Colonization and Sex Ratios of *Pratylenchus alleni* in Soybean Roots under Two Soil Moisture Regimes[1]

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Abstract: Population size and sex ratios of *Pratylenchus alleni* in soybeans were studied under two different moisture regimes in Hagener loamy fine sand. Soil moisture was maintained from field capacity to 50% below field capacity in the dry regime and from field capacity to 25% above field capacity in the wet regime. The initial peak of colonization of soybeans by *P. alleni* was in the top 5-cm of taproot 14 days after seeding. There were more *P. alleni* per unit length of taproot in the dry than in the wet regime during the first 7 days, and this trend continued in the top 5-cm of the taproot for 21 days. Nematode density was greater in taproots than in fibrous roots. The ratio of males to females recovered from roots was significantly higher in the dry than in the wet regime.

Soil moisture is doubtless among the more important variables governing nematode populations. There are, however, few data regarding moisture and nematode populations, especially with *Pratylenchus* spp. Kable and Mai (6) found that population increase of *P. penetrans* was greatest at moderate soil tensions (*p*F 2–3) and that there was an interaction with soil moisture and type; the greater the amount of silt and clay present, the greater was the soil moisture tension necessary for good population growth.

Large numbers of *P. alleni* Ferris have been found in some soybean fields in Iowa, but little is known about the parasitism of this nematode on any crop (4, 5). The data reported herein resulted from studies on the colonization of soybeans by *P. alleni* under two moisture regimes.

MATERIALS AND METHODS

Hagener loamy fine sand, from Keokuk County, Iowa, naturally infested with 185 ± 20 *Pratylenchus alleni*/250 cc soil was passed through a five-mesh sieve. The soil was mixed thoroughly and placed in 10.2-cm diam pots, each planted with four seeds of *Glycine max* (L.) Merrill Amsoy soybeans. Moisture was maintained at approximately field capacity for the first two days to promote uniform seed germination. After that the two moisture regimes, as calculated from field capacity determinations (14), were maintained in all tests from field capacity to 50% below field capacity in the dry regime and from field capacity to 25% above field capacity in the wet regime. The soil moisture tensions (Fig. 1) were determined by the gravity method (14) for low moisture tensions, and by the pressure membrane apparatus (11) for higher moisture tensions. The dry regime soil was moistened to field capacity and the pots and contents were weighed two to six times daily, depending upon temperature, sunlight, and growth stage of the plant. When weighing indicated that the soil was at 50% field capacity, water was added to restore field capacity. Appropriate weighings and moisture adjustments were also made with soil in the wet regime. Each moisture treatment group contained three replicate pots for each sampling period of 4, 7, 14, 21, and 28 days after seeding. The experiments were performed in temperature controlled greenhouses where the soil tem-

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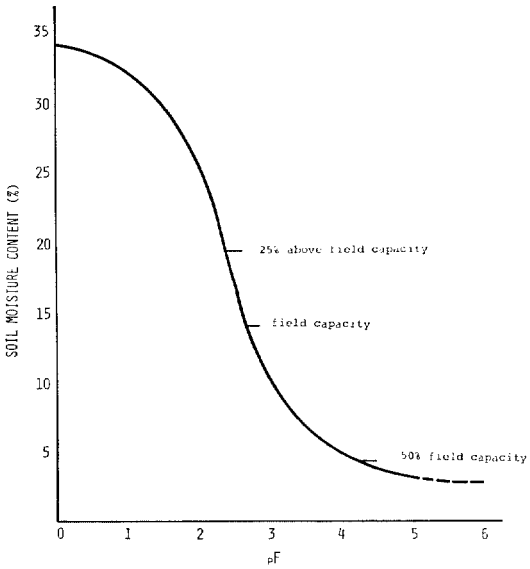

FIG. 1. Soil moisture curve for Hagener loamy fine sand.

perature was maintained at 25–29 C which, in preliminary tests, was more satisfactory for nematode activity than 16–21 C. A 16-hr photoperiod was used. The following data were taken at each sampling period: tap root length, number of secondary roots, number of bacterial nodules, whole-plant fresh weight, fresh and dry weight of roots, number of *P. alleni* in the roots, number of nematodes in the soil, and relative recovery of juveniles, males, and females of *P. alleni* from the roots by thin water-film incubation. Because only data concerning sex ratios and colonization were either meaningful or pertinent to this paper, only those data are presented. At each sampling the soil was saturated with water and the plants carefully removed from the pots and appropriate data recorded. The roots in each replication were mixed thoroughly and divided into two equal portions. One portion was used for *P. alleni* extraction and root dry weights; the other portion was used in colonization studies.

Internal root colonization by *P. alleni* was studied microscopically by two methods. In one method, the upper 5-cm of taproots from each pot were fixed in F.A.A., dehydrated in tertiary butyl alcohol, embedded in paraffin, sectioned longitudinally at 12 μ, and stained with safranin and fast-green (12). In the other, the entire taproot was stained for 30 sec in hot cotton blue in lactophenol, destained with hot lactophenol, and examined as whole or hand-sectioned mounts. The numbers of *P. alleni* and eggs per cm of root were counted in both methods. Where available, the 10 oldest fibrous roots were treated similarly. Data were analyzed by analysis of variance and the T-test.

Nematode extraction was essentially by the method of Christie and Perry (2). For extraction of nematodes from roots, roots were cut into 2-mm sections and placed in a petri dish with just enough water to cover the root pieces. Extractions were counted daily for 5 days with daily changes of water. The experiment was repeated by using a Hagener loamy sand from Fayette County, Iowa, with similar results not reported here.

