Influence of Root Biomass on Number of *Pratylenchus penetrans* Within Host Roots

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ABSTRACT

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The influence of alfalfa root biomass on the number of *Pratylenchus penetrans* in alfalfa roots was examined. Differential root biomasses were obtained by planting different numbers of seeds in 120 cm^3 of autoclaved loamy sand. Nematodes were either present in the soil at planting or were added to the soil surface 2 days later. A negative correlation existed between root biomass and the number of *P. penetrans* recovered per gram of root tissue. Knowledge of this phenomenon is of importance for the proper interpretation of population densities of endoparasitic nematodes in relation to predictive crop loss assessment.

Root biomass has been recognized as potentially important in the understanding of nematode-incited plant diseases. Limited research, however, has been conducted to test the effect of root biomass on nematode density in roots. Seinhorst and Kozlowska (7) proposed that the greater tolerance of older vs younger carrots to *Rotylenchus uniformis* was probably due to an increase in root biomass relative to nematode inoculum density. O'Brien and Fisher (5) offered a similar explanation to account for the lower level of disease that occurs on barley rather than wheat growing in soil infested with *Heterodera avenae.* They suggested that barley develops more seminal roots during early seedling growth and therefore may contain fewer larvae per root tip. In another study, 1-or 5-wk-old apple seedlings in pots were inoculated with *Pratylenchus penetrans* (3). Two weeks later, the younger seedlings contained significantly more nematodes per gram of root and were more severely stunted than were the older seedlings. Again, the investigators proposed that the larger root system of the older seedlings resulted in fewer penetrations per gram of root.

In the above studies, differences in root quantity were accompanied by differences in root quality (ie, young and old apple or carrot seedlings differ anatomically, whereas, barley and wheat differ genetically). The present investigation examined the effect of root biomass on numbers of nematodes in roots of alfalfa. The evaluation used methods which minimized differences in root quality.

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MATERIALS AND METHODS

Point infestation. One-hundred and twenty cubic centimeters of an autoclaved loamy sand (3) containing 8% water was placed in 150-ml beakers. The beakers were tapped lightly to settle the soil, and water was then added to increase the moisture content to 16% (−0.1 bar, the inflection point for this soil's moisture characteristic and therefore the optimum water potential for nematode movement [9]). One, two, six, or 12 alfalfa seeds (*Medicago sativa* L. 'DuPuits') were planted symmetrically in a shallow ring (2.2 cm in diameter) inscribed on the soil surface. The seeds were pushed a few millimeters into the soil, and each beaker was sealed in a plastic bag containing a moist paper towel. There was a minimum of five replications per treatment. Because preliminary experiments indicated higher variability in the data obtained from the 1- and 2-seeds-per-beaker treatments, additional replications (up to eight) were used for these treatments as materials were available. Germination ranged from 80–100% and therefore the number of seedlings per beaker often was less than the number of seeds planted. The beakers were randomly positioned in a growth chamber operated at 20 C and with 15 hr of 21 klux light per day. Two days later, approximately 750 *P. penetrans* in 1 ml of water (larvae and adults grown on alfalfa callus) were pipetted onto the surface of the soil at the center of the beaker. Because the seeds had been positioned in a circle, all seedlings were located 1 cm from the point of infestation and 2 cm from the closest beaker wall. The amount of inoculum added was determined by counting the nematodes in five 1-ml samples. Four days after the addition of nematodes, the seedlings were removed, washed, and weighed. Each root was placed in a 5-cm-diameter petri dish containing 5 ml of water. Six days later the nematodes in the water were counted. Nematode recovery was assumed to be a function of the number of nematodes within the roots. Nematodes were not extracted from the soil. The point infestation trial was repeated once.

Dispersed infestation. Loamy sand (3) infested with *P. penetrans* was obtained from an 8-L pot containing an alfalfa plant infected with *P. penetrans*. The top of the plant was removed, and the roots were cut into ~2-mm segments and mixed into the soil. All procedures other than the method of nematode infestation were as described for the point-infestation trials. The experiment was terminated 6 days after seed placement. Variation in the amount of inoculum added per beaker was determined by adding infested soil to five beakers and extracting the nematodes from the soil in a modified Baermann funnel. The dispersed infestation trial was repeated once.

RESULTS AND DISCUSSION

In the point infestation experiments, the number of *P. penetrans* recovered per beaker was a linear function of root biomass (Fig. 1A), with the total recovery increasing with increasing root biomass. There was a negative correlation ($r = -0.92$, $P = 0.001$), however, between root weight per beaker and the number of *P. penetrans* recovered per gram of root (Fig. 1B). The variation in numbers of nematodes, correlation coefficients, and slopes of the regression lines are recorded in Table 1.

