Influence of Some Environmental Factors on Populations of Pratylenchus minyus in Wheat

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Abstract: The distribution and density of Pratylenchus minyus and possible relationships of several environmental components, including ammonium nitrate, were investigated in a wheat field in South Australia. Seasonal variation as measured every 2-4 weeks was eliminated from the observations by periodic regression. Correlation and regression analyses were then used to investigate the association of host plant, rainfall, temperature, and the fungus Gaeumannomyces graminis with P. minyus. Other than seasonal effects, soil moisture and G. graminis were the only components associated with P. minyus. Ammonium nitrate usually was correlated with fewer P. minyus in wheat roots. Much higher numbers of P. minyus were observed in seminal than in crown roots of wheat. Key Words: Gaeumannomyces graminis, seasonal variation, ammonium nitrate, crown roots, seminal roots, population dynamics.

Previous studies dealing with the influence of environmental factors on plant and soil nematodes under field conditions indicate that soil moisture is a major factor (16, 18, 19, 27). Inorganic fertilizers, soil type, soil pH, salinity, cation exchange capacity, oxygen, and the dominant vegetation are also important (9, 11, 12, 14, 16, 17, 18, 27, 28).

Many investigations dealing with the effects of environmental factors on nematodes have been conducted in the laboratory. These experiments helped to formulate hypotheses as to what may occur in the field. However, in the field, variables may vary independently or together to produce a particular result (15).

The first objective of this study was to identify factors responsible for variations in the number of *Pratylenchus minyus* Sher and Allen in a wheat field when no disease symptoms were present. Observations were also made on the influence of NH_4NO_3 on *P. minyus*, since there are reports that nitrogen compounds possess nematicidal properties (5, 21, 22, 23, 24). The numbers of *P. minyus* in crown and seminal roots were also noted.

MATERIALS AND METHODS

Environmental study: A 0.8-ha wheat field at Cooke Plains in South Australia was sampled on 33 occasions at 2-4 week intervals from 10 January 1970 to 14 January 1972. Plants were removed from soil to a depth of 30 cm. Eight sites (replicates) within the field were sampled at each sample date. Five plants were collected from each site, placed in a plastic bag, and taken directly to the laboratory. The root systems of five plants were utilized for immature plants, but only portions of root samples were used from mature plants. Between wheat crops, rotation crops such as timothy and alfalfa were sampled in a similar manner.

Tops of plants and washed roots were weighed. Root samples were stained in 0.15% cotton-blue lactophenol at 52 C for 12 h and examined at 50-100X under a stereomicroscope. *Pratylenchus minyus* and *Gaeumannomyces graminis* (Sacc.) Arx and Olivier var. *tritici* J. Walker [= Ophiobolus graminis (Sacc.) Sacc.] were the dominant nematode and fungal species infecting the wheat roots. The lengths of fungal hyphae were measured in stained roots in order to have data that could be analyzed statistically and obtained concomitantly with the nematode counts.

The following environmental factors were measured: (i) plant weight (PW), (ii) rainfall (RAIN), (iii) temperature range (TR), (iv) mean temperature (MT), and (v) lengths of fungal hyphae (FUN). A squareroot transformation was applied to the plant-weight data because a quadratic relationship existed between the plant weight and nematode data. Rainfall was recorded at a weather station 6 km from the field site. Daily rainfall values were converted to \log_{10} (x + 1), and the 30-day

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period prior to sampling was divided arbitrarily into 10-day intervals. The sum of the values for the first 10-day interval was multiplied by 1/6, the middle 10-day sum was multiplied by 1/3, and the 10-day sum prior to sampling was multiplied by 1/2, a calculation which gave more weight to rain occurring nearer to the sampling time. Temperature ranges and mean temperatures for each 30-day period were recorded at the weather station. Fungal hyphae measurements and nematode counts were converted to $\log_{10} (x + 1)$.

Influence of NH_4NO_3 on Pratylenchus minyus: A 0.45-ha plot adjacent to the environmental site was treated with NH_4NO_3 at 0, and 44.8 kg of N/ha applied in bands at planting time in early July 1971. There were six replicates in a randomized design. Wheat plants were collected every 2 weeks from September 10 to December 1 and processed for nematodes in the same manner as described in the environmental study.

