Density-Dependent Parasitism of *Xiphinema diversicaudatum* by *Pasteuria penetrans* in a Naturally Infested Field

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ABSTRACT

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Parasitism of *Xiphinema diversicaudatum* by the nematode pathogen *Pasteuria penetrans* was studied in a naturally infested peach grove in Italy. The nematode density and the numbers of specimens infected or encumbered with spores were determined at monthly intervals during 2 yr in six replicated plots. Nematode densities varied between 32.4 ± 13.8 and 121.7 ± 26.5 specimens per 100 cm^3 of soil. The percentages of nematodes parasitized by *P. penetrans* were low, varying between 0.7 ± 0.7 and 9.3 ± 1.6. Both parameters remained relatively constant in time, fluctuating at 2- to 3-mo delays with maximum 2.6- and 9.8-fold increases, respectively. Similar trends were observed for the percentage of nematodes with spores and the mean number of spores per nematode. Density-dependent trends for nematode abundance and rates of parasitism by *P. penetrans* were shown by Bulmer's and Pollard's tests. When the grove was sampled to determine the spatial distribution of *X. diversicaudatum* densities and parasitism, *X. diversicaudatum* were found in 87.5% of the samples. Parasitism by *P. penetrans* was observed in 80.3% of the samples containing *X. diversicaudatum*. A subset of 43 (76.8%) observations from the spatial sampling was described by the equation $0.026 \ln(y) - 0.0038y = 0.000043x - 0.0052 \ln(x) + 0.04$, where x is the number of nematodes per 100 cm^3 of soil and y is the percentage of parasitism by *P. penetrans*.

Additional keywords: biological control, Lotka-Volterra equation, modeling, population dynamics.

Pasteuria penetrans (Actinomycetes) and related *Pasteuria* spp. are obligate, host-specific pathogens of nematodes (12,17,21). In several trials, *P. penetrans* suppressed root-knot nematodes, even at low application rates (12,21). It is one of the major candidates for the biological control of several plant-parasitic nematodes of economic importance (12,17,21).

P. penetrans produces resting spores that are resistant to desiccation and high temperatures (21). The spores are released into the soil as parasitized nematodes decay, and they adhere to the cuticle of susceptible nematode hosts. Upon contact with a host, the spores produce a germ tube that penetrates the cuticle. Spherical vegetative microcolonies form, which partially or totally digest the host body content before sporulating (12,17,21). A laborious procedure of drying and grinding plant roots containing parasitized nematodes is currently used to collect large numbers of *P. penetrans* spores (21). Exploiting *P. penetrans* and/or other *Pasteuria* species as biological control agents requires suitable methods for in vitro mass cultivation (3,26). The lack of sufficient numbers of spores for soil inoculum is the main obstacle to the direct evaluation of the effect of *P. penetrans* on a nematode population in the field.

Little is known about the relationship between the population dynamics of host nematodes and *P. penetrans* in nature. Understanding the mechanisms regulating host and parasite abundance in natural ecosystems would provide useful information for managing indigenous populations or introducing laboratory-reared *P. penetrans* to reduce nematode populations.

The obligate nature of parasitism and host specificity of *P. penetrans* (12,17) suggest a possible density-dependent relationship between *P. penetrans* and the abundance of its nematode hosts. "Density dependence" is a general term used to describe a situation in which the growth of a population is restricted in nature by adverse factors whose influence increases at high population levels and decreases as the population declines (15). Density dependence can occur in time or in space. Temporal density dependence is detected through repeated long-term time samplings or laboratory microcosm experiments (10,11). The spatial analysis of host and antagonist abundance can identify a second type of relationship known as spatial density dependence, in which population densities fluctuate over space rather than time, reflecting the reproduction of the host and antagonist or their motility and aggregation potentials (10).