These results indicate that the nematodes dispersed among the roots present and therefore caused proportionately more infections in beakers containing small root biomasses than in beakers containing large root biomasses. Parasite motility is the essential character responsible for this phenomenon. The motility of *P. penetrans* and other nematodes is well documented (4,6,8–10). However, the significance of movement over small distances possibly has been underestimated, because root biomass is seldom used as a variable in root invasion studies. In this study, differential root biomasses were obtained by planting different numbers of seeds in the same volume of soil. Varying root biomass by changing plant age, genotype, or history would likely yield similar results.

These data are clearly relevant to pot experiments; ie, root biomass can be expected to influence the number of penetrations and thus the severity of disease which occurs at a given density of nematodes in soil. If a similar relationship between root biomass and penetration occurs in the field, it may help explain the observations of a number of research workers that young plants are more severely damaged by plant parasitic nematodes than are older plants (1).

In a hypothetical system consisting of one highly motile nematode (ie, a nematode which can easily move to and penetrate one root) and different root biomasses (one to 10 roots per beaker), one would expect the probability of a particular root being penetrated to be inversely proportional to the number of roots per beaker (Probability = No. of Roots $^{-1}$). For example, the probability of an individual root being penetrated would be 1.00, 0.50, 0.33, and 0.10 in the 1-, 2-, 3-, and 10-roots-per-beaker treatments, respectively. A log-log transformation would transform the curve into a straight line with a slope value of -1 (log probability $= -1$ log no. of roots). The data reported in this paper fit a similar model. However, the slope value of the transformed line was always less negative than -1. It is likely that factors which favor nematode movement would shift the slope value toward -1 whereas factors inhibiting nematode movement would shift the slope value toward 0. In fact, a slope value of 0 would be expected had the nematodes been stationary.

TABLE 1. Effect of root biomass on the number of *Pratylenchus penetrans* recovered per gram of root from point and dispersed infestation experiments[a]

Infestation method	Trial no.	Nematodes (no. added/beaker ± standard deviation)	Correlation coefficient (r)[b]	Slope
Point	1	757 ± 16	−0.92	−0.71
	2	736 ± 51	−0.90	−0.65
Dispersed	1	75 ± 9	−0.91	−0.57
	2	149 ± 50	−0.55	−0.33

[a] Different root biomasses were obtained by planting 1, 2, 6, or 12 alfalfa seeds per beaker. The number of *P. penetrans* recovered per gram of root was calculated by dividing the number of *P. penetrans* recovered per beaker by the corresponding root weight per beaker.

[b] Correlation coefficient based on transformed data (log-log). All values are significant at $P \leq 0.01$.

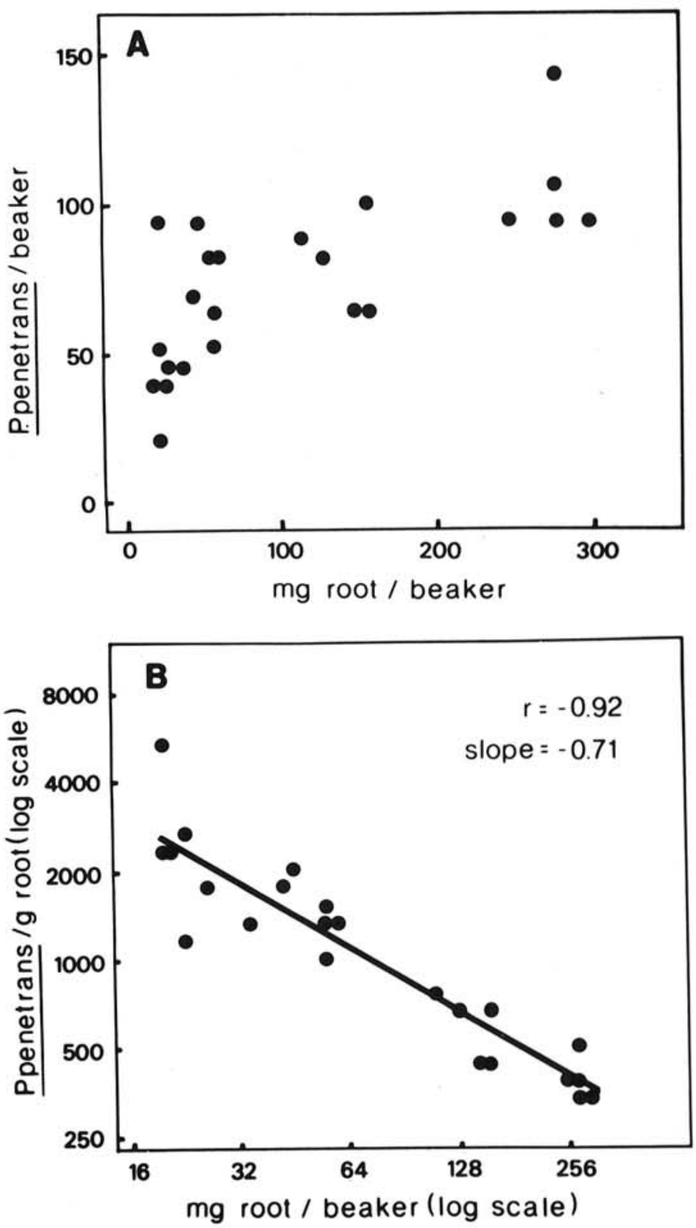

Fig. 1. Effect of root biomass on the number of *Pratylenchus penetrans* recovered **A,** per beaker **B,** per gram root. Different root biomasses were obtained by planting 1, 2, 6, or 12 alfalfa seeds per beaker. In this trial, nematodes were added at a point on the surface of the soil. Results from all trials are summarized in Table 1.