A modification of the vertical migration technique (25) was utilized to study the effect of NH₄NO₃ on nematode movement. Plastic tubes 0.5 cm in diam. and 2 cm long were covered with butter muslin at one end and filled to a depth of 1 cm with sterile sand (of particle size 150-250 μ m) which had been treated with NH₄NO₃ at 0 or 40 μ g N/gm. The covered ends were immersed in sterile water of pH 7 at 20 C in individual concave blocks arranged in a randomized design with five replicates. Approximately 100 P. minyus were pipetted onto the upper surface of the sand in each tube. The percentage of nematodes migrating through the columns was determined at 2, 4, 8, 12, and 24 h. At 24 h, the nematodes remaining in the tubes were counted.

Distribution of Pratylenchus minyus in crown and seminal roots of wheat: The number of P. minyus in the crown and seminal roots of wheat in a 0.25-ha plot adjacent to the environmental site were recorded at 2-week intervals from 10 September 1971 to 1 December 1971. The soil contained 90% sand, 1% silt, and 9% clay in the top 30 cm, and the average seeding depth of 5 cm facilitated separation of the two root systems. The sampling procedures and treatment of samples were

similar to those described for the environmental study.

RESULTS

Correlation and regression analyses were used to examine the influence of the environmental variables on the numbers of *P. minyus* (NEM). The following regression equation was calculated:

NEM = 3.505 + 0.124 PW - 0.072 TR[1]

However, all the variables were significantly influenced by a periodic or seasonal component fluctuating with time in a similar fashion. Therefore, a second-degree harmonic curve was used to adjust for the seasonal influence (20). The coefficients for the harmonic curve were estimated by least squares methods:

 $\begin{array}{rl} \text{NEM} &= 2.383 - 0.841 \, \sin \, (0.0172 \\ \text{TIME}) \, - \, 0.158 \, \cos \, (0.0172 \, \text{TIME}) \\ - \, 0.368 \, \sin \, (0.0344 \, \text{TIME}) - \, 0.007 \\ \cos \, (0.0344 \, \text{TIME}) \end{array}$

[2]

The estimated curve and the observed nematode data are shown in Fig. 1. That part of an observation not explained by the harmonic curve (i.e. the residual) was the adjusted data (superscript 1). The regression equation based on adjusted data was: $NEM^{1} = -0.162 FUN^{1} + 0.118 RAIN^{1}$ [3]

The residual mean square for equation [3] was 0.1297, and the standard errors were 0.093 for the FUN¹ coefficient and 0.051 for the RAIN¹ coefficient.

Equation [3] indicated that soil moisture, as determined by rainfall, influenced the population fluctuation of *P. minyus*. A negative relationship existed between the number of nematodes and *G. graminis*.

The NH_4NO_3 treatments suppressed the reproduction of *P. minyus* in wheat roots and their mobility in the soil tubes (Tables 1 and 2). Nematodes were much more concentrated in seminal roots and average densities were often 10 times greater in seminal roots than in crown roots (Table 3).

DISCUSSION

Plant weight was the major component prior to adjustment of the data, a factor which was expected since *P. minyus* is an



FIG. 1. Log numbers of nematodes (x) and expected values (—), as determined by a second degree harmonic (equation 2), plotted against time period of 10 January 1970 to 14 January 1972.

obligate plant-parasite. However, the plantweight data were closely associated with the harmonic curve (Fig. 1) and adjusting the data for seasonal effects also removed much of the influence by the plant. This procedure facilitated examination of other variables whose influence on *P. minyus* may not have been as apparent as that of the host plant. Approximately 72% of the total variance of the nematode population was accounted for by the periodic component, and it was assumed that the second-degree

TABLE 1. Effects of NH_4NO_3 application on numbers of *Pratylenchus minyus* in wheat roots.

Sample date 1971	Nitrogen treatmentsª (kg N/ha)			
	0	44.8		
Sept. 10	1,230 ^b	1,122 ^b		
Sept. 23	1,738	1,259		
Oct. 7	1,479	1,549		
Oct. 21	1,950	955*		
Nov. 4	2,138	1,175*		
Nov. 18	2,455	1,349		
Dec. 1	1,329	955		

Asterisk () indicates a significant difference (P = 0.05).