A number of studies have demonstrated the persistence of *P. penetrans,* but they did not provide sufficient information to explain the changes observed in parasitism levels. In a South African field study, parasitism of *Meloidogyne* spp. was related to nematode density, but *P. penetrans*-infested fields showed the highest nematode populations (20). In South Australia, *P. penetrans* was frequently found in old and medium-aged vineyards with low densities of root-knot nematodes, suggesting the occurrence of a natural biological control (22). Temporal sampling of a *P. penetrans*-infested population of an ectoparasitic nematode, *Helicotylenchus lobus,* in California did not show a direct effect of the parasite on the host density, although *P. penetrans* parasitized an average of 50% of the nematodes at all sampling times. In this system, an increase in host density was followed, after 2 mo, by an increase in parasitism (7). In Florida, a population of the ectoparasitic nematode *Belonolaimus longicaudatus* showed relatively uniform rates of parasitism by *P. penetrans* over time (8). Finally, observations of *Meloidogyne* spp. in *P. penetrans*-infested fields in Spain showed that rates of parasitism were correlated with temporal fluctuations in the nematode populations (24).

The aim of the present study was to verify the occurrence of density-dependent parasitism in a population of the virus vector *Xiphinema diversicaudatum* (Micoletzky) Thorne naturally associated with a specific *P. penetrans* pathotype. The parasite had spore measurements larger than those of the original *P. penetrans*

pathotype from root-knot nematodes assigned by Sayre and Starr to *P. penetrans* sensu stricto (17). It will be herein referred to as *P. penetrans,* indicating through this term a "*Pasteuria penetrans* member" different from the microorganism originally described.

The population dynamics of both host and parasite were studied by analyzing repeated time samplings and using a generic model in which parasitism and host density mutually affect each other by a nonlinear feedback mechanism (25). This study was based on the spatial changes in *X. diversicaudatum* density and parasitism. Data from a study of more than 2 yr are presented, together with additional light microscopy observations on the biology of parasitism. Statistical tests for density dependence and nonlinear modeling were performed on the time and spatial distribution data, respectively.

MATERIALS AND METHODS

Nematode population study and time sampling. A population of *X. diversicaudatum* naturally associated with *P. penetrans* was sampled monthly in a 10-yr-old peach grove at Borgo d'Ale (Vercelli) from January 1990 through December 1991. A total volume of 2.5 L of soil (sand, 85.0%; loam, 8.3%; clay, 6.7%; pH 7.2) was collected from the plant rhizosphere in two opposite sites 40 cm from the trunks of six trees. The two subsamples were collected at depths of 10–35 cm, mixed, and placed in polyethylene bags. The samples were stored at 4 C until extraction by Cobb's sieving and decanting technique with sieves of 1.5-mm and 110-μm pore diameter (19). Assays were always performed within 1 wk of soil collection. Nematodes were counted with a Hawksley counting chamber at 125×.

After nematodes were counted, approximately 30 (17–55) *X. diversicaudatum* juveniles and adults per replicate were killed by gentle heat in a water droplet on a glass slide covered by a coverslip and examined under a Leitz Orthoplan microscope at 312× and 500× for *P. penetrans* parasitism. Nematodes containing spores (Fig. 1A), thalli (Fig. 1B), and/or other intermediate *P. penetrans* vegetative stages within their bodies were considered parasitized. The numbers of males, females, and juveniles with spores adhering externally to the cuticle were also recorded for each sample.

Spatial sampling. Taylor's power law (23) and the monthly means and variances of density and parasitism from the 2 yr of sampling were used to determine the number of spatial samples needed to obtain a 15% SE (standard error) to mean ratio level for parasitism (14). Fifty-six samples were collected in seven replicated rows at three- to four-tree intervals (eight samples per row). Soil was collected on 15 September 1992 from a single sampling site per tree at depths of 10–35 cm in the rhizosphere 40 cm from the trunk base. The nematodes were extracted as described, killed in an equal volume of 5% cold formalin solution, and stored at 4 C until counted. The data collected included nematode densities, rates of parasitism by *P. penetrans,* number of specimens infected by other fungi, and nematode stage and sex.