A root exudate or other factor of root origin which oriented nematode movement toward roots (attraction) might also shift the slope value toward −1. Although most studies indicate that attraction is limited to several centimeters (2), Prot (6) reports that *Meloidogyne* larvae may move vertically toward tomato roots as much as 50 cm in 3 days. Degree or distance of attraction may be a function of root biomass; ie, a large root system in the field may draw nematodes from a proportionately greater distance than does a small root system. If this is the case, the results reported here would be relevant only to enclosed systems in which attraction is limited by container size.

Wallace (10) discusses several ways in which nematode population density in roots may influence subsequent penetration. For example, attraction to roots may increase following initial penetration; penetration may be facilitated by movement through a previously formed penetration hole; and a heavily penetrated root may become less attractive. Density-dependent factors such as these may have influenced the observed relationship between root biomass and penetration.

The slope values in the dispersed-infestation experiment (−0.57,−0.33) appeared to be less negative than in the point infestation experiment (−0.71, −0.65). However, statistical analysis was not performed on the combined data because those of trial 2 of the dispersed-infestation experiment were considerably more variable than those of the other trials. A difference in slope values might be expected due to the relationship between the number of seeds per beaker and the mean distance between the nematodes and closest root. In the dispersed-infestation experiment the mean distance between the nematode and closest root decreased as more roots were added per beaker. In the point infestation the mean distance between nematode and closest root was not influenced by the number of roots added. It follows that in the one-seed-per-beaker treatment in the dispersed-infestation experiment, most nematodes would have to move farther to penetrate than in the same treatment of the point-infestation experiment. This difference would lessen as more seeds per beaker were added.

The experiments were terminated shortly after planting, both to minimize the effect of *P. penetrans* on root growth and to minimize plant-plant interactions which might differentially affect root quality. It was assumed that *P. penetrans* did not affect plant growth. Regarding plant-plant interactions, a suppression of root growth was noted in two trials in which 12 seeds per beaker were planted. For example, in trial 2 of the dispersed-infestation experiment, the mean root weight per seedling was 21.7, 22.4, 19.7, and 16.9 mg in the 1, 2, 6, and 12 seeds per-beaker treatments, respectively. This reduction in growth rate may have been associated with qualitative changes that may have inhibited or increased nematode penetration.

LITERATURE CITED

1. BARKER, K. R., and T. H. A. OLTHOF. 1976. Relationships between nematode population densities and crop responses. Annu. Rev. Phytopathol. 14:327-353.
2. GREEN, C. D. 1971. Mating and host finding behavior of plant nematodes. Pages 247–266 in: B. M. Zuckerman, W. F. Mai, and R. A. Rohde, eds. Plant Parasitic Nematodes, Vol. II. Academic Press, New York.
3. JAFFEE, B. A., and W. F. MAI. 1979. Growth reduction of apple seedlings by *Pratylenchus penetrans* as influenced by seedling age at inoculation. J. Nematol. 11:161-165.
4. KABLE, P. F., and W. F. MAI. 1968. Influence of soil moisture on *Pratylenchus penetrans.* Nematologica 14:101-122.
5. O'BRIEN, P. C., and J. M. FISHER. 1978. Factors influencing the number of larvae of *Heterodera avenae* within susceptible wheat and barley seedlings. Nematologica 24:295-304.
6. PROT, J-C. 1976. Ampliltude et cinétique des migrations du nématode *Meloidogyne javanica* sous l'influence d'un plant de tomate. Cah. ORSTOM, ser. Biol. 11:157-166.
7. SEINHORST, J. W., and J. KOZLOWSKA. 1977. Damage to carrots by *Rotylenchus uniformis,* with a discussion on the cause of increased tolerance during development of the plant. Nematologica 23:1-23.
8. TOWNSHEND, J. L., and L. R. WEBBER. 1971. Movement of *Pratylenchus penetrans* and the moisture characteristics of three Ontario soils. Nematologica 17:47-57.
9. WALLACE, H. R. 1958. Movement of eelworms 1. The influence of pore size and moisture content of the soil on the migration of larvae of the beet eelworm, *Heterodera schachtii* Schmidt. Ann. Appl. Biol. 46:74-85.
10. WALLACE, H. R. 1966. Factors influencing the infectivity of plant parasitic nematodes. Proc. R. Soc. Ser. B. 164:592-614.