RESULTS

COLONIZATION: The initial colonization by *P. alleni* in the top 5-cm of the soybean taproots peaked at 14 days (Table 1). The top 5-cm of the taproot was chosen for study because preliminary work indicated that 85–94% of the initial taproot invasion occurred in this area. There was little colonization of the initial secondary roots formed (Table 1). Nematodes that first colonized the roots did not remain in the top 5-cm, as evidenced by the decreasing numbers observed after 14 days (Table 1). Eggs were first seen in the roots 7 days after seeding and were hatching by 21 days.

There were significantly more nematodes in the top 5-cm of the taproot in the dry regime than in the wet regime (Table 1).

TABLE 1. Number of *Pratylenchus alleni* in the taproot and oldest secondary roots of soybeans growing in Hagener loamy fine sand for 28 days under wet and dry moisture regimes.[a]

	Top 5 cm tap root		Ten oldest secondary roots		Secondary root length	
Days after seeding	Wet No./cm	Dry No./cm	Wet No./cm	Dry No./cm	Wet cm	Dry cm
4	0.4	1.4	—[b]	—[b]	—[b]	—[b]
7	11.0	30.0	0.1	0.6	2.1	0.9
14	30.0	133.0	1.0	0.6	3.7	4.9
21	19.0	32.1	0.1	0.2	5.0	8.5
28	21.0	19.7	0.2	0.3	17.1	9.4
Av.	16.3	43.2**	0.3	0.4	7.0	5.9
	Eggs/cm in Top 5-cm of Tap Root					
4	0.0	0.0	—[b]	—[b]		
7	0.8	0.0	0.0	0.0		
14	21.0	54.0	0.1	0.0		
21	5.0	15.0	0.1	0.0		
28	6.0	2.0	0.2	0.1		
Av.	6.6	14.0*	0.1	0.02		

[a] Wet regime regulated from field capacity to 125% field capacity and dry regime regulated from field capacity to 50% field capacity.
[b] Secondary roots not yet formed.
* Significant at $P = 0.05$ level when compared with the wet treatment.
** Significant at $P = 0.01$ level when compared with the wet treatment.

Plant dry weight was significantly ($P = 0.01$) higher in the wet than in the dry regime. There also was a greater nematode density in the top 5-cm of taproot than in the fibrous roots (Table 1). *P. alleni* per cm of secondary roots decreased after 14 days, by which time the number and length of fibrous roots greatly increased.

SEX RATIOS: Numbers of males, females and adults extracted from roots were significantly ($P = 0.01$) greater in the dry than in the wet regime (Table 2). The ratio of males to females of *P. alleni* extracted from soybean roots was also significantly ($P = 0.01$) greater in the dry than in the wet soils, indicating that production of males may be favored or that females may be inhibited in drier conditions (Table 2). The number of adults to juveniles were about equal in the dry regime, but there were about twice as many juveniles as adults in the wet regime. The proportion of juveniles and females generally increased in the roots over the 28-day experiment.

TABLE 2. Males, females, and juveniles of *Pratylenchus alleni* extracted from soybean roots during 28 days under two moisture regimes[a] in Hagener loamy fine sand.

Moisture regime	Nematodes[b]			
	Males	Females	Adults	Juveniles
Wet	629	1,753	2,382	4,642
Dry	2,075**	2,524**	4,599**	4,362
	Ratios			
	Male: Female		Adults: Juveniles	
Wet	1.0:2.8		1.0:1.9	
Dry	1.0:1.2**		1.0:0.9**	

[a] Wet regime regulated from field capacity to 125% field capacity and dry regime regulated from field capacity to 50% field capacity.
[b] Per gram dry weight of roots.
** Significant at $P = 0.01$ level when compared with wet treatment.

DISCUSSION

Kable and Mai (6) found that invasion of alfalfa roots by *Pratylenchus penetrans* in a sandy soil was greatest under moist conditions (pF 1.85–2.5) and that it also occurred when the soil was saturated (pF 0.0) and under dry conditions (pF 3.0). In our study, more *P. alleni* occurred in soybean roots under dry (field capacity to temporary wilting) than under wet (field capacity or above) conditions. The 50% field capacity used in this study was at the temporary wilting stage. Field soils are not at constant pF values, and thus it is possible that the *P. alleni* that invaded soybean roots more actively in the drier than in the wet series did so at considerably lower localized pF in the moisture gradients around the roots. Other factors also, such as root attractants and aeration, however, may be affecting the observed behavior.

The results presented show that the pro-

portion of males to females was closer to a 1:1 ratio in the dry than in the wet regime, indicating that the dry regime favors either male production, inhibition of females or differential survival. In the wet series, the ratios deviated in favor of more females. Other factors have been associated with sex ratio changes. Many studies indicate that host nutrition probably is important in shifting sex ratios, usually with a shift towards males with conditions unfavorable for plant growth, or when less food, as a result of crowding of nematodes, is available per nematode (1, 3, 7, 8, 9, 13, 15, 16). One exception is *Anguina agropyronifloris*, in which females predominated under crowded conditions (10).

The results reported here agree with those obtained with other predominantly soil nematodes in that more males were produced either through crowding or conditions that adversely affected the growth of the plant, since plants in the dry regime had significantly less dry weight than did those in the wet regime.

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