Statistical procedures. Time sampling data were interpolated by a cubic spline method with continuous second derivatives (16). Tests for normality of distribution of the spatial and time samples included chi-square and Shapiro-Wilk's W (16). Bulmer's and Pollard's tests were used to detect density-dependent trends. Both tests were preferred to regression analysis because of the absence of pronounced trends in the data series (15) and to maximize the likelihood of rejecting the null hypothesis of density independence. Bulmer's test was performed with the reciprocal of Von Neumann's ratio: $R = V/U$, where $V = \Sigma(x_{i-1} - x_i)^2$, $U = \Sigma[x_i - \text{mean}(x)]^2$, $i = 1, \ldots, 23$, and x is the tested variable. Density dependence at a given P value is shown by the test when $R < R_L$, where $R_L = 0.25 + (n - 2)X_L$ ($n = 24$, and X_L represents tabulated percentage points at P) (4). Pollard's test was performed with Pearson's correlation coefficient, $r(x,d)$, between each set of variables and the corresponding monthly differences (x_i is the variable tested; $d_i = x_{x-i} - x_i$; and $i = 1, \ldots, 23$). The test consists of a series of random permutations of the original monthly differences (d_i). A simulated series of variables is then calculated starting from x_1, which is added to the first permuted difference to obtain the second (simulated) value of the density in the series. Subsequent simulated density values are obtained sequentially by adding the permuted differences to each preceding value. The random permutations of the differences left unchanged the first and last variable values, thus allowing the calculation of a corresponding simulated $r_s(d,x)$. This procedure is replicated several times by comparing the simulated correlation coefficients with the observed $r(d,x)$ in order to verify whether the time changes depend on the population size or are density independent. The test evaluates the probability that in a population, the observed x_i values, and the corresponding differences, arise from a significant particular arrangement with respect to all the possible arrangements of x_i. In the hypothesis of random changes in the population from the initial to the last values, no significant differences should be observed in the distribution of the simulated $r_s(d,x)$; values greater than the real correlation coefficient are expected at a significant frequency. In a density-dependent population, only a low frequency of simulated $r_s(d,x)$ equal to or greater than the real correlation coefficient should result. The simulations were performed with a random number generator and an 80386 Intel microprocessor personal computer. The test shows the likelihood of density-dependent changes, significant at $P \leq 0.05$, when less then 5% of the simulated $r_s(d,x)$ values exceed the real $r(d,x)$ calculated from the original data series (15).

Modeling. The nematode density and parasitism data from the spatial distribution sampling were used to describe parasitism by a Lotka-Volterra model (25). The equation

$$a\ln(y) - by = cx - d\ln(x) + k, \quad (1)$$

where x is the number of nematodes per 100 cm^3 of soil, y is the percentage of parasitized nematodes, and a, b, c, d, and k

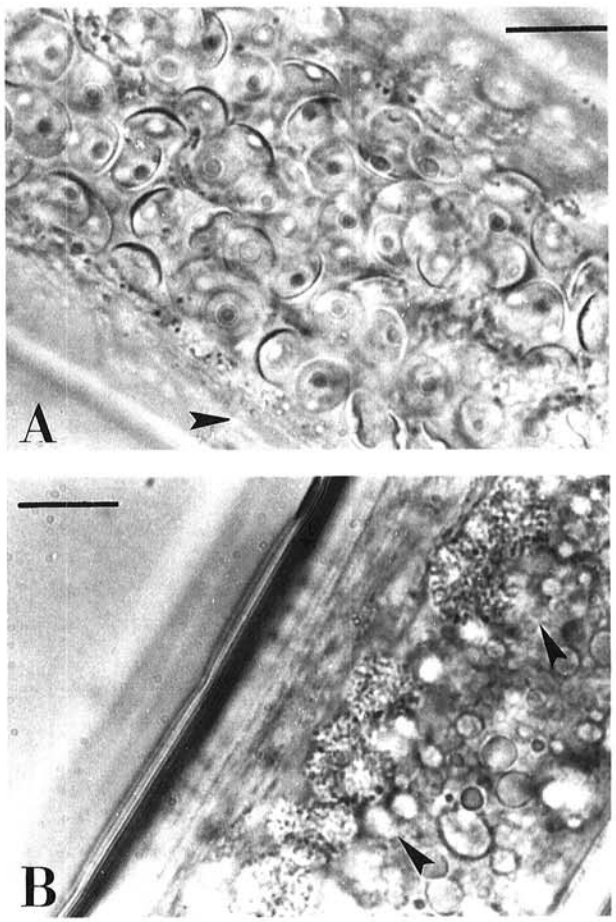

Fig. 1. A, Spores and **B,** dichotomic vegetative thalli of *Pasteuria penetrans* within parasitized *Xiphinema diversicaudatum* (bars = 10 μm).

