PERGAMON

Soil Biology & Biochemistry 33 (2001) 429–438

Soil Biology & Biochemistry

www.elsevier.com/locate/soilbio

Biogeophysical factors influencing soil respiration and mineral nitrogen content in an old field soil

M.C. Savin[a,*], J.H. Görres[a], D.A. Neher[b], J.A. Amador[a]

[a]*University of Rhode Island, Laboratory of Soil Ecology and Microbiology, Department of Natural Resources Science, 210B Woodward Hall, 9 East Alumni Avenue, Suite 5, Kingston, RI 02881, USA*

[b]*University of Toledo, Department of Biology, 2801 West Bancroft Street, Toledo, OH 43606, USA*

Received 31 December 1999; received in revised form 27 June 2000; accepted 20 July 2000

Abstract

Microbivorous grazers are thought to enhance nutrient mineralization. The predicted effect of microbivory on nutrient cycling depends on the pore habitat model used. We evaluated CO_2 evolution and mineral N content of an old field soil to test two alternative habitat hypotheses. The *exclusion hypothesis* predicts that nematodes are separated from their microbial food resources in water-filled pores when soils dry, resulting in slower rates of biogeochemical transformations. The *enclosure hypothesis* predicts that nematode densities increase relative to their forage in smaller, isolated water volumes when soils dry, accelerating rates of biogeochemical transformations. We investigated the effect of soil moisture on the relationship between microbial biomass, microbivorous and predaceous nematodes, soil respiration and mineral N concentrations in an old field five times during the course of a year.

We could evaluate the validity of the two habitat hypotheses for the entire field only in August 1997 because that was the only sampling date when maximum water-filled pore diameters were smaller than microbivorous nematode body diameters in all sampled field locations. The mean microbivorous and predaceous nematode abundances for the field in August were greater than 6300 kg^{-1} and 80,000 kg^{-1}, respectively. Accordingly, the *exclusion hypothesis* was rejected. Predaceous nematode abundance was markedly higher in August than at any other sampling date. The high abundance of predators present suggests that detrital resources were not limiting productivity and that predators and microbivores were in enclosures, allowing predators to efficiently access their prey. Spatial maps, in agreement with linear correlation analyses, suggest that under our driest sampling conditions, soil respiration and mineral N content were controlled by microbivory and predation. © 2001 Elsevier Science Ltd. All rights reserved.

Keywords: Nutrient mineralization; Carbon; Nitrogen; Matric potential; Microbivorous grazing; Free-living nematodes; Enclosure hypothesis

1. Introduction

Soil respiration is a common and simple measure of biological activity in soil. In particular, C mineralization has been thought of as a convenient surrogate for N dynamics because C mineralization is thought to be coupled to N mineralization via the C:N ratios of detrital resources and their consumers (Smith, 1994; Mary et al., 1996; Myrold, 1998). Mineralized C is released as CO_2, while mineralized N may be immobilized by plants and microbes, resulting in measures of N mineralization which reflect net but not gross N mineralization. If C and N mineralization were coupled, measurements of C mineralization could be used to estimate N mineralization. Temporal variation of CO_2 evolution can be predicted by models that consider soil moisture and temperature (Edwards, 1975; Wildung et al., 1975; Norman et al., 1992; Bowden et al., 1998; Wagai et al., 1998). However, spatial and temporal heterogeneity of the soil environment plus the influence of biotic factors (Burgess and Webster, 1980; Rao and Wagenet, 1985; Groffman and Tiedje, 1988; Robertson et al., 1988; Schimel et al., 1989; Cambardella et al., 1994; Görres et al., 1998) and the effect of root respiration make it difficult to measure or predict the variation of C mineralization at the field scale. In particular, microbial community structure and population dynamics are affected by microbivory and may thus affect mineralization (Coleman et al., 1977; Anderson et al., 1981; Ingham et al., 1985; Hunt et al., 1987; Verhoef and Brussaard, 1990; Wardle and Yeates, 1993; Griffiths, 1994).

In a related laboratory study in which plants were excluded from soil, C and N mineralization were uncoupled regardless of sampling date or matric potential (Savin et al., 2000). Uncoupling may occur because of changes in

* Corresponding author. Tel.: +1-401-874-2902; fax: +1-401-874-4561.
E-mail address: msavin@etal.uri.edu (M.C. Savin).

0038-0717/01/$ - see front matter © 2001 Elsevier Science Ltd. All rights reserved.
PII: S0038-0717(00)00182-6

temperature, microbial community composition, substrate quality, or as the result of grazing of microbial biomass. The grazing hypothesis predicts that microbivory enhances C and N mineralization by two mechanisms. C mineralization is amplified indirectly if grazers keep microbial biomass below the carrying capacity of the soil ecosystem, thereby maintaining density-dependent compensatory growth rates (Traunspurger et al., 1997). N mineralization is enhanced directly when grazers excrete nitrogen in excess of their needs and indirectly by increasing nitrogen cycling by the microbial community (Verhoef and Brussaard, 1990). The net effect of grazing on N mineralization is to increase rates by about 30% (Hunt et al., 1987; Verhoef and Brussaard, 1990). Since soil fauna do not contribute much to soil respiration directly (Sohlenius, 1980; Hassink et al., 1993), uncoupling may result from the dissimilar direct contributions of N and C mineralization by grazers.

Although microbivorous grazing may affect C and N mineralization rates, the effect of grazing on C and N mineralization is moderated by water-filled pore structure (Elliott et al., 1980; Hassink et al., 1993). Nematodes are aquatic animals and require water to migrate (Wallace, 1958). Local moisture variations may alter the ability of nematodes to access microorganisms and thus control their ability to affect C and N mineralization. To improve our understanding of the effect of food web interactions on C and N mineralization, the relationship between water-filled pore space and grazing needs to be examined.

There are two habitat hypotheses for aquatic grazers applicable in a drying soil. The biogeochemical consequences of these two hypotheses differ:

1. The *exclusion hypothesis* (Darbyshire, 1976; Elliott et al., 1980) suggests that microbivorous grazers become less abundant as moisture decreases. Microfauna are excluded from their food resources in water-filled pores when diameters become less than grazer body diameters (Elliott et al., 1980). This means that, as grazers are progressively excluded from pore spaces containing microbial food resources as soil dries, the contribution of microbivorous grazing to mineralization rates diminishes. Specifically, one would expect decreased C and N mineralization and a change in the ratio of mineralized C and N as soil dries. When all grazers have been excluded from accessing microorganisms, C and N mineralization are expected to be coupled via the C:N ratios of microorganisms and detrital resources.
2. The *enclosure hypothesis* (Görres et al., 1999) suggests that as moisture decreases, grazers may become enclosed, or isolated, in water-filled pores in close proximity with their food resources. In an enclosure, microbivorous grazing should increase with increased density of grazers. As a result, C mineralization is expected to increase. Gross N mineralization may also increase. However, because N cannot diffuse from the enclosure, it is subject to immobilization and net N mineralization may therefore be low. When enclosures have formed and detrital resources are not limiting microbial activity, C mineralization does not depend on matric potential or water content, but rather on the number of microbivores present in the enclosures.

Some studies have shown that bacterial, protozoan and nematode activity was limited in dry soil, but increased directly following water additions and the release of substrates (Schnürer et al., 1986). However, the *enclosure hypothesis* is supported by data that suggests that nematodes are present in soils even at low matric potentials (Griffiths et al., 1995; Görres et al., 1999; Savin et al., 2000) when they should have been excluded. In addition, laboratory experiments under constant moisture and temperature showed an increase in C mineralization per unit microbial biomass with increased microbivorous nematode abundance in soil incubated at −50 kPa (Savin et al., 2000). This relationship is consistent with the *enclosure*, but not the *exclusion hypothesis*.

The objective of the study was to evaluate field patterns of soil respiration and mineral N content in terms of the two alternative habitable pore space hypotheses. In the field, C inputs change over the course of a year. In addition, moisture and temperature are variable and equilibrium conditions may not be reached. Moisture dynamics in the field are influenced by plant activity which may affect fine-scale distribution of water. The pattern of de-watering in the field is likely to be different compared to laboratory studies where water is drained from soil on tension tables (Savin et al., 2000). To assess the validity of the two hypotheses in the field, we sampled an old field five times during the course of a year. We wanted to determine if the influence of grazers on nutrient dynamics could be detected despite the presence of vegetation and variation in meteorological conditions. We expected the abundance of microbivorous nematodes and water-filled pore space to be correlated positively with soil respiration and mineral N content if the *exclusion hypothesis* is correct. If the *enclosure hypothesis* is correct, we expected that nematode abundance would be independent of matric potential and that soil respiration, but not necessarily mineral N content, would be related to microbivorous nematode abundance in dry soil. In addition to microbivorous nematode abundance, we evaluated other factors which may influence soil respiration and mineral N content, such as predaceous nematode abundance and microbial biomass C and N. To assess the relationship between mineral N content, soil respiration and controlling variables, we employed linear models and visualized field distributions by mapping variables using kriging.

2. Materials and methods

2.1. Study area

A 20×40 m^2 plot was subdivided into fifty 4×4 m^2 plots

in an old field at the Peckham Farm Research Area of the University of Rhode Island in Kingston, RI. The soil is a Hinckley sandy loam (sandy-skeletal, mixed, mesic Typic Udorthent) and had not been cultivated for at least 9 years prior to this study. Dominant vegetation includes timothy grass (*Phleum pratense* L.), brome grass (*Bromus inermis* Leyss.), orchard grass (*Dactylis glomerata* L.), Kentucky blue grass (*Poa pratensis* L.), rose (*Rosa multiflora* Thunb. ex Murr.), cinquefoil (*Potentilla recta* L.), brambles (*Rubus* spp.) and golden rod (*Solidago* spp.).

Mean ± s.d. of soil O.M. as determined by loss on ignition at 550°C was 56.2 ± 12.3 mg g^{-1} ($n = 120$), ρ_B was 1.06 ± 0.12 g cm^{-3} ($n = 200$), and pH at a 1:10 soil:water ratio (wt:wt) was 5.0 ± 0.6 ($n = 93$). Soil C (22.4 ± 5.5 mg g^{-1}) and N (1.4 ± 0.4 mg g^{-1}) were measured using a Carlo Erba C and N analyzer (NA 1500 series 2, Milan, Italy), yielding a soil C:N ratio of 16.1 ± 2.5 ($n = 80$).

2.2. Soil respiration

In situ soil respiration was measured at coordinates generated randomly within 40 randomly chosen plots six times in May, July, August, September, October and November of 1997 and three times in March, April and May of 1998. The positions of all sample points were determined using polar topographic surveying. Soil temperature to a depth of 5 cm and air temperature readings were taken in the late morning (10:00–12:00) during all nine sampling periods.

Cylindrical chambers (7 cm dia., 7 cm high) fitted with a rubber septum for gas sampling were driven about 1 cm into the soil surface. Samples of the air in the chamber were removed after 20–30 min using 20 ml gas-tight syringes. CO_2 evolution was previously determined to be linear over this time interval (data not shown). At six randomly determined points, a chamber was inserted into the soil and a gas sample taken immediately to determine background CO_2 levels. The chambers at those locations were removed and reinserted for subsequent measurement of gas flux. Samples in the gas-tight syringes were brought back to the laboratory, injected at a slight positive pressure into previously evacuated, 20 ml glass headspace vials that were sealed with a rubber septum and an aluminum crimp collar, and analyzed immediately using gas chromatography as described in Görres et al. (1997).

2.3. Sample collection

Forty intact soil cores (5 cm diameter, 10 cm length) were removed from the same coordinates used for soil respiration on five of the sampling dates (May 13, August 12 and November 11, 1997, and March 2 and May 19, 1998). Roots at the bottom of the cores were clipped, and the core bottoms covered with aluminum foil. The cores were brought into the laboratory within 2 h of collection and stored in the dark at 4°C. Soil (140–200 g moist soil) was shipped on ice by overnight courier within 2 days of collection to the University of Toledo (Toledo, OH) for nematode abundance and trophic group distribution determinations.

2.4. Soil physical properties

Matric potential was predicted from volumetric moisture based on a soil moisture characteristic curve measured in the field. The soil moisture characteristic curve was obtained from matric potential and volumetric moisture data measured on 22 occasions at 13 points in the field. Matric potential was measured with Irrometer moisture blocks (Riverside, CA) and volumetric moisture measured by time domain reflectometry or TDR (Trase, Soil Moisture Equipment, Santa Barbara, CA). Maximum water-filled pore diameters were calculated from matric potential using the following form of the capillary rise equation, valid for mineral soils (Hillel, 1971):

$$d = 300/\psi_m \quad (1)$$

where d is pore size in μm and ψ_m is the matric potential in kPa.

2.5. Inorganic N determination

A 2 M KCl solution was added to 1 g moist soil at a 1:10 soil:extract (wt:vol) ratio to extract NH_4^+ and NO_3^- (Keeney and Nelson, 1982). The soil suspension was shaken for 1 h, filtered through a Whatman #42 filter and the filtrate analyzed colorimetrically for NH_4^+ and NO_3^- using an Alpkem Rapid Flow Analyzer (RFA-300, Alpkem Corp., Clackamas, OR).

2.6. Microbial biomass C and N

The fumigation–extraction method (Vance et al., 1987) was used to measure microbial biomass C and N. Unfumigated soil (20 g moist) was extracted with 0.5 M K_2SO_4 using a 1:2 soil:extract (wt:vol) ratio, shaken for 30 min on an oscillating shaker, and filtered through Whatman #42 filter paper. Soil (20 g moist) was fumigated for 24 h with ethanol-free chloroform and then extracted in the same manner as unfumigated soil. The filtrate was analyzed using a Total Organic Carbon Analyzer (Model TOC-5000A, Shimadzu Scientific Instruments, Inc. Columbia, MD). An aliquot of the filtered extract was then oxidized (Cabrera and Beare, 1993) for measurement of microbial biomass N. Microbial biomass C was calculated as the difference between C in fumigated and unfumigated soil expressed per gram dry soil and multiplied by a correction factor of 2.64 (Vance et al., 1987). Microbial biomass N was calculated as the difference between N in fumigated and unfumigated soil expressed per gram dry soil and divided by a correction factor of 0.54 (Brookes et al., 1985). November microbial biomass C values were not included because the results were suspiciously low relative to biomass N values.

Table 1
Geostatistical parameters for soil properties in an old field in August 1997 and May 1998

Month	Soil property	Model	Nugget (C_0)	Sill ($C_0 + C$)	Q[a]	Range (m)	r^2	Krig mean[b]
August 1997	Soil respiration (mg C^2 kg^{-2} d^{-2})	Linear	20	20	NA[c]	NA	NA	20
	C_{mic} (mg C^2 kg^{-2})	Spherical	1100	7300	0.85	6	0.56	150
	Mineral N (mg N^2 kg^{-2})	Spherical	0.01	9.8	1.0	12	0.76	4.0
	Microbivorous nematodes (kg^{-2})	Linear	5.0×10^7	5.0×10^7	NA	NA	NA	6700
	Predaceous nematodes (kg^{-2})	Linear	1.8×10^{10}	1.8×10^{10}	NA	NA	NA	88,000
	Maximum water-filled pore diameters (μm^2)	Linear	4.2	4.2	NA	NA	NA	6.2
May 1998	Soil respiration (mg C^2 kg^{-2} d^{-2})	Spherical	5.0	89	0.94	18	0.85	23
	C_{mic} (mg C^2 kg^{-2})	Spherical	1800	8900	0.80	25	0.82	108
	Mineral N (mg N^2 kg^{-2})	Linear	28	28	NA	NA	NA	7.8
	Microbivorous nematodes (kg^{-2})	Linear	8.2×10^7	8.2×10^7	NA	NA	NA	9600
	Predaceous nematodes (kg^{-2})	Linear	2.5×10^6	2.5×10^6	NA	NA	NA	1600
	Maximum water-filled pore diameters (μm^2)	Spherical	1.0	1100	1.0	16	0.91	46

[a] $Q = C/(C_0 + C)$.
[b] Units for krig means are the square root of those listed in the table.
[c] Not applicable.

2.7. Nematode abundance and classification

Prior to extraction, all soil samples were stored at field moisture levels and 15°C. Nematodes were extracted from soil (140–200 g moist soil) using Cobb's sifting and gravity method (Thorne, 1961; Ayoub, 1980) modified by triplicate passes through 840, 250, 150, 75 and 44 μm mesh sieves. The final pass through the sieves was followed by centrifugal-flotation (Caveness and Jensen, 1955) modified by using a 1:1 (vol:vol) sugar solution and centrifuging for 1 min (Neher and Campbell, 1994). Taxonomic families were assigned to a trophic group (plant parasites, bacterivores, fungivores, omnivores/predators) according to Yeates et al. (1993). Bacterivores and fungivores were combined for data analyses and reported as microbivorous nematodes. Voucher specimens were preserved in 10% formalin and 1% glycerin, and sealed with Parafilm® (Neher and Campbell, 1994; Neher et al., 1999).

2.8. Statistical analysis

A Pearson product moment correlation analysis was conducted to determine which soil properties correlated with soil respiration and inorganic N in each sampling month. A forward stepwise regression model was fit to the data for each sampling month to determine which properties were significant predictors of soil respiration and inorganic N ($P \leq 0.05$). Properties evaluated in the statistical tests included soil respiration, inorganic N, microbial biomass C and N, microbivorous and predaceous nematodes, maximum water-filled pore diameters, soil O.M. and bulk density (SigmaStat for Windows, SPSS Inc., Chicago, IL).

2.9. Geostatistical analysis

All sampling positions were determined using polar topographic surveying and the coordinates of the points appended with the corresponding physical and biological property values. Geostatistical analysis was conducted using GS + (version 3.1, Gamma Design Software, Plainwell, MI). Only isotropic semivariograms were considered and the spherical model was chosen unless there was no apparent spatial relationship, i.e. the nugget was equivalent to the sill. In those cases, a linear model approximating the sample variance was chosen. Semivariance parameters that were used for map interpolation are listed in Table 1. Usually, six or seven lag classes were used to calculate semivariance and average lag spacing for all sampling months was 3.4 m. Lag spacing was chosen so that the smallest interval class contained at least 20 data pairs. Lag class distance intervals were greater than twice the minimum lag spacing in all months. The number of pairs in each class varied between 21 and 137 with greater than 66 pairs in all lag distance classes greater than the smallest interval class.

Kriging is an unbiased (i.e. mean residual should equal zero), weighted linear (estimates are weighted linear combinations of available data with weights summing to one) interpolation method which minimizes total variance by relying on semivariogram functions (Isaaks and Srivastava, 1989). Maps were computed using block kriging at a block size of 2 m, using (x, y) intervals of about 0.5 m. Additionally, the number of nearest-neighbors considered was set to six and the search radius was 20 m. Coordinates for the maps are reported as (x, y) with negative values plotted on the x-axis because positive values resulted in a mirror image of the field plot. Maps of the field plot were constructed using four contour intervals for the z-coordinate.

3. Results

The temporal course of soil respiration followed soil

temperature, except when C mineralization was limited by moisture, indicated by a drop in matric potential in July 1997 (Fig. 1A and C). Soil mineral N content generally tracked soil respiration, except in November when soil respiration was low, but mineral N content was higher compared to previous sampling dates (Fig. 1C). Microbial biomass C followed a similar pattern to soil respiration and showed about a two-fold difference seasonally. Although microbial biomass N did not increase from August to November, as did mineral N content, the trends in N content were similar.

To evaluate the applicability of the *enclosure* and *exclusion* hypotheses to nutrient dynamics, matric potential had to be less than −20 kPa which is equivalent to water-filled pore openings of less than 15 μm. In August, matric potential for the entire field was below −40 kPa, representing water-filled pore diameters less than 7 μm. Previous to the August sampling date, matric potential had been as low as −150 kPa (Fig. 1A). Several precipitation events had brought the matric potential back up to an approximate field average of −50 kPa (Fig. 2). In May 1998, about 30% of the field was below the matric potential threshold of −20 kPa, corresponding to pore openings of less than 15 μm. The average matric potential was greater than −20 kPa for the remaining sampling dates. Mean microbivorous nematode abundances were 6300 kg^{-1} and 10,000 kg^{-1} in August 1997 and May 1998, respectively (Fig. 1B). We found at least 1000 nematodes per kg soil with at least 300 microbivores per kg in every sample location on both dates. In addition, we generally found greater than 5000 and greater than 100 predaceous nematodes per kg in all sample locations in August 1997 and May 1998, respectively.

If the *enclosure hypothesis* is correct, spatial patterns of respiration and possibly mineral N should coincide with microbivorous nematodes at low moisture. At the local scale in August, high mineral N content occurred with high soil respiration, high abundance of microbivores and low predator densities (Fig. 3). Low CO_2 evolution occurred in locations of high predator densities and low microbivore abundances (Fig. 3). At the field scale in August 1997, microbivore abundance was a significant predictor of soil respiration in a stepwise linear regression model (data not shown). Soil respiration was also correlated positively with microbial biomass C ($P < 0.03$), and microbivorous nematodes ($P < 0.09$). It was correlated negatively with predaceous nematodes ($P < 0.04$). Mineral N was not predicted by any biotic factors in the step-wise linear regression model (data not shown). However, it was correlated positively with microbivorous nematodes ($P < 0.07$) and C mineralization ($P < 0.05$), but not with predators ($P < 0.46$) or microbial biomass ($P < 0.92$).

Mean values of soil properties in May 1998 were similar to those in August 1997, except for matric potential and the ratio of microbivorous to predaceous nematodes (Fig. 1). Yet, in May 1998, neither stepwise regression nor

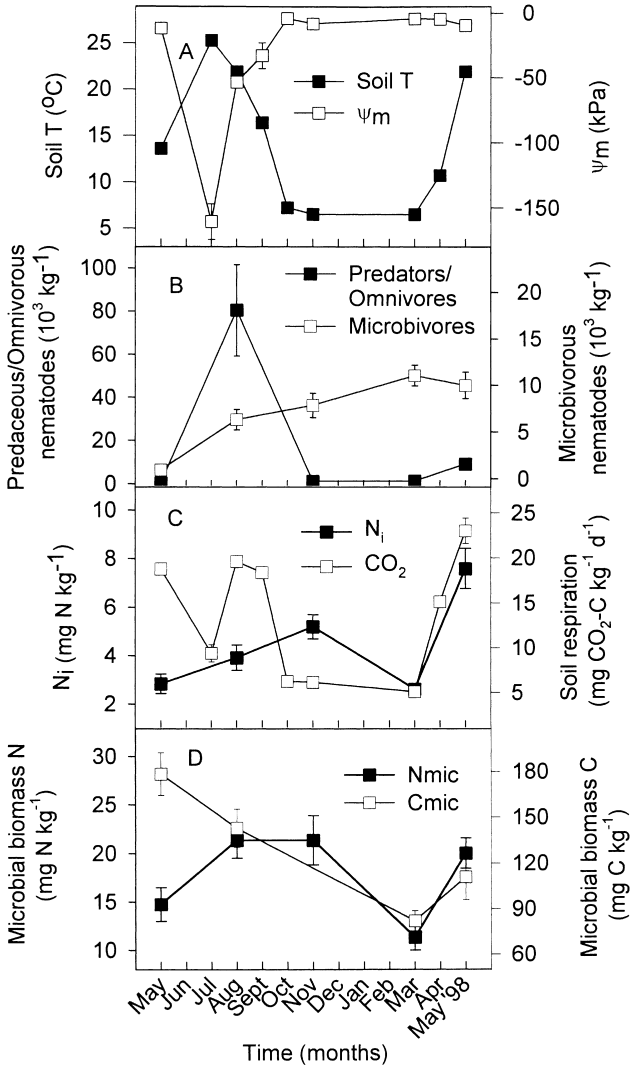

Fig. 1. (A) Matric potential and soil temperature; (B) microbivorous and predaceous/omnivorous nematodes; (C) mineral N (N_i) and soil respiration (CO_2); and (D) microbial biomass C and N in an old field as a function of time. Error bars represent one standard error and are smaller than the symbols where they are not seen.

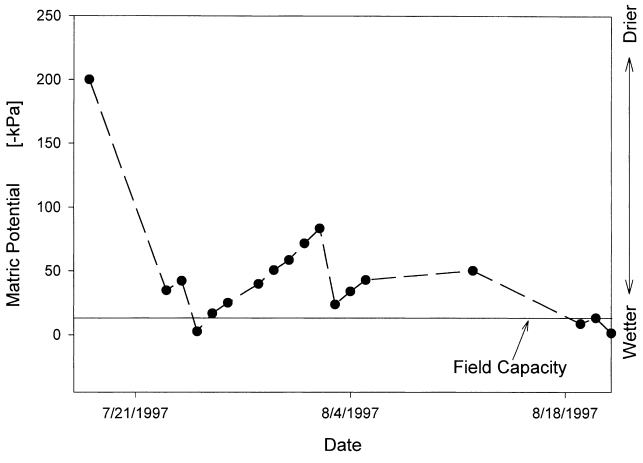

Fig. 2. Matric potential in an old field in July and August 1997.

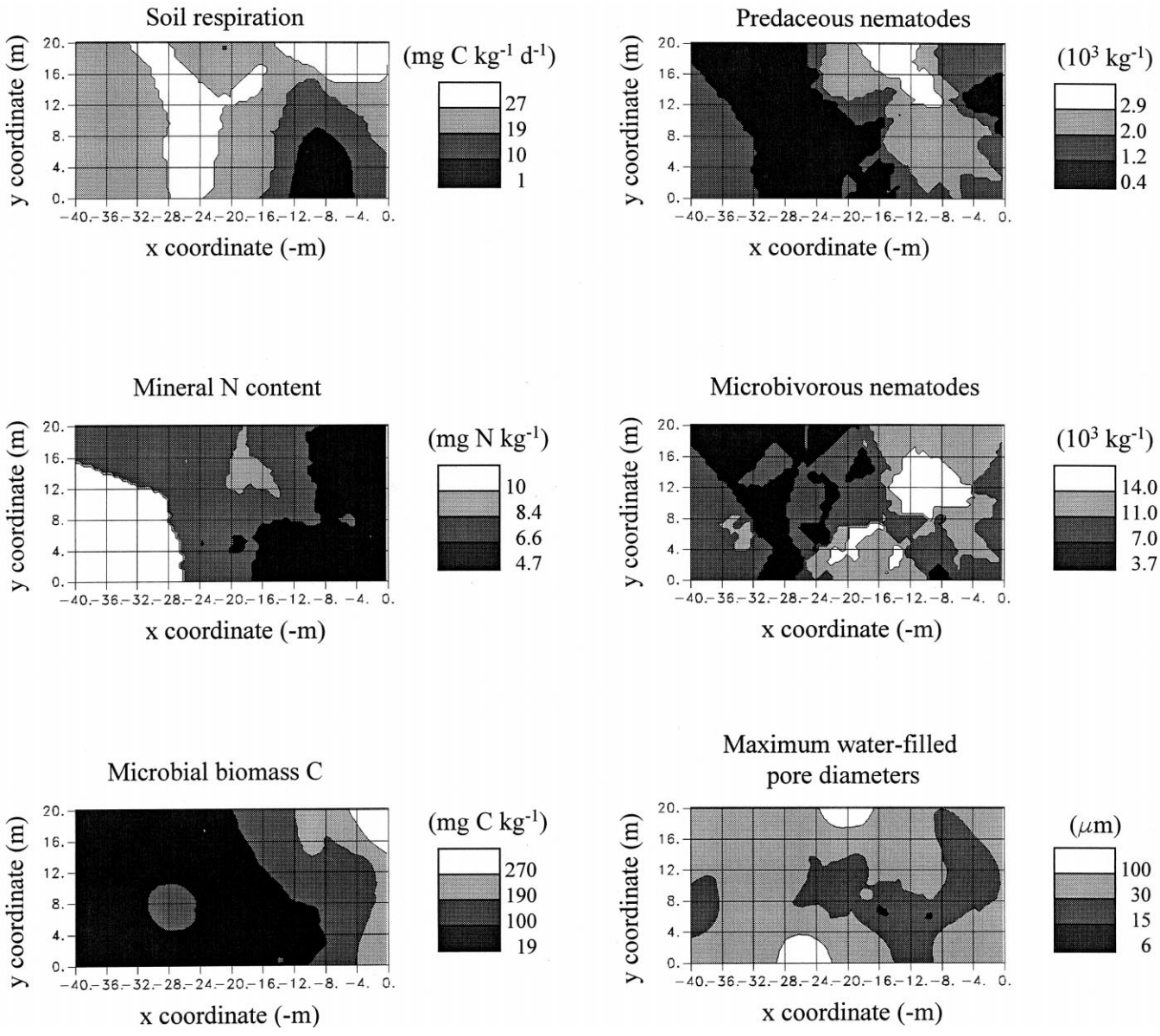

Fig. 3. Kriged maps of soil respiration, mineral N (N_i), microbial biomass C (C_{mic}), maximum water-filled pore diameters, and predaceous and microbivorous nematodes in an old field in August 1997.

correlation analysis identified any relationships between soil respiration or accumulated mineral N and biotic soil properties (data not shown). The lack of apparent trophic relationships in May is mirrored by the lack of coincidence in local patterns (Fig. 4).

4. Discussion

CO_2 evolution and moisture content were comparable to those in a previous study (Görres et al., 1998). Although temperature is consistently more influential than moisture, moisture fluctuations can explain additional variations in CO_2 evolution, especially at higher temperatures (e.g. Wildung et al., 1975; Bowden et al., 1998; Wagai et al., 1998). Generally, soil CO_2 evolution decreases at low water potential even when the soil temperature would otherwise support high C mineralization (Edwards, 1975; Norman et al., 1992). However, at one soil temperature there were still wide spatial variations in soil respiration rates in the field (Figs. 3 and 4). These localized differences may be the result of spatial variation in ecological mechanisms, including abiotic factors, controlling mineralization.

N mineralization tends to decrease with decreased temperature (Stanford et al., 1973; Addiscott, 1983; Grundmann et al., 1995; Sierra, 1997). The continued high mineral N content in November may be explained by different mechanisms. Low respiration (Fig. 1C) indicates that both microbial mineralization and plant root activity and uptake of N were low at the time of sampling.

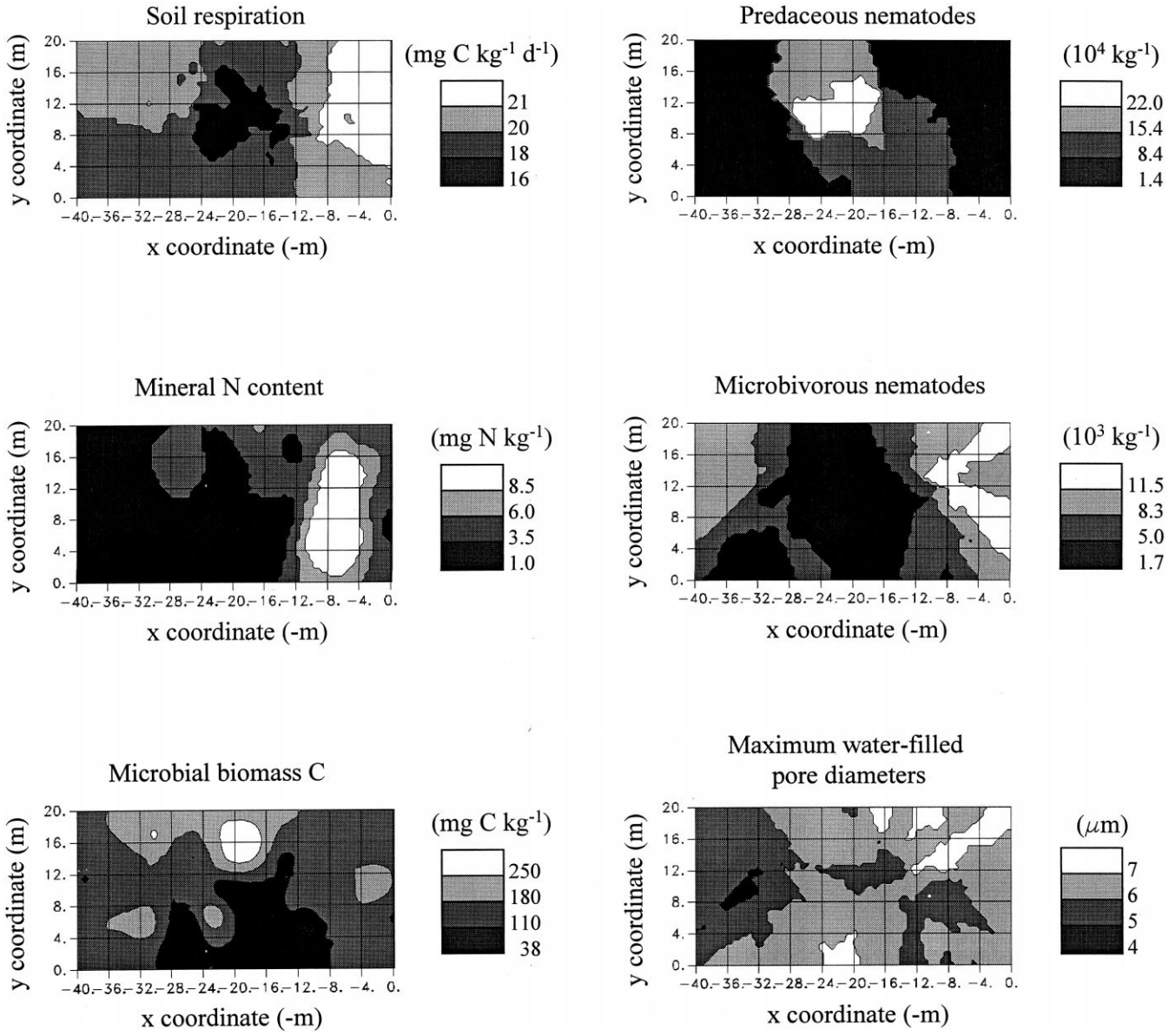

Fig. 4. Kriged maps of soil respiration, mineral N (N_i), microbial biomass C (C_{mic}), maximum water-filled pore diameters, and predaceous and microbivorous nematodes in old field in May 1998.

However, N mineralized from organic matter, including dead microbial biomass, previous to the sampling date may have contributed to the accumulation of mineral N in the absence of immobilizing organisms. Alternatively, microbivorous nematodes (Fig. 1B) could have released N from microbial biomass (Woods et al., 1982; Anderson et al., 1983).

To reject the *exclusion hypothesis*, microbivorous nematodes must exist in areas where water-filled pore spaces had capillary diameters less than 15 μm, i.e. where soil matric potential was less than −20 kPa. We base this size threshold on Jones et al. (1969) and on the size distribution for microbivorous nematodes in Görres et al. (1999), obtained for soil taken from the same field. In that study, 95% of microbivorous nematodes had diameters greater than 15 μm. The average matric potential was greater than −20 kPa in May and November 1997 and March 1998, so that evaluation of the *exclusion* or *enclosure hypotheses* was restricted to August 1997 and May 1998. Mean microbivorous nematode abundances for these two sampling dates (Fig. 1B) demonstrate that the *exclusion hypothesis* does not hold true for the field average. However, field distributions of nematodes (Robertson and Freckman, 1995; Ettema et al., 1998) and of soil moisture (Amador et al., 2000) are spatially variable. Therefore, it is possible that exclusion does occur in portions of the field with low matric potentials. In order to evaluate this possibility, we examined the spatial distribution of moisture and microbivorous nematode abundance.

Microbivorous and predaceous nematodes were present at all sampling points (Figs. 3 and 4). Exclusion, especially

of the generally larger predaceous nematodes, would have been expected for both dates. These results support the rejection of the *exclusion hypothesis* at both the local and the field scale. The *exclusion hypothesis* is thus of little value in explaining the possible role of microbivorous nematodes on C and N mineralization. Could the *enclