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The effect of prolonged diverse mineral fertilization on nematodes inhabiting the rhizosphere of spring barley

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Abstract

The studies embraced nematodes occurring in the rhizosphere of spring barley grown in experimental field divided into small plots fertilized for many years with the same set of mineral fertilizers: N, P, K, NPK, CaPK, CaNP, CaNK, CaNPK, and Ca or not fertilized at all (0). The study was carried out for three years. The first samples were taken in the 22nd year of fertilization. The fertilization did not differentiate greatly the structure of nematode communities. In all combinations of fertilizers plant and bacterial feeding nematodes dominated. Hyphal feeding nematodes occurred in all combinations but were less abundant than plant and bacterial feeding groups, omnivores and predators occurred only in some samples and their densities were low. Treatment was not observed to cause recurrent changes in total density and the density of plant and bacterial feeding nematodes. Such changes were noticed in the case of hyphal feeding nematodes. High density of them was connected with NPK application. The dominated genus of this group *Aphelenchoides* was significantly more abundant in NPK treatment than in the control (0). The application of Ca alone had a negative effect on *Pratylenchus*. This genus was significantly less abundant in Ca than in NPK, N, P or K treatments. The frequency of individual species of *Pratylenchus* was not affected by type of fertilization.

Keywords: Trophic groups, nematode community, *Aphelenchoides, Pratylenchus*, prolonged mineral fertilization.

Effets à long terme de la fertilisation avec divers engrais chimiques sur des nématodes de la rhizosphère de l'orge.

Résumé

L'influence de la fertilisation avec divers engrais sur la composition des communautés de nématodes et sur la densité des groupes trophiques a été étudiée dans des parcelles expérimentales. Chaque parcelle était fertilisée avec un des engrais suivants : N, P, K, NPK, CaPK, CaNP, CaNK, CaNPK et Ca. Une de ces parcelles n'était pas fertilisée (0). Les premiers échantillons de racines d'orge avec du sol ont été prélevés après 22 années d'expérimentation. Les prélèvements ont été effectués pendant trois années, en mai et juillet.

La fertilisation n'a pas différencié considérablement les communautés. Dans chaque cas de fertilisation, des phytophages et des bactériophages étaient présents. Les fongivores ont été également présents dans toutes les combinaisons mais à des densités plus faibles que celles des phytophages et bactériophages. Les prédateurs et les omnivores n'ont pas été observés dans toutes les combinaisons et leurs densités ont été plutôt faibles. La fertilisation n'a pas causé des changements récurrents de la densité des phytophages et des bactériophages. Dans le cas des fongivores, ce type de changement a été observé ; les densités élevées dans ce groupe sont associées à l'application de NPK. Le genre dominant dans ce groupe, *Aphelenchoides*, a été significativement plus abondant dans les parcelles avec NPK que dans celles sans engrais. Le chaulage sans application d'engrais chimiques a eu une influence défavorable sur l'abondance des nématodes appartenant au genre *Pratylenchus* ; ceux-ci étaient moins abondants dans les parcelles avec Ca que dans les parcelles avec N, P, K ou NPK. La fertilisation n'a pas affecté significativement la fréquence des espèces particulières de *Pratylenchus*.

Mots-clés : Communauté des nématodes, fertilisation avec engrais chimiques, groupes trophiques, *Aphelenchoides, Pratylenchus*.

INTRODUCTION

Mineral fertilization can affect nematodes in different ways. One of them is direct influence; according to some authors (Kimpiński *et al.*, 1976; Rodriguez-Kabana, 1986; Walker, 1971) fertilizers containing nitrogen in some conditions have a detrimental effect on nematodes. Indirect influence among others can be through plants or changes in soil pH. Mineral fertilizers are applied to increase the biomass of cultivated plants, therefore very often the growth of the root system is stimulated, providing more feeding sites for parasitic nematodes (Ross, 1959). Fertilization can also change the nutrient value of plants and in this way effect parasitic nematodes. Several studies indicate that the development of some parasitic nematodes depends on the nutrient status of plants (Bird, 1970; Davide & Triantaphyllou, 1967; Khan, 1985; Marks & Sayre, 1964; Moriarty, 1962). The presence of nutrients in the soil can result in better growth of bacteria and then in an increase of bacterial feeding nematodes (Griffiths *et al.*, 1992). The application of fertilizers can cause changes in soil pH; urea increases pH while ammonium nitrate decreases pH, lime which increases pH is added to neutralize the acidifying effect of some fertilizers (Huhta *et al.*, 1982). Changes in soil pH can influence some groups of nematodes. An increase in the abundance of parasitic nematodes in the acidified soil could be expected because it was proved for some parasitic species that they develop better in low pH (Brzeski & Dowe, 1969; Kuiper & Leeuw, 1963; Morgan & Mac Lean, 1968). An increase of hyphal feeding nematodes number in acid soil has been explained by the increase of fungi reacting positively to low pH (Bååth *et al.*, 1980; Wainwright, 1979).