are constants, was used to describe the densities and parasitism values observed in the field. Its application was based on the consideration that if similar density and parasitism time changes occurred independently all over the field, it would be possible to reconstruct the cycle on the density and parasitism phase space by examining a sufficient number of observations. The computed values of x and y were obtained through 800 iterative solutions of the form of equation 1 given by the system

$$x_{t+1} = x_t + ax_t - bx_t y_t \quad (2)$$

$$y_{t+1} = y_t = cx_t y_t - dy_t, \quad (3)$$

where $t = 1, \ldots, 800$ (1). The general biological meaning of each constant is as follows: a accounts for the host growth rate in the absence of other limiting factors; b accounts for the host decrease caused by parasitism; c accounts for the parasite growth rate as influenced by the host density; and d accounts for the parasite death rate, whereas k is an integration constant (2,6). Although no antagonist of *P. penetrans* is actually known, constant d also includes all the factors reducing spore density in soil, e.g., removal by repeated infections or molting, spatial dispersal, resistance within infected nematodes (5), and natural mortality.

Equation 1 has no explicit solutions, and its points must be iteratively calculated from initial values for density and parasitism. When plotted against time, equation 1 produces a series of regularly displaced fluctuations of density and parasitism. When density and parasitism are plotted together, a satellite-like closed curve is formed. As a function of time, the points move counterclockwise along this orbit, describing a cycle. When, through the iterative constant assignment, a cyclic equation is obtained, different initial values of x and y can be used. Changing these initial values influences only the horizontal and vertical dimensions of the cycle orbit but not its shape. A property of equation 1 is that the ratios d/c and a/b correspond to two "equilibrium values" for x and y, respectively. At these particular values, no changes in x and y occur over time, and the orbit is restricted to a single (equilibrium) point (6).

The basic assumptions considered for the treatment of the observed spatial data with equation 1 were that 1) the percentage of parasitism was representative of the spore density in soil, 2) each observation represented an independent homogeneous microcosm, 3) the parasitism and nematode density relationship was cyclic, and 4) the samples represented different points at different moments of the cycle.

The data fitting was performed through iterative constant assignments, starting from the initial values of density and parasitism assigned in order to maximize the goodness of fit. To evaluate the model's adherence to the observed data, Student's t test and Pearson's coefficient of correlation were computed between the observed and the calculated values of parasitism. These values were obtained by linear interpolation at each observed density between the closest intervals of the discrete parasitism values calculated with equations 2 and 3. The iterations were sequentially repeated with an 80386 Intel microcomputer until a significant correlation was found between the observed and the calculated values of parasitism. After the constants were assigned, both equations were used to calculate the maxima and minima reached by the system with different initial densities and/or parasitism rates. Mathematical and statistical analyses were performed through dedicated software packages (13,16).

RESULTS

Time sampling. Adult and juvenile *X. diversicaudatum* were recovered from all the samples collected each month. Mean monthly densities ranged from 32.4 ± 13.8 (mean ± SE) to 121.7 ± 26.5 specimens per 100 cm^3 of soil. Nematode population densities peaked three times in 1990 at 2- to 3-mo intervals, declined gradually during the winter of 1990, and subsequently peaked twice at 5- to 6-mo intervals in the spring and fall of 1991 (Fig. 2). Nematode densities from all the monthly replicated observations ($n = 144$) had a log-normal distribution (mean = 78.3 ± 24.4; $W = 0.9718$; nine classes).

At each monthly sampling, *P. penetrans* adhered to and parasitized *X. diversicaudatum* juveniles, males, and females. When observed by light microscopy, nematodes showed spores, thalli, and different intermediate sporulating stages of *P. penetrans* within their bodies. Parasitized nematodes of all stages survived penetration and development of the parasite and, in a few cases, could even molt or reproduce before *P. penetrans* completed its life cycle. More than 90% of parasitized nematodes filled with spores had spores throughout their entire bodies and appeared incapable of feeding or migrating.

The average percentage of *X. diversicaudatum* parasitized by *P. penetrans* varied between 0.7 ± 0.7 and 9.3 ± 1.6. Rates of parasitism peaked during May and September 1990, declined gradually during the winter of 1990, and had multiple peaks from March through December 1991 (Fig. 2).