^bEach nematode value is based on the sum of five plants and is the geometric mean of six replications.

TABLE 2. Percentage of *Pratylenchus minyus* migrating through sandy soil treated with NH_4NO_3 .

Time (h)	NH ₄ NO ₃ treatments ^a (µg N/gm of soil)		
	0	40	
2	2.3	2.3	
4	31.5	19.0*	
8	57.1	46.5**	
12	66.3	58.3*	
24	73.2	76.2	

*Percentage data subjected to angular transformation; each value is the mean of five observations. Asterisks *, ** indicate a significant difference at P = .05 and .01, respectively.

harmonic curve adequately represented the seasonal effect. The regression equation, based on the adjusted data, indicated that rainfall and G. graminis influenced P. minyus, but only for about 15% of the variance. Also, 15-20% of the between-time variation in the NEM variable was not accounted for by the environmental variables used here.

Soil moisture, as determined by rainfall, significantly influenced populations of P. minyus. The numbers of nematodes were highest at approximately days 310 and 690

Sample date	Fresh weight in gm ^a		Number of nematodes ^b		Nematode density ^e	
	CR	SR	CR	SR	CR	SR
Sept. 10	5.3	1.5	252	867	57	622
Sept. 23	8.7	1.6	469	948	64	657
Oct. 7	9.3	0.9	591	705	78	981
Oct. 21	8.5	1.6	380	891	48	619
Nov. 4	5.2	0.9	576	851	120	1,254
Nov. 18	6.0	0.4	944	624	194	1,503
Dec. 1	4.1	0.2	938	146	242	815

TABLE 3. The number and concentration of *Pratylenchus minyus* in crown roots (CR) and seminal roots (SR) of wheat at Cooke Plains.

"Each value is the arithmetic mean of 12 observations in sets of 5 plants.

^bNematode values are geometric means of 12 observations.

"Densities are derived from dividing nontransformed nematode values by appropriate root weight.

when soil moisture tension was about 2 bars, and lowest in mid-summer at days 70 and 430 when soil moisture tension was about 100 bars (Fig. 1). Kable and Mai (13) showed that under soil moisture conditions of 115 bars, reproduction, root invasion, and survival of *Pratylenchus penetrans* were greatly suppressed.

A negative relationship existed between P. minyus and G. graminis. Williams (26) observed that populations of the cereal cyst nematode, Heterodera avenae, fell when G. graminis was prevalent in spring wheat but increased when fungal infection was slight. (2) suggested that G. graminis Cooke suppressed reproduction, invasion, female development, and egg production of H. avenae in barley. The associations of P. minyus with fungi, however, have usually been positive. For example, the presence of P. minyus increased the severity of wilt in peppermint, and nematode reproduction increased when Verticillium dahliae was present (6, 7, 8). Benedict and Mountain (1) noted significant suppressions in yields of winter wheat as a result of root rots when P. minyus and Rhizoctonia solani were present.

The detrimental effect of NH_4NO_3 treatments on *P. minyus* (Table 1) agreed with results from other studies in which root-lesion nematodes decreased in numbers when NH_4NO_3 and other nitrogen compounds were applied (22, 23). Nitrogen is thought to act indirectly on nematodes by increasing the incidence of fungi which attack nematodes (3, 4). However, the application of NH_4NO_3 to the alkaline soil (pH 8.0) at Cooke Plains would increase the NH_3 content in the soil. Ammonia has nematicidal properties (5, 21, 23, 24) and even small amounts in soil could hinder nematode movement, as might have been the case in the soil tubes (Table 2), with subsequent inhibition of root invasion.

Nematodes were much more concentrated in seminal roots which developed before crown roots and were invaded first by *P. minyus* (Table 3). Williams (26) found many more *H. avenae* in seminal roots of spring wheat than in the remainder of the root system. Seminal roots have greater physiological activity than crown roots in cereals (10) and appear to be a preferred site for nematode invasion and reproduction.

The objective was to identify several key environmental factors influencing *P*. *minyus* in wheat. Although equation [3] was not tested with another set of data, the approach used herein should be useful in formulating predictive models for nematode pest-management and advisory programs.

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