In the literature there are many papers dealing with the effect of mineral fertilization on nematode communities but conclusions arrived at by different authors are not unanimous and even sometimes contradictory. Several studies have indicated that total number of nematodes increased after application of lime or urea (Bassus, 1967; Kozłowska & Domurat, 1975; Marshall, 1974; Hyvöven & Huhta, 1989). In contrast to this, Sohlenius & Wasilewska (1984) found a decrease of nematode abundance in the soil fertilized with nitrogen as De Goede & Dekker (1993) after liming. Kozłowska & Domurat (1977a, b) and Wasilewska (1976) did not notice great effect of NPK treatment on total number of nematodes. Many authors (Bååth *et al.*, 1978; Griffiths *et al.*, 1992; Hyvöven & Huhta, 1989; Sohlenius & Boström, 1986) observed an increase of bacteria feeding nematodes after nitrogen application. There is a divergence of opinion as regards the effect of nitrogen fertilization on hyphal feeding nematodes. Bassus (1960), Bååth *et al.* (1978), Kozłowska & Domurat (1977a), Sohlenius & Boström (1986), Sohlenius & Wasilewska (1984) showed that introduction of nitrogen into the soil caused a decrease of hyphal feeding nematodes, while Kozłlowska & Domurat (1977b) observed an increase of this group in the plots fertilized with NPK. Special emphasis was paid on plant parasitic nematodes because of their economic importance. Kozłowska & Domurat (1977a, b), Sohlenius & Boström (1986) showed an increase of parasitic nematodes after nitrogen application. In contrast to this, Wasilewska (1976) did not notice the effect of NPK on parasitic nematodes.

All these investigations concern the effect of short-term mineral fertilization on nematode communities. It is expected that a prolonged application of mineral fertilizers could more distinctly influence the nematode communities than short-term fertilization and thus allow to precise the behaviour of nematode species or trophic groups. A study on nematode communities in barley crops treated for over 20 years with different combinations of mineral fertilizers was carried out. The results of research on nematode communities inhabiting bulk soil were described by Kozłowska (1989), those dealing with communities in the rhizosphere are presented in this paper.

MATERIAL AND METHODS

Site

The study was conducted on the experimental field of the Warsaw Agricultural Academy in Obory, at the Łyczyn farm near Warsaw. The experimental field, with total of 14 ha, has been treated since 1960 with a set of mineral fertilizers N, P, K and Ca in a complete factorial block design. The treatments were divided in 4 blocks regarded as random replications. Across the strips fertilized with the same set of mineral fertilizers ran four stretches of crop fields on which potatoes, spring barley, winter rape and rye were cultivated in crop rotation system. This gave a combination of small plots, 120 m^2 each. Fertilizers were introduced into the soil in spring every year; nitrogen in form of ammonium nitrate, phosphorus as triple sulphosphate, potassium as 60% potassium salt. Calcium was introduced in the form of slacked lime every 4 year. The doses of fertilizers calculated per pure component, were as follows: N-120, P_2O_5-100 and K_2O-50 kg/ha, and Ca-2 200 kg/ha (Oset, 1982).

The soil of the experimental field was of lessive type, its upper layer being composed of light loamy sand overlaying light boulder loam (Oset, 1982).

Sampling

The plots selected for the study were treated with the following combination of fertilizers: O (control), N, P, K, NPK, CaPK, CaNP, CaNK, CaNPK and Ca. In the first year of the study no samples were taken from the field fertilized with potassium or phosphorus alone.

