been reported on peanut outside South Africa. In many parts of the world *D. destructor*, also known as the potato-rot nematode, is an important pest of potato tubers and bulbs of flowers (9).

The purpose of this study was to determine under greenhouse conditions 1) the effect of initial population densities (Pi) of *D. destructor* on final population densities (Pf) in roots, hulls, and seeds of peanut and 2) the effect of *D. destructor* Pi and Pf on number of pods and seeds per plant; sec-

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TABLE 1. Reproduction of *Ditylenchus destructor* in soil, roots, hulls, and seeds of Sellie peanut against initial nematode density (Pi), 140 days after planting.

Pi (nemas/pot)	Nemas/pot soil	Nemas/root system	Nemas in hulls/plant	Nemas in seeds/plant	Rate of nematode increase (Pf/Pi)	
					in hulls/plant	in seeds/plant
10	390	188	3,771	322	377.1	32.2
50	276	781	15,261	735	305.2	14.7
100	161	2,210	38,846	675	388.5	6.8
250	69	1,808	33,400	774	133.6	3.1
500	804	1,722	61,474	6,575	122.9	13.2
1,000	4,318	3,871	130,511	7,956	130.5	8.0
2,000	1,309	2,880	118,853	10,179	59.4	5.2
4,000	1,444	1,539	47,351	4,159	11.8	1.0
8,000	643	3,864	106,796	10,086	13.3	1.3

Numbers are means of eight replicates.

ond generation germination before harvest; fresh root, hull, and seed weight; and hull and seed disease severity of peanut.

MATERIALS AND METHODS

The following procedures were used in two greenhouse experiments, unless otherwise indicated. Nematode-free seeds of peanut cultivar Sellie were planted in 3-liter (20-cm-d) plastic pots filled with steamed sandy soil (93% sand, 4% silt, 3% clay) augmented with *Rhizobium* nitrogen-fixing bacteria. Pots were placed in a completely randomized design. Seedlings were thinned to one per pot 14 days after planting and inoculated with *D. destructor* 21 days after planting. Inoculum of *D. destructor* of various life stages was obtained from monoxenic cultures on callus tissue initiated from peanut leaves (17). Nematodes were pipetted in 10-ml aqueous suspensions into depressions in the soil around the roots of seedlings. Plants were fertilized weekly by irrigation with tap water containing a hy-

droponic nutrient solution (6.5% N, 2.7% P, 13% K), and maintained at 20–25 C with a 13-hour photoperiod. Plants were harvested 140 days after planting. Mature pods were rated for severity of disease symptoms 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. Mature pods were opened and disease severity of the seeds was determined by (N blemished or unsound seeds/total N seeds) × 10. Blemished seeds were characterized by a dark micropyle, dark veins, and discolored testa. Unsound seeds were characterized by a hypocotyl of 1–2 mm and were distinguished from second-generation seedlings with established root and shoot. Second-genera-

TABLE 2. Regression equations of reproduction of *Ditylenchus destructor* on Sellie peanut against initial nematode density (Pi), 140 days after planting.

<i>D. destructor</i>	Equation	
N/root system	$= (e^{1.169+0.840[\log_e(Pi+1)]}) - 1$	$r = 0.725, P = 0.01$
N in hulls/plant	$= (e^{2.367+1.230[\log_e(Pi+1)]}) - 1$	$r = 0.846, P = 0.01$
N in seeds/plant	$= (e^{0.188+1.056[\log_e(Pi+1)]}) - 1$	$r = 0.784, P = 0.01$
Rate of nematode increase (Pi/Pf)		
in hulls/plant	$= (e^{7.852-0.531[\log_e(Pi+1)]}) - 1$	$r = 0.912, P = 0.01$
in seeds/plant	$= (e^{4.066-0.538[\log_e(Pi+1)]}) - 1$	$r = 0.825, P = 0.01$

TABLE 3. Initial nematode density (Pi) of *Ditylenchus destructor* against fresh root, hull and seed weight; number of pods and seeds per plant; pod and seed appearance; and second-generation germination of seeds of Sellie peanut, 140 days after planting.