The percentage of nematodes with *P. penetrans* spores adhering to the cuticle ranged from zero to 22.6 ± 6.9 with peaks at 3- to 5-mo intervals. The mean number of adhering spores per nematode ranged from zero to 3.4 ± 1.8, showing a peak during May 1990, a gradual increase from August 1990 through May 1991, a marked decrease during June 1991, and two peaks during July and October 1991 (Fig. 3).

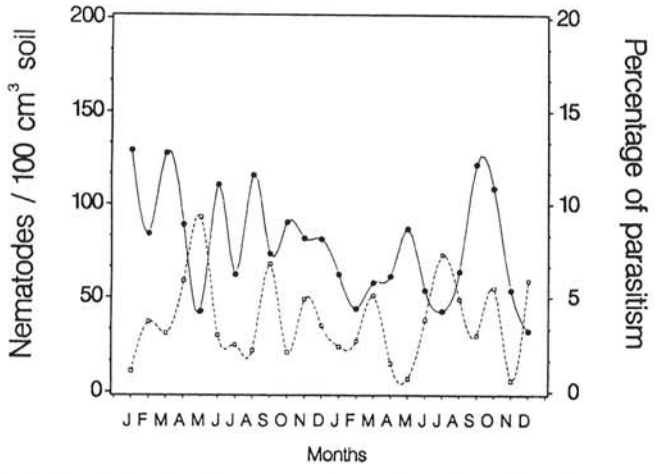

Fig. 2. Population densities (nematodes per 100 cm^3 of soil) of *Xiphinema diversicaudatum* (●) and percentages of *Pasteuria penetrans*-infected specimens (□) in a naturally infested field. Each observation is the mean of six replicates.

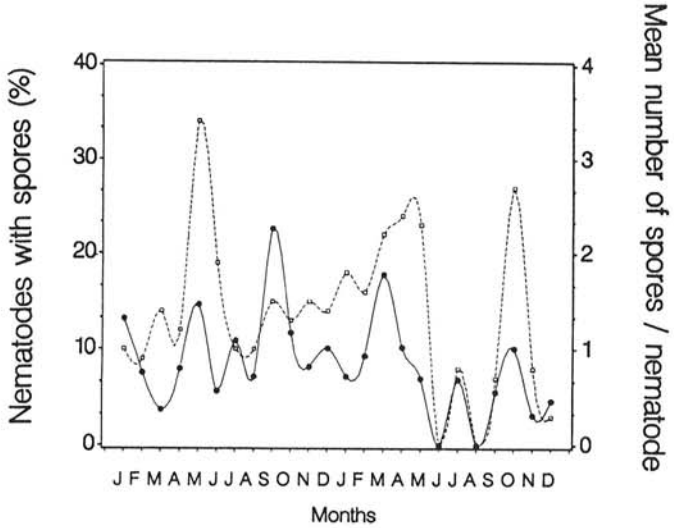

Fig. 3. Percentages of *Xiphinema diversicaudatum* with adhering *Pasteuria penetrans* spores (●) and mean numbers of spores counted per nematode (□). Each observation is the mean of six replicates.

TABLE 1. Pearson's correlation coefficients among the densities of *Xiphinema diversicaudatum* and *Pasteuria penetrans* parasitism variables[a]

	DEN	PAR	SPO	AVS	MAX	MAL	FEM
PAR	0.0700						
SPO	−0.0308	0.6728***					
AVS	−0.1139	0.3547**	0.5889***				
MAX	−0.1229	0.4451***	0.6662***	0.9735***			
MAL	0.1470	0.4590***	0.3561**	0.2587	0.2955*		
FEM	0.0772	0.4090**	0.2359	0.0298	0.0847	0.3330*	
JUV	0.5169***	0.2262	0.0947	0.0297	−0.0037	0.2052	0.3004*

[a]Samples were collected in September from 56 equally spaced peach trees. *, **, and *** = significance at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively. DEN = nematodes per 100 cm^3 of soil; PAR = percentage of parasitized nematodes; SPO = percentage of nematodes with adhering spores; AVS = mean number of spores per nematode; MAX = maximum number of spores per nematode with spores; MAL = percentage of males; FEM = percentage of females; and JUV = percentage of juveniles.