The material used in the study – the roots of spring barley, was collected twice during the vegetation season, in May and July. Each time 20 plants were removed from every plot. The samples were taken in 1982, 1983, 1984, respectively at the 22, 23 and 24 year of fertilization. The nematodes were extracted from the roots and the closely adhering soil – rhizosphere by the modified Baermann method (Flegg & Hooper, 1970). Ten grams of roots were cut into pieces of approx. 1 cm long and were placed on sieves. The nematodes were extracted for 7 days with a change of water every two days. The extracted nematodes were counted and mature specimens of *Pratylenchus* were determined to species, all other specimens to genus level. The nematodes were then divided into feeding groups according to the classification proposed by Yeates *et al.* (1993).

The pH of soil samples was determined in triplicates using mixtures of soil and distilled water (1:1).

Statistics

The effect of fertilization on the density of all nematodes in the communities was tested with the use of Friedman variance analysis (Siegel, 1956), (significance level $\alpha \leq 0.05$).

The effect of different fertilization on genera composition of nematodes was analyzed using correspondence analysis and canonical correspondence analysis (CANOCO, Ter Braak, 1988). The analyses were based on absolute nematode densities on which square-root transformation was made. Genera occurring in $\leq 20\%$ of the most common genus were down weighted in proportion to their frequency (Ter Braak, 1988).

The effect of fertilization on density of nematode trophic groups was analyzed by using multi-way ANOVA (computer program Systat).

The differences between the density of *Pratylenchus* as well as *Aphelenchoides* in the treatments were tested by Mann-Whitney U-test (significance level $\alpha \leq 0.05$). The same test was used for analyzing the differences in pH values between the control and the treatments with CaNPK, CaPK, CaNK, Ca and between the control and N, P, K, NPK.

The significance of differences between frequency of species belonging to the genus Pratylenchus was determined with the χ^2 test (Siegel, 1956).

RESULTS

pH of soil

The results of pH analysis in the soil treated with different combinations of fertilizers are illustrated in *figure* 1. In all treatments with Ca pH was significantly higher ($p \leq 0.05$) than in the control. On the contrary in the plots fertilized with N, P, K and NPK pH of the soil in these plots was significantly lower than in the control ($p \leq 0.05$).

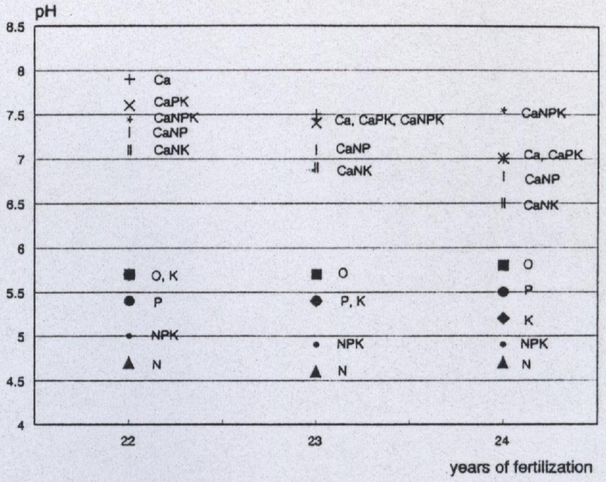

Figure 1. – Soil pH (H_2O) in the treatments (Means of triplicate assays).

Density of nematodes

Density of nematodes in the rhizosphere of barley was greatly variable (*table* 1 and *table* 2). In all combinations of fertilization very low numbers in the order of several specimens were accompanied by high numbers in the order of several hundred specimens per 1 g of roots. The differences between density of nematodes on variously fertilized plots were not significant ($p = 0.6$).

Community analyses

A canonical correspondence analysis with year (22, 23, 24 years of fertilization), season (spring, summer) and treatment (LIME – treatments with Ca but without N: CaPK, Ca; NITR – treatments with N but without Ca: N, NPK; LIMENITR – treatments with Ca and N: CaNPK, CaNP, CaNK; NONE – treatments without N and Ca: O, P, K) as environmental variables showed that 84% of the variation in genera composition was explained by year and season and only 16% was explained by the fertilization treatments. Soil pH was excluded from analyses because of its high correlation with the fertilizer treatments. To reveal the specific effect of the fertilizer type, the variables year and season were treated as co-variables. The first axis separated the NITR from the LIME and LIMENITR treatments (*fig.* 2). The second axis separated the NONE treatments from the treatments with either Ca and N present. The eigenvalue of the first axis was very small (0.034), but it significantly explained part of the remaining variation (Monte Carlo Permutation test, $p = 0.04$). The highest correlations were found with NITR ($r = 0.66$) and pH ($r = -0.64$).