Pi (nemas/ pot)	Fresh wt. (g/plant)			Fresh seed weight/plant (%)†	Pods/ plant	Seeds/ plant	Disease severity		Second- generation germination (%)
	Root	Hull	Seed				Pod‡	Seed§	
0	3.79	4.45	9.02	100.0	10.1	18.0	0.00	0.00	0.6
10	5.03	5.04	10.50	116.4	12.0	21.3	0.74	0.55	0.0
50	5.36	4.80	9.10	100.9	10.8	19.3	2.98	1.52	5.2
100	3.92	4.59	7.84	86.9	10.3	17.9	3.74	3.84	2.2
250	3.88	4.02	7.28	80.7	9.1	15.8	4.33	2.39	10.1
500	4.50	3.55	5.30	58.8	7.9	13.3	6.13	5.74	21.8
1,000	4.76	4.61	6.65	73.7	9.5	16.5	6.75	6.47	21.2
2,000	3.23	4.48	4.65	51.6	7.6	13.5	6.34	7.56	25.2
4,000	3.48	4.48	6.41	71.1	8.1	15.7	6.80	5.15	22.9
8,000	3.64	5.39	5.81	64.4	9.0	15.1	6.56	7.94	23.2

Numbers are means of eight replicates.

† Fresh seed weight of infected seeds expressed as a percentage of the fresh seed weight of uninfected seeds.

‡ Based on a scale where 0 = clean pod and 10 = black discoloration extending over more than 80% of pod surface.

§ Number of blemished or unsound seeds per plant divided by total number of seeds per plant multiplied by 10.

tion seedlings were lost in the field at harvesting.

Nematodes were extracted from 200-cm³ soil subsamples by a modified decanting and sieving method (8) with 710- μ m-pore and 45- μ m-pore sieves, followed by the centrifugal-flotation method (10). Nematodes were extracted from 5 g fresh roots by an adapted centrifugal-flotation method (5). Nematodes were extracted from 5 g fresh hulls and seeds by soaking the tissues in shallow water in petri dishes for 24 hours at 22 C (3).

Nematode numbers were transformed (log_e) before statistical analyses. Initial and final nematode densities; fresh root, hull, and seed weight; number of pods and seeds per plant; second-generation germination; and pod and seed disease severities were compared using regression analyses.

In experiment 1, plants were inoculated with 0, 10, 50, 100, 250, 500, 1,000, 2,000, 4,000, and 8,000 *D. destructor* per pot (Pi). Treatments were replicated eight times. At harvest, second-generation seedlings and number of pods and seeds (quiescent and unsound) per plant were counted; fresh root, hull, and seed weights were determined; pods were indexed; and blemished and unsound seeds were counted to determine disease severity of the plant. Nematodes were extracted from one subsample each of soil, fresh roots, hulls, and seeds per pot and counted.

In experiment 2, 50 plants were inoculated with 100 *D. destructor* per pot and another 50 with 1,000 to obtain a range of Pf. Twenty uninoculated plants served as controls. At harvest, mature pods from the inoculated plants were indexed for disease

TABLE 4. Regression equations of seed weight, pod and seed appearance, and second generation germination of Sellie peanut against initial density (Pi) of *Ditylenchus destructor*, 140 days after planting.

	Equation		
Fresh seed weight/plant (g)	$= (e^{2.443 - 0.065[\log_e(Pi+1)]}) - 1$	$r = -0.803$	$P = 0.01$
Fresh seed weight/plant (%)†	$= (e^{4.770 - 0.073[\log_e(Pi+1)]}) - 1$	$r = 0.800$	$P = 0.01$
Pod disease severity‡	$= (e^{0.189 + 0.238[\log_e(Pi+1)]}) - 1$	$r = -0.813$	$P = 0.01$
Seed disease severity§	$= (e^{0.003 + 0.253[\log_e(Pi+1)]}) - 1$	$r = 0.791$	$P = 0.01$
Second-generation germination (%)	$= (e^{-0.121 + 0.451[\log_e(Pi+1)]}) - 1$	$r = 0.907$	$P = 0.01$

† Fresh seed weight of infected seeds expressed as a percentage of the fresh seed weight of uninfected seeds.

‡ Based on a scale where 0 = clean pod and 10 = black discoloration extending over more than 80% of pod surface.

§ Number of blemished or unsound seeds per plant divided by total number of seeds per plant multiplied by 10.

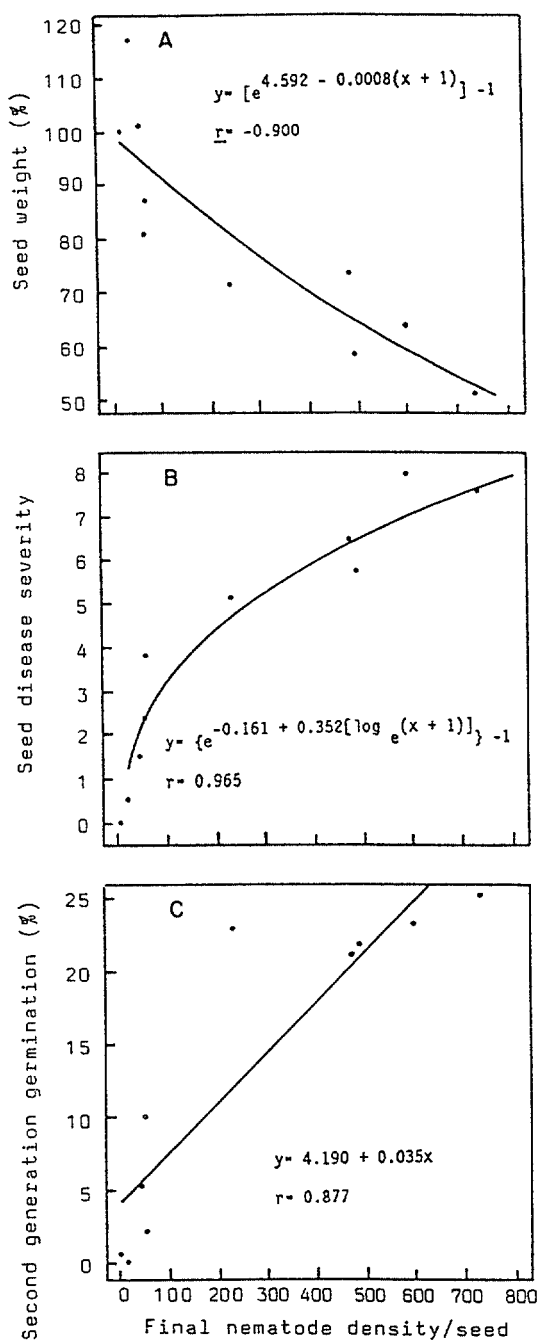


FIG. 1. Relationship of final density of *Ditylenchus destructor* per seed. A) Fresh seed weight (expressed as a percentage of fresh seed weight of uninfected seeds). B) Seed disease severity. C) Second-generation germination (expressed as a percentage of total number of seeds per plant) of Sellie peanut, 140 days after planting. Significant at $P = 0.01$.

severity, bulked, and grouped into six classes as described earlier. Within each class, pods and seeds were counted, fresh hull and seed weights were determined, and blemished and unsound seeds were counted to determine the seed disease severity of the plant. Nematodes were extracted from 10 fresh hull and seed subsamples per class. The mature pods of the uninoculated plants were treated similarly and classed as controls.

RESULTS

Experiment 1

Reproductive potential: Final population densities of *D. destructor* in roots, hulls, and seeds per plant increased ($P = 0.01$) as a function of increasing Pi (Tables 1, 2). The observed Pf were highest at Pi 1,000 (roots and hulls) and Pi 2,000 (seeds) whereafter they plateaued or decreased. Final population densities in hulls were 9.3 (at Pi 500) to 57.5 (at Pi 100) times higher than in seeds and 17.6 (at Pi 100) to 41.3 (at Pi 2,000) times higher than in roots. At Pi 500 and higher, Pf in seeds per plant were 2.1–3.8 times higher than in roots. Reproductive rates (Pf/Pi) of *D. destructor* in hulls and seeds were inversely related ($P = 0.01$) to Pi (Table 1) and are described by multiplicative model regression equations (Table 2).

Damage potential: Fresh root and hull weights and numbers of pods and seeds per plant were not affected ($P = 0.01$) by *D. destructor* (Tables 3, 4). Fresh seed weight decreased ($P = 0.01$), whereas pod and seed disease severity and seedling germination increased ($P = 0.01$) as a function of increasing *D. destructor* Pi. A Pi of 250 and higher suppressed fresh seed weight 20–50%. At Pi 50 and higher, pod disease severity was 3–7 and 15–80% of seeds were blemished or unsound. Second-generation germination ranged from 10–25% at Pi 250 and higher.

Fresh seed weight decreased ($P = 0.01$), whereas seed disease severity and seedling germination increased ($P = 0.01$) with increasing *D. destructor* Pf per seed (Fig. 1A–

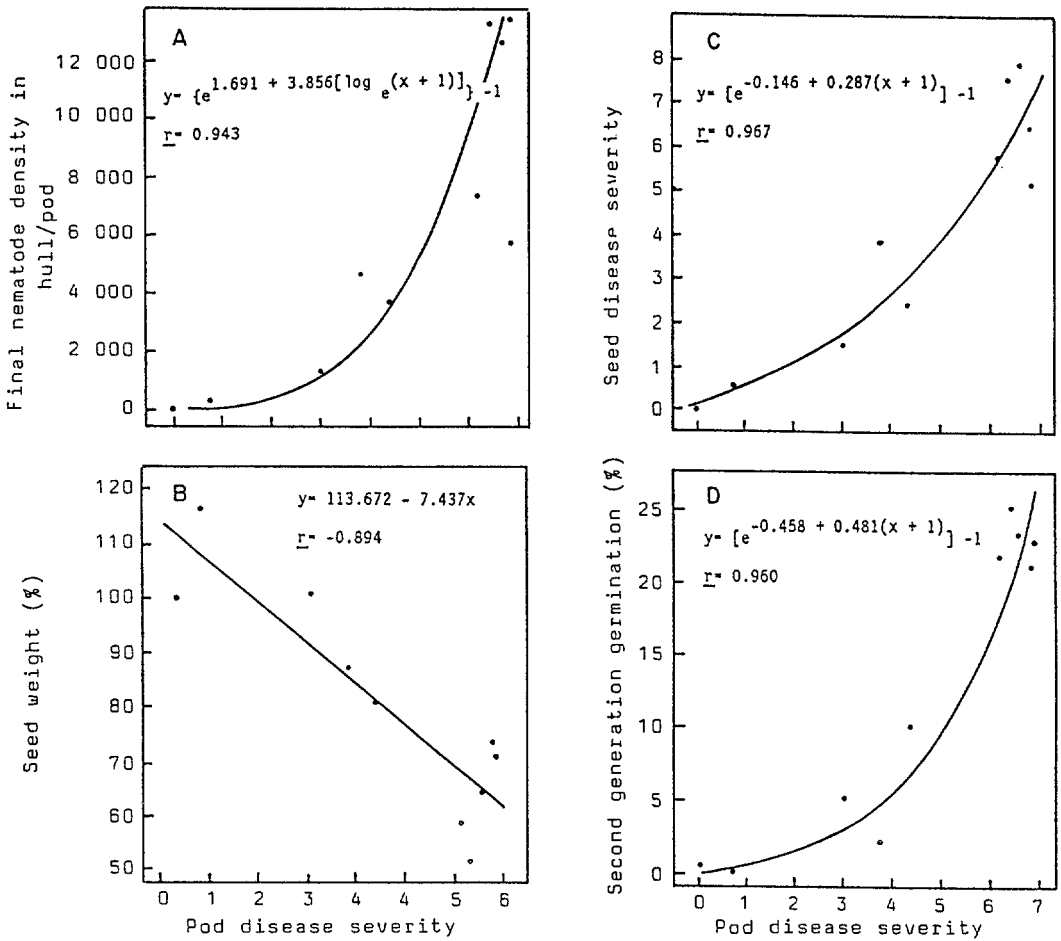


FIG. 2. Relationship of pod rating. A) Final densities of *Ditylenchus destructor* in hulls per pod. B) Fresh seed weight (expressed as a percentage of fresh seed weight of uninfected seeds). C) Seed disease severity. D) Second-generation germination (expressed as a percentage of total number of seeds per plant) of Sellie peanut, 140 days after planting. Significant at $P = 0.01$.

C). Pod disease severity increased ($P = 0.01$) with increasing *D. destructor* Pf in hulls per pod (Fig. 2A). Pod disease severity was negatively ($P = 0.01$) correlated with fresh seed weight (Fig. 2B) and positively ($P = 0.01$) correlated with seed disease severity (Fig. 2C) and second-generation germination (Fig. 2D).

Experiment 2

Reproductive potential: Final population densities of *D. destructor* in hulls per pod were 1.6 (pod disease severity 4) to 4.6 (pod disease severity 8) higher than in seeds per pod (Table 5).

In experiments 1 and 2, a relationship existed between Pf of *D. destructor* in hulls

and seeds. This relationship was described by a multiplicative model regression equation: $y = (e^{-1.024+0.714[\log_e(x+1)]}) - 1$ ($r = 0.823$, $P = 0.01$) in experiment 1, and $y = e^{-0.317+0.820[\log_e(x+1)]} - 1$ ($r = 0.993$, $P = 0.01$) in experiment 2 ($x = \text{Pf/g hulls}$, $y = \text{Pf/g seeds}$).

Damage potential: Fresh hull weight per pod and numbers of pods and seeds per plant were not affected ($P = 0.01$) by *D. destructor*. Inoculation with 0, 100, and 1,000 nematodes per plant resulted in pod disease severities of 0, 2.44 ± 2.19 , and 6.93 ± 1.77 , respectively. Fresh seed weight decreased ($P = 0.01$) and seed disease severity increased ($P = 0.01$) with increasing Pf per seed (Fig. 3A, B).

TABLE 5. Mean numbers of pods and seeds and numbers *Ditylenchus destructor* in hulls and seeds of Sellie peanut per pod disease index class, 140 days after planting.

Pod disease severity†	Pods	Seeds	Nemas in hulls/pod‡	Nemas/seed§	Nemas in seeds/pod
C	139	247	0	0	0
0	206	371	24	8	14
2	84	156	699	127	236
4	52	89	843	305	522
6	209	381	3,722	532	970
8	301	521	6,054	767	1,328
10	23	37	3,691	838	1,348

Numbers are the means of 10 replicates.
 † Rated as follows: 0 = clean pod; 2 = black discoloration at one end; 4 = black discoloration at both ends; 6 = black discoloration along one longitudinal vein; 8 = black discoloration along both veins; and 10 = black discoloration over more than 80% of pod surface.
 ‡ Nematodes in hull/pod ($e^{0.384+4.57[\log_e(x/2+2)]} - 1$, $r = 0.976$, $P = 0.01$ (x = pod disease severity).
 § Nematodes/seed ($e^{0.129+3.698[\log_e(x/2+2)]} - 1$, $r = 0.983$, $P = 0.01$ (x = pod disease severity).
 || C = uninoculated pods.

Final population densities in hulls and seeds (Table 5) increased ($P = 0.01$) with increasing pod disease severity. Pod disease severity was negatively ($P = 0.01$) correlated with fresh seed weight (Fig. 4A) and positively ($P = 0.01$) correlated with seed disease severity (Fig. 4B).

DISCUSSION

Population densities of *D. destructor* increased and caused severe damage on Sellie peanut under greenhouse conditions. These results are consistent with field observations where large numbers of *D. destructor* were associated with high yield losses.

Inoculation of peanut callus tissue with 50 *D. destructor* juveniles and adults isolated from peanut resulted in a 600-fold increase in nematode numbers within 5 weeks (17). In our study, the rate of nematode increase at Pi 50 in soil, roots, hulls, and seeds combined was 341-fold. The true rate of nematode increase was probably much higher, since nematodes inside the pegs were not counted and soaking fresh hulls and seeds for 24 hours in tap water yields only about 50% of the total population (3). The short life cycle of *D. destructor*, 6–7 days at 28 C

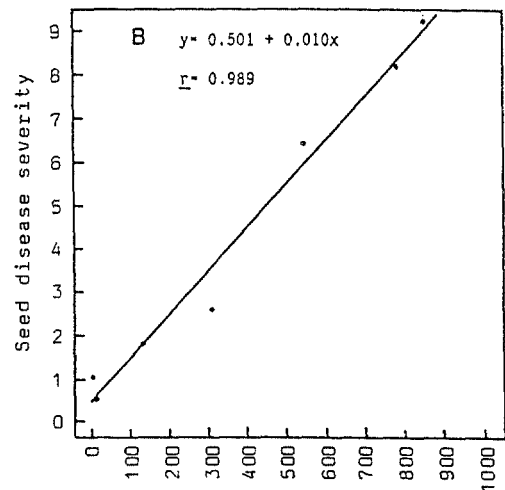
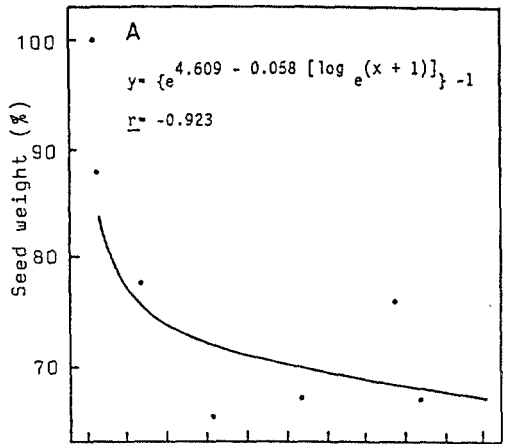


FIG. 3. Relationship of final density of *Ditylenchus destructor* per seed. A) Fresh seed weight (expressed as percentage of fresh seed weight of uninfected seeds). B) Seed disease severity of Sellie peanut, 140 days after planting. Significant at $P = 0.01$.

(6), can explain this high reproductive potential.

Ditylenchus destructor varied in ability to reproduce in the different plant parts. Final populations in hulls were much higher than in seeds. A histopathological study (12) showed that *D. destructor* feeds on all tissues of the hull except the thin mesocarp layer, whereas feeding in the seed occurs only on the embryo and the thin testa surrounding the cotyledons.

Ditylenchus destructor affected the yield of peanut seed through increased germination into second-generation seedlings be-

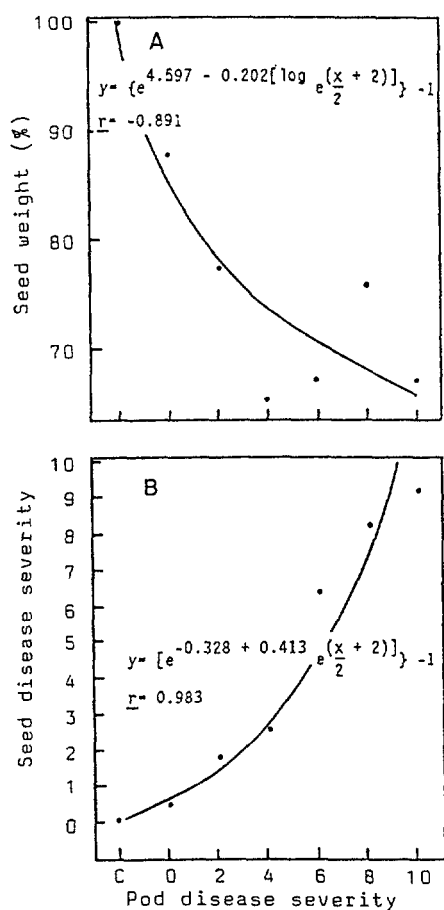


FIG. 4. Relationship of pod rating. A) Seed weight (expressed as percentage of fresh seed weight of uninfected seeds). B) Seed disease severity of Sellie peanut, 140 days after planting. Significant at $P = 0.01$. C = uninoculated plants.

fore harvest, suppression of fresh weight of harvested seeds, and increased blemished and (or) unsound seeds. Response to infection increased with increasing Pi and Pf. Our results provide preliminary models for predicting *D. destructor* population development and damage and for identifying economically important levels. Quantitative differences in models between the two experiments can be attributed to the different techniques used. The models based on the results of experiment 1, with the widest range of Pi, are preferred when making predictions. Further research on the influence of environmental, agronomic, and edaphic factors is required to refine the models.

The economic importance of *D. destructor* on peanut is determined by the loss in income from a peanut field infested with the nematode. This loss depends on the peanut seed grading prices. In South Africa, all peanut seeds are traded through the Oilseeds Board and are graded according to specific regulations (15). These regulations require that a consignment of peanut seed containing more than 15% blemished or unsound seeds is downgraded from edible or crushing seed to undergrade seed. Current (1988–89) market prices for edible and crushing seed are \$309/t and \$197/t, respectively (conversion R1.00 = \$0.35, Aug. 1989). The value of undergrade seed is less than \$4/t. In the greenhouse, a Pi of 50 *D. destructor* in the rhizosphere of a peanut seedling and a Pf of 20 *D. destructor* per seed were required to reach this damage limit. In the field, the average Pf of *D. destructor* per seed in 640 infected samples of blemished or unsound seeds from all major peanut-producing areas in South Africa was 160 (7).

A Pi of 50 *D. destructor* per 3,000 cm³ soil is near the reliable detection level. Little information is available on the survival strategy of *D. destructor* in the soil in the absence of a host plant. If *D. destructor* survives in the soil as eggs (16), or undergoes complete dehydration and enters a state of anhydrobiosis, specialized and extremely accurate techniques will be necessary for preplant soil population assays. As an alternative for soil population assays, indicator plants may be grown in soil for 6–10 weeks. The use of several maize (*Zea mays* L.) and grain sorghum (*Sorghum vulgare* Pers.) genotypes, recently identified as moderate hosts for *D. destructor* (1), should be investigated as bioassay plants. During the growing season, however, peanut pod disease indexing may be utilized to monitor nematode populations and damage. Our results show that pod disease indexes are correlated with *D. destructor* population densities in hulls and seeds, second-generation germination, fresh seed weight, and seed disease severity. Peanut pods are formed throughout the growing season,

and the first mature ca. 100 days after planting. At a critical time, early matured pods could be indexed and the disease index used for estimating damage at harvest. On the basis of this assessment, a management program could then be implemented. The advantage of this method is that it is easy, rapid, and more reliable than pre-plant soil population assays and it is not based on predictions of environmental parameters. Our data shows that an average pod disease index of 2 indicates that 15% or more of the seeds inside the pods are blemished or unsound.

In South Africa, the annual production of peanut averaged about 0.6 t/ha over the last decade, which is lower than in most other peanut-producing countries. The lower yield until now has been attributed almost solely to drought in dryland fields and black pod rot, caused by *Chalara elegans* NagRaj and Kendrick, in irrigated fields. Our results show that *D. destructor* could also play an important role in limiting peanut yield in South Africa.

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