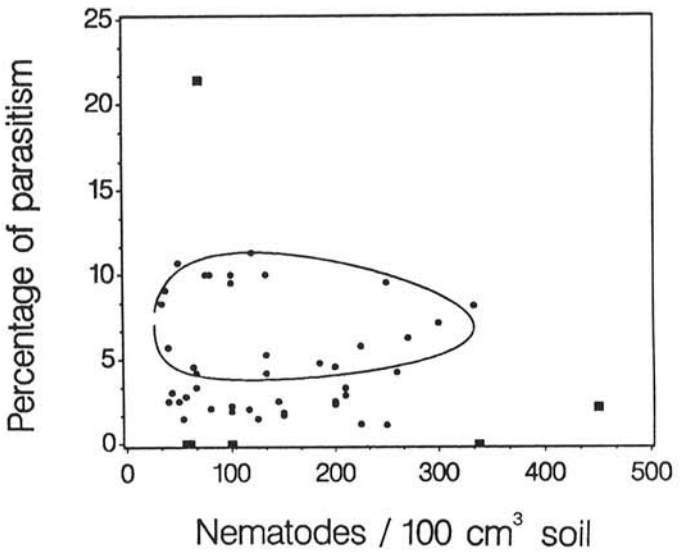

Fig. 4. Relationship between density of *Xiphinema diversicaudatum* and percentage of infection by *Pasteuria penetrans* in a naturally infested soil. Data are from a spatial sampling of 56 equally spaced trees. Each observation represents a 2.5-L soil sample. Calculated density and parasitism values from the equation $0.026 \ln(y) - 0.0038y = 0.000043x - 0.0052 \ln(x) + 0.04$ (where x is the number of nematodes per 100 cm^3 of soil and y is the percentage of parasitism; $x_1 = 27.2$, and $y_1 = 6.9$; continuous line, 800 points) are fitted to a subset of 43 observations (●). ■ = Observations with null parasitism values and two samples outside equation 1 density and parasitism maxima not considered for fitting.

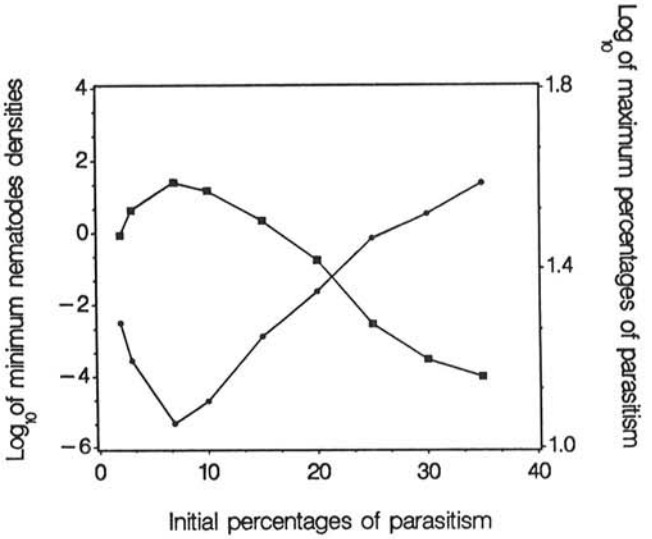

Fig. 5. Effect of increasing initial values of *Pasteuria penetrans* parasitism on the minimum densities achieved by *Xiphinema diversicaudatum* (■) and the maximum percentages of parasitism (●) obtained from the equation $0.026 \ln(y) - 0.0038y = 0.000043x - 0.0052 \ln(x) + 0.04$ (where x is the number of nematodes per 100 cm^3 of soil, y is the percentage of parasitism, and $x_1 = 27.2$ specimens per 100 cm^3 of soil for all points).

The 2-yr means of the monthly changes in host densities and parasitism ($x_{i=1}/x_i$, where x = the monthly mean of the tested variable) were 1.0 (0.5–2.6) and 1.8 (0.1–9.8), respectively. Relative to the previous month, nematode abundance and rates of parasitism increased in 9 and 11 mo, respectively, during the 2-yr study. The annual changes in density and parasitism were 0.48 and 2.1 (January 1991 and 1990) and 0.39 and 1.7 (December 1991 and 1990), respectively. No correlation was found between density and parasitism during the 2 yr of sampling. When each year was considered separately, the 1990 density and parasitism rates showed a significant inverse correlation after natural log transformation ($r = -0.6380$; $P < 0.05$).

Statistical analysis of *X. diversicaudatum* densities and percentages of parasitism by means of Bulmer's test showed density-dependent trends for the mean monthly densities ($P < 0.01$), the mean percentages of parasitized and spore-encumbered nematodes ($P < 0.001$), and the mean number of spores per nematode ($P < 0.01$). When the analysis was performed on each replicated plot, five of the six showed density-dependent trends for nematode abundance ($P < 0.01$), and all showed density-dependent trends for parasitism ($P < 0.001$). Pollard's tests performed with 3,700 simulations showed density-dependent trends for the mean values of densities ($P < 0.05$) and parasitism ($P < 0.001$). When the analysis was performed on each replicated plot, five showed density-dependence for host abundance ($P < 0.05$), and all showed density-dependent trends for parasitism ($P < 0.01$).

Spatial sampling. Adult and juvenile *X. diversicaudatum* were found in 87.5% of samples; mean density was 125.7 ± 13.4 (0–450) nematodes per 100 cm^3 of soil. Parasitism by *P. penetrans* was observed in 80.3% of samples; mean percentage was of 4.1 ± 0.5. Nematodes with adhering spores were found in 37.5% of samples; mean percentage was 1.2 ± 0.3. The average number of spores per nematode was 2.4 ± 1.3. The percentages of males and females within the population were 10.4 ± 1.0 and 9.8 ± 0.9, respectively. Nematodes with signs of mycoparasitism were found in 35.7% of total samples. The mean percentage of nematodes with zoospores and mycelium of *Lagenidium* sp. or other septate mycelia within the body was 1.3 ± 0.3. Parasitism by *P. penetrans* was correlated with the percentage of males and females in the nematode population but not with the percentage of juveniles (Table 1).

Modeling. Equation 1 was fitted to a subset of 43 (76.7%) observations from the spatial samples. Samples with null values for density or parasitism and two samples out of the model ranges were excluded. A significant fit was observed after 800 values of x and y were obtained through iterative calculations. The points were obtained starting from initial values of density and parasitism of $x = 27.2$ and $y = 6.9$; constants a, b, c, and d values were 0.026, 0.0038, 0.000043, and 0.0052, respectively (Fig. 4). The equilibrium values for density (d/c) and parasitism (a/b) were 120.9 and 6.84, respectively. The minimum and maximum calculated values for host density and parasitism were 25.5–333.5 nematodes per 100 cm^3 of soil and 3.7–12.7%, respectively. The calcu-

lated and the observed *X. diversicaudatum* percentages of parasitism were correlated ($r = 0.9078$; $P < 0.001$). No statistical difference was found between these two data sets when compared by Student's t test ($t = 1.3481$).

Changes in the simulated rates of nematode parasitism yielded an increase or decrease in the maximum density and parasitism values expected because of larger or smaller orbit amplitudes. Larger orbits tended to cause the cycle to approach parasitism and density axes. For a 3.5-fold simulated increase in parasitism, the system yielded a minimum nematode density of fewer than 0.001 specimens per 100 cm^3 of soil (Fig. 5).

DISCUSSION

The results from the time and spatial samplings support the hypothesis of a temporal, density-dependent relationship between *P. penetrans* and *X. diversicaudatum*. *P. penetrans* persisted in soil and was the main antagonist affecting *X. diversicaudatum,* but parasitism rates were low. The nematode population reached high density levels, fluctuating between 50 and 150 specimens per 100 cm^3 of soil, but never reached the growth rates observed for this species in greenhouse trials (9).

The statistical tests showed density-dependent trends in *P. penetrans* and *X. diversicaudatum* populations but did not allow any further evaluation of the extent to which parasitism influenced or was regulated by the host density. Nematode population growth is regulated in nature by initial densities, food availability, and host plant susceptibility (18). A number of laboratory tests in controlled conditions are required to discern which of these factors affect the nematode and parasite interaction.

In the time series, parasitism and nematode densities showed similar slopes at 3- to 4-mo delays with irregular fluctuations. Changes in the nematode population growth rates can be responsible for the lack of regularity in the density and parasitism changes observed over time. Seasonal changes in constant *a,* which accounts for the host population growth rate in equation 1, have no effect on the spatial distribution data, which result from samples collected at the same time.

The application of the Lotka-Volterra model to the dynamics of *X. diversicaudatum* and *P. penetrans* provides a basis for hypotheses on the mechanisms regulating host and parasite abundance in a natural ecosystem. This model appears to be more appropriate for describing the interaction of *P. penetrans* and *X. diversicaudatum* than models describing the parasite dynamics through a contact (transmission) term between healthy and parasitized hosts (1,11) because spores of *P. penetrans* are spread in soil and no direct transmission from parasitized to healthy nematodes occurs (12,17). A second model based on nematode and parasite propagule densities appears more suitable to describe the host and *P. penetrans* dynamics (11). The equations used considered basic biological parameters such as spore density and mortality, production and transmission rates, and host recruitment. The lack of methods for counting the numbers of *P. penetrans* spores in soil is the main obstacle to the application of this model (1). One such model was recently applied to the parasite *Hirsutella rhossiliensis* and its nematode hosts over short time intervals on an experimental laboratory scale (11). Unlike *H. rhossiliensis,* which can be cultured in vitro, *P. penetrans* spores cannot be produced in large quantities, and the number obtained from parasitized *X. diversicaudatum* is low. These circumstances impede the application of similar experimental procedures and limit the study of nematode and *P. penetrans* interactions to naturally infested fields.

The Lotka-Volterra model describes a theoretically stable situation difficult to observe in nature on a time scale because of the dimensions of the microcosm and other natural disturbances, which interfere with the host and parasite abundance and the regularity of their subsequent changes in time. The correlation between observed and calculated spatial sampling data indicates a cyclic, temporal density-dependent relationship between *P. penetrans* and *X. diversicaudatum*. There is a dynamic relationship between host and parasite in which parasitism is a function of host density and host density is a function of parasitism. The low levels of mycoparasitism observed suggest that the differences between expected and observed parasitism values were not related to factors such as concomitant antagonists and probably reflected host motility, root abundance or damage, or sampling error.

A property of equation 1 is that any changes in the density and parasitism values do not affect the system, which evolves toward cycles of different amplitudes (6). The different simulated parasitism rates used in computations with equation 1 showed that, after a suitable time period, parasitism can reduce host density and growth to values so low that nematodes can be considered locally extinct. On this basis, it can be proposed that stable and persistent cycles range between minimum and maximum threshold levels required to avoid the extinction of both organisms. This effect of equation 1 agrees with the properties of other models describing parasite-host interactions and minimum host and parasite density thresholds (1,11) and could explain the low levels of parasitism observed in *P. penetrans*-infested fields. High parasitism values would dramatically increase the likelihood of a host and parasite local extinction. This effect of the *Pasteuria*-nematode interaction agrees with observations of naturally infested fields, in which both host and parasite persisted in time at parasitism levels usually not higher than 50–60% (7,8, 20,22,24). When data from the spatial sampling are considered, it is worthwhile to note that in a limited number of observations (12.5%), nematodes were not found, whereas only two samples were far apart from the plot area defined by equation 1 maxima for density and parasitism (Fig. 4). This observation, and the model properties, suggest that the persistence of both host and parasite is the most common situation occurring in naturally infested fields, opposed to a temporary local extinction of the host.

The cyclic relationship described by equation 1 supports current efforts to exploit *P. penetrans* as a biological control agent for nematodes. The results of the computations performed with different initial parasitism levels suggest that introducing suitable amounts of *P. penetrans* spores could dramatically reduce the subsequent development of the host nematode population. Although this observation results from the modeling of a naturally infested field, it provides a basis for interpreting some successful trials of biological control achieved by inundative release of *P. penetrans* spores (21).

The reconstruction of a cyclic relationship through spatial samplings appears to be a suitable method for the study of other *Pasteuria* spp. and host nematode relationships, provided a previous evaluation of confidence bounds has been conducted (14). The standard errors observed were within the 15% bounds used to determine the number of samples to collect. These results agree with other field studies on the spatial distribution of nematodes (14) and confirm the efficacy of spatial samplings in the study of obligate nematode parasites (10).

Parasitism by *P. penetrans* allowed the hosts to survive infection, molt, and, in a few cases, even reproduce. Parasitism, however, was correlated with the fraction of adults in the population because of low reproductive rates associated with high levels of infection.

LITERATURE CITED

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