The effect of prolonged diverse mineral fertilization

Table 1. – Effect of treatment on the density of nematode trophic groups (number per g of roots) in the rhizosphere of barley, 22 (A), 23 (B) and 24 (C) years after first fertilization. Data of spring samples.

Treatment	Plant feeding			Hyphal feeding			Bacterial feeding			Omnivores			Predators			Total density		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
O	30.1	12.6	53.4	1.1	2.3	14.5	6.1	12.3	60.9	0.8	0.6	0.0	0.0	0.3	0.0	38.1	28.1	128.8
N	431.0	41.5	36.1	19.5	3.4	0.5	16.5	14.6	5.1	0.0	0.0	0.0	0.0	0.0	0.0	46.7	59.5	41.7
P	–	19.1	62.2	–	1.8	2.3	–	30.7	11.3	–	0.3	0.0	–	0.0	0.0	–	51.9	75.8
K	–	11.0	1498.4	–	0.9	0.4	–	21.8	60.0	–	0.1	0.0	–	0.0	0.0	–	33.8	1558.8
Ca	10.5	9.2	26.9	3.7	2.2	7.8	10.0	22.2	62.8	0.0	0.2	0.3	0.0	0.0	0.0	24.2	33.8	97.8
NPK	191.2	17.3	151.9	27.8	10.2	24.7	6.1	19.0	100.0	0.0	1.3	0.0	0.0	0.0	0.0	225.1	47.8	276.6
CaPK	34.6	7.8	479.5	3.4	0.6	7.5	11.2	11.1	153.5	0.3	0.3	0.1	0.0	0.0	0.0	49.5	19.8	640.6
CaNK	3.0	15.0	83.9	2.6	0.5	6.9	9.7	13.2	56.0	0.0	0.0	5.3	0.0	0.0	0.0	15.3	28.7	152.1
CaNP	140.6	18.1	9.2	2.6	0.9	1.4	8.9	28.2	6.7	0.6	0.3	0.2	0.0	0.0	0.0	152.7	47.5	17.5
CaNPK	49.2	2.4	33.8	4.7	0.4	6.9	5.0	9.0	41.3	0.3	0.4	0.0	0.0	0.2	0.5	59.2	12.4	82.5

"–" not determined.

Table 2. – Effect of treatment on the density of nematode trophic groups (number per g of roots) in the rhizosphere of barley, 22 (A), 23 (B) and 24 (C) years after first fertilization. Data of summer samples.

Treatment	Plant feeding			Hyphal feeding			Bacterial feeding			Omnivores			Predators			Total density		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
O	603.3	26.0	98.2	9.2	8.4	25.7	54.8	30.6	102.5	6.7	0.0	3.2	0.0	0.0	0.0	674.0	65.0	229.6
N	18.0	55.8	394.0	10.3	58.7	41.8	6.1	150.9	58.1	0.3	1.8	0.0	0.0	0.0	0.0	34.7	267.2	493.9
P	–	54.5	724.2	–	17.0	6.2	–	74.5	68.7	–	0.0	0.0	–	0.0	0.0	–	146.0	799.1
K	–	4.0	205.6	–	8.3	0.0	–	17.7	22.5	–	0.0	0.0	–	0.0	0.0	–	30.0	228.1
Ca	57.6	3.8	42.5	44.4	4.4	11.0	92.2	10.3	164.5	7.9	0.4	0.2	0.0	0.0	0.0	202.1	18.9	218.2
NPK	157.8	34.8	840.4	10.5	74.1	31.6	7.1	113.1	484.3	0.0	0.0	0.0	0.0	0.0	0.0	175.4	222.0	1356.3
CaPK	100.9	36.8	87.1	24.7	28.3	6.0	22.4	42.6	22.0	0.6	0.0	0.0	0.0	0.0	0.0	148.6	107.7	115.1
CaNK	68.8	15.2	41.7	7.6	8.4	0.3	23.6	44.9	81.0	0.4	0.5	0.0	0.0	0.0	0.0	100.4	69.0	123.0
CaNP	344.2	19.4	73.5	14.0	20.0	14.7	37.7	73.4	83.2	0.0	0.0	0.0	0.0	0.0	0.0	171.4	112.8	171.4
CaNPK	20.5	28.3	524.0	12.0	20.4	15.0	20.8	56.2	530.0	0.4	0.3	0.0	0.0	0.3	0.0	53.7	105.5	1069.0

"–" not determined.

bacterial feeding nematodes were more abundant in the third year compared to the second year (Tukey test, $p \leq 0.05$).

Hyphal feeding nematodes in contrast to the two groups described above reacted to the treatment. Hyphal feeding nematodes were significantly more abundant in NPK than in CaNK and K treatment (Tukey test, $p \leq 0.05$). The density of nematodes belonging to this group also depended significantly on the season. They were more abundant in summer than in spring (Tukey test $p \leq 0.05$).

The density of hyphal feeding nematodes as a whole group did not differ significantly in 0 and NPK treatment. In contrast to this the dominating genus in this group, *Aphelenchoides*, was significantly lower in 0 than in NPK treatment (Mann-Whitney U-test, $p \leq 0.05$).

Nematodes belonging to the genus *Pratylenchus*

The effect of fertilization on nematodes of the genus *Pratylenchus* should be analyzed in detail. First, because *Pratylenchus* dominated in the examined material (*table* 3 and *table* 4) and second, because species belonging to this genus are regarded as one of the main parasites of barley. The density of this genus varied widely with year and season however in some extent depended on the treatment. The density of *Pratylenchus* in the plots treated with sole lime was significantly lower than in N, NPK, P or K treatment (Mann-Withney U-test, $p \leq 0.05$).

The frequency of *Pratylenchus* species in the variously fertilized plots is presented in *table* 6. The most frequent *Pratylenchus* species in the studied material were: *Pratylenchus crenatus* Loof (1960) and *P. neglectus* Rensch (1924). Much less frequent were *P. fallax* Seinhorst (1968), *P. penetrans* Cobb (1917) and *P. pinguicaudatus* Corbett (1969). The species *P. flakkensis* Seinhorst (1968), *P. pratensis* de Man (1880), *P. pseudopratensis* Seinhorst (1968) and *P. thornei* Sher & Allen (1953) were observed sporadically. The frequency of the individual species on the variously fertilized plots was similar (*table* 6). Statistical analysis revealed the lack of significant differences in the frequency of the individual *Pratylenchus* species in the studied combination of

Table 3. – Effect of treatment on the density (number per g of roots) of the dominant nematode genera in the rhizosphere of barley, 22 (A), 23 (B) and 24 (C) years after first fertilization. Data of spring samples.

Genus		O	N	P	K	Ca	NPK	CaPK	CaNK	CaNP	CaNPK
Plant feeding											
	A	28.5	429	–	–	9.6	190.8	32.5	2	140.6	47.8
Pratylenchus	B	9.7	40.1	18.3	10.4	7.6	8.5	6.5	14.4	17.1	0.4
	C	45.3	35.9	14.8	1498.4	20	151.9	478.3	76.9	5.9	29.1
Hyphal feeding											
	A	0.7	11.5	–	–	2.5	22	0.3	1.3	0.3	2
Aphelenchoides	B	1.5	2	1.3	0.4	1.4	6.6	0	0.1	0	0.2
	C	8.8	0.4	0	0	1.1	12.3	0	0	0.3	3.2
	A	0.4	6	–	–	1.2	4.4	0.9	1	0	2.3
Aphelenchus	B	0.5	0.4	0.5	0.4	0.2	0.6	0	0.2	0.3	0
	C	2.2	0	1.5	0	3.6	12.3	3.6	6.6	0.7	1.5
Bacterial feeding											
	A	1.5	4.5	–	–	0	1.7	0.6	0.1	0.7	3.7
Acrobeloides	B	2	2.4	3.5	0.3	0.8	0	0.6	1.2	3	0.4
	C	0	0	0	0	0	0	0	0	0	0
	A	0.4	0.5	–	–	6.5	0	0	0	0	0
Cephalobus	B	3	7.9	24.9	14.8	16	13.2	6.3	9.5	18.9	5.8
	C	46.6	4.2	5.8	56.6	22.6	29.7	142.8	20	2.4	24.7
	A	0.4	1.5	–	–	0.3	0	3.1	1.7	1.3	0
Chiloplacus	B	0.3	0.1	1	1.1	0.6	1.3	1.6	0.3	2.3	2.3
	C	3.3	0	0	0	10.7	0	0	9.3	0.5	5.2
	A	3	4	–	–	1.6	1.3	4.1	3.3	3.3	1.3
Eucephalobus	B	2.3	0.1	0	0.9	1	0.6	0.3	0.8	0.5	0
	C	7.7	0.3	3.2	3.3	21.5	12.5	10.7	18.7	2.7	8
	A	0	2	–	–	1.6	1.2	2.8	4	1	0
Panagrolaimus	B	3.6	2.8	0.3	4.6	2	0	0	0	0	0.2
	C	2.2	0.6	1.5	0.1	4.4	15.5	0	0	0.9	0.5

"–" not determined.

fertilization (value of χ^2 obtained in χ^2 test had probability $0.05<p<0.1$).

DISCUSSION

This study could not detect a significant effect on genera composition of nematode communities occurring in rhizosphere of barley which had been fertilized with mineral fertilizers for over 20 years. However, some part of nematode communities variation was correlated with fertilization, mainly with the application of nitrogen. Canonical correspondence analysis revealed that some genera were associated with specific fertilizer treatment, but it should be mentioned that the most of these general, except *Aphelenchoides* were detected rather rarely therefore it is difficult to conclude that their occurrence was really due to the treatments.

The obtained data indicated that nematode communities inhabiting the rhizosphere of barley were dominated by plant feeding and bacterial feeding nematodes. Hyphal feeding nematodes occurred in lower abundance compared to these two groups. This is in agreement with the observation made by Griffiths *et al.* (1991, 1992) who also found that nematode communities of barley rhizosphere were composed mainly of plant and bacterial feeding nematodes. In our study in the bacterial feeding group dominated: *Acrobeloides*, *Cephalobus*, *Chiloplacus*, *Eucephalobus* and *Panagrolaimus*. The association of these genera with the roots of higher plants was noticed by others authors (Domurat, 1970; Griffiths *et al.*, 1991, 1992; Kozłowska & Domurat, 1975; Mianowska-Dmowska, 1980; Roguska-Wasilewska, 1961).

Rhizosphere is a dynamic system which is affected by the nutrient level of the surrounding soil (Merckx *et al.*, 1987). Therefore after introduction of mineral fertilizers some changes in the rhizosphere can be expected. In fact, Griffiths *et al.* (1992) observed that application of nitrogen caused an increase of bacterial activity in the rhizosphere of barley and in result an increase of bacterial feeding nematodes. The data obtained in this study do not confirm the results presented by Griffiths.

Table 4. – Effect of treatment on the density (number per g of roots) of the dominant nematode genera in the rhizosphere of barley, 22 (A), 23 (B) and 24 years after first fertilization. Data of summer samples.

Genus		O	N	P	K	Ca	NPK	CaPK	CaNK	CaNP	CaNPK
Plant feeding											
Pratylenchus	A	600	13.7	–	–	53.9	155.9	100.3	68.4	344.2	10.7
	B	23.8	38.6	33	3.1	3	25.2	24.3	9.6	12.4	18.8
	C	52.5	374.9	717.9	203.1	37.4	840.4	86.8	38	54	523.3
Hyphal feeding											
Aphelenchoides	A	0	6.9	–	–	18.1	6.6	4.7	3.2	2.7	3.3
	B	1.7	37.3	4	0.3	0.9	71.1	1	2.2	7.7	14.2
	C	17.5	21.8	6.2	0	2.1	31.6	5.2	0	1.5	0
Aphelenchus	A	3.3	0.9	–	–	13.7	0.6	4.7	0.8	4.3	2.1
	B	5	16.4	8	5.7	1.3	2.2	15.6	3.2	10	5.7
	C	0	0	0	0	0	0	0	0	4.4	15
Bacterial feeding											
Acrobeloides	A	0	1.2	–	–	10.4	1.3	6	2.8	1.3	4.8
	B	0	3.6	0	2	0.4	11.1	3.6	1.5	1.5	0
	C	0	0	0	0	0	0	0	0	0	0
Cephalobus	A	7.8	0	–	–	14.8	2.6	10	7.2	6.7	2.4
	B	12.8	72.8	39	10	5.6	30.8	19.8	19.5	8.7	23.1
	C	72.5	54.5	50	12.5	144.5	390.6	16.8	74.7	41.5	410
Chiloplacus	A	12.2	0.3	–	–	5.9	0.6	0	2.4	0	5.1
	B	5	21.8	12	0	2.2	17.7	0	10	12	4.8
	C	0	3.6	0	0	8.7	0	0	3.5	8.8	5
Eucephalobus	A	15.9	0.9	–	–	57.8	2	0	9.2	26	7.3
	B	6.1	36.4	20.5	5.7	0.4	35.5	7.1	8.5	20.2	6.3
	C	22.5	0	12.5	10	6.7	31.2	5.2	2.8	30	45
Panagrolaimus	A	11.1	3.4	–	–	0	0.6	4	0	2.7	0.9
	B	0	10.9	1	0	0	0	6.7	1.2	12	16.6
	C	0	0	6.2	0	4.6	62.5	0	0	2.9	70

"–" not determined.

Some studies have indicated that plant parasitic nematodes are affected by the nutrient status of plants (Bird, 1970; Davide & Triantaphyllou, 1967; Khan, 1985; Marks & Sayre, 1964; Moriarty, 1962). It was proved that several species – among them the species belonging to *Pratylenchus* – developed better in host plants receiving complete nutrition than in the plants which were in deficiency treatment. Our results do not confirm this opinion because there was no significant difference between the density of *Pratylenchus* in NPK treatment and in deficiency treatments (N, P or K). The results obtained in this study are rather in line with those which indicate that an application into the soil of calcium alone is unfavorable for the nematodes of the genus *Pratylenchus* (Kincaid *et al.*, 1970). We found that *Pratylenchus* developed worse in the rhizosphere of the plants treated with Ca alone than in plants receiving full nutrients NPK or at least one element: N, P or K.

Since in the available literature there is not much information dealing with the effect of different mineral fertilization on hyphal feeding nematodes it is difficult to interpret significantly higher density of this group in NPK treatment in comparison to K and CaNK treatment. High density of hyphal feeding nematodes in NPK treatment seems to be connected with the genus *Aphelenchoides* which dominated in this group. *Aphelenchoides* was positively correlated with nitrogen treatment and thus with low pH since pH of the soil decreased after application of nitrogen. This confirms the observation made by Kozłowska (1989), who analyzed the communities of nematodes in bulk soil in the field on which this study was conducted. She found more *Aphelenchoides* in N and NPK treatments in comparison to the control. Our results are also in line with those obtained by others authors who studied nematodes in acidified soil (Dmowska, 1993; Hyvöven & Persson, 1990; Ruess & Funke, 1992). They noticed positive reaction of *Aphelenchoides* on low pH. This also corresponds with the data of De Goede & Dekker (1993) who in turn observed lower density of *Aphelenchoides* in soil treated with lime which increased the pH to 3.9 than in untreated soil (pH 3.2). It is possible that the increase of *Aphelenchoides* in soil with low pH is due to higher number of fungi which are a food source of

Table 5. – Results of multi-way ANOVA for the effect of treatment, season and year on the density of plant (P), hyphal (H) and bacterial (B) feeding nematodes in the rhizosphere of barley. Letters (A, B, C, D, E, F and G) indicate separate analyses.

Source of variation	P		H		B	
	F	p	F	p	F	p
A.						
all treatments	1.177	.340	2.464	.028	0.919	.521
season	0.223	.640	14.159	.001	5.981	.020
year	5.890	.006	0.299	.743	5.965	.006
B.						
N, P, K, Ca, O	1.310	.314	1.370	.295	0.510	.729
season	0.020	.889	5.560	.034	4.860	.045
year	2.700	.102	0.000	.998	1.620	.234
C.						
CaNPK, NPK, CaPK, CaNK, CaNP	0.970	.448	4.070	.016	0.940	.465
season	1.450	.244	8.180	.010	5.050	.018
year	3.720	.044	0.630	.547	4.070	.059
D.						
Ca-treatments against non-Ca-treatments	0.626	.433	3.160	.081	0.032	.860
season	0.610	.439	13.760	.001	6.791	.012
year	5.150	.009	0.170	.848	5.356	.008
E.						
N, NPK against O	0.270	.612	3.286	.095	0.516	.486
season	1.859	.198	3.320	.093	1.985	.184
year	1.807	.206	0.919	.425	2.048	.172
F.						
Ca, CaPK against O	0.575	.463	0.110	.745	0.138	.714
season	1.070	.321	4.932	.046	1.524	.241
year	1.052	.372	0.594	.568	5.611	.019
G.						
N, NPK against Ca, CaPK	2.678	.119	4.266	.050	0.637	.435
season	0.190	.668	8.204	.010	2.762	.114
year	3.196	.065	0.313	.735	3.167	.066

Shady fields with $p < 0.05$ – significant influence of variables.

Table 6. – Effect of treatment on the frequency of *Pratylenchus* species in the rhizosphere of barley. Data of spring and summer samples for three years.

Species	Combination of fertilization									
	O n(6)	N (6)	P (4)	K (4)	Ca (6)	NPK (6)	CaPK (6)	CaNK (6)	CaNP (6)	CaNPK (6)
P. crenatus	6	5	3	3	5	5	4	4	6	5
P. fallax	1	1	2	1	3	4	3	1	2	2
P. flakkensis	1	1	0	2	2	0	1	0	1	0
P. neglectus	2	4	3	2	3	4	3	4	5	3
P. penetrans	4	1	1	2	4	2	4	4	3	2
P. pinguicaudatus	0	2	1	1	1	1	0	2	2	1
P. pratensis	0	1	1	2	0	1	0	0	0	0
P. pseudopratensis	0	0	1	1	0	0	1	0	0	2
P. thornei	1	0	0	0	0	0	1	1	2	1

$n = 6$ for samples which were taken twice a year for three years.
$n = 4$ for samples which were taken twice a year for two years.

this genus. It has been proved that acidity of the soil favors the development of some fungi (Bååth *et al.*, 1980; Kmitowa, 1979; Wainwright, 1979).

Based on the obtained data it cannot be univocally said which species of *Pratylenchus* are related to a specific combination of fertilization since insignificant differences in the frequency of the individual species on the variously fertilized plots could be determined.

CONCLUSIONS

This study did not reveal a clear effect of prolonged mineral fertilization on nematodes in rhizosphere of barley. Only nitrogen application caused some increase of hyphal feeding nematodes, especially *Aphelenchoides*. The density of *Pratylenchus* was higher when barley was fertilized with NPK, N, P or K then when only lime was introduced into the soil.

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Effects of starvation and mechanical manipulation of leaf litter on faecal pellet production and assimilation in some millipedes from southern Africa: Implications for feeding strategies

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Abstract

Laboratory experiments were performed to establish the effects of starvation and mechanical grinding of leaf litter on the faecal pellet production and assimilation rates for three species of spirostreptid millipede common in the riparian savannas of south-eastern Botswana.

When food was present individuals produced between 6 and 20 faecal pellets per day. Starved animals stopped pellet production within 48 hours and retained some food in the gut. Individuals fed whole leaves, medium ground or fine leaf litter produced faecal pellets at similar rates but in *Zinopohora* sp. the mass of individual pellets tended to increase with a decrease in food particle size. Assimilation rate of fine ground litter was independent of, and generally higher than the previous assimilation rate on medium ground litter.

The implications of starvation and intermittent pellet production on the evolution of digestive and foraging strategies in savanna millipedes and the effects of faecal pellet size on rates of decomposition are discussed.

Keywords: Diplopoda, feeding, faecal pellet production, assimilation, savannas.

Effects du jeûne et de manipulations mécaniques de la litière de feuilles sur la production de fèces et sur l'assimilation chez quelques espèces de myriapodes d'Afrique du Sud : implications dans les stratégies alimentaires.

Résumé

Des expériences de laboratoire ont été menées pour établir les effects du jeûne et d'un broyage mécanique de litière de feuilles sur la production de fèces et les taux d'assimilation chez trois espèces de myriapodes spirostrepsides communes dans les savanes riveraines du sud-est du Botswana.

En présence de nourriture, les individus produisent entre 6 et 20 pelotes fécales par jour. Des individus soumis au jeûne arrêtent la production de fèces pendant 48 heures et retiennent une partie de la nourriture dans leur tube digestif. Des individus nourris avec des feuilles entières, des morceaux de taille intermédiaire ou de la litière finement broyée produisent des fèces en quantités similaires, mais chez *Zinophora* sp., la masse de chaque pelote fécale tend à augmenter avec la diminution de la taille des particules alimentaires. Le taux d'assimilation était généralement plus élevé avec de la litière fine qu'avec des particules de taille moyenne.

Les implications du jeûne et de la production intermittente de fèces sur l'évolution des stratégies digestives et de recherche alimentaire des myriapodes de savane et les effets de la taille des pelotes fécales sur les taux de décomposition sont discutés.

Mots-clés : Diplopoda, nutrition, production de fèces, assimilation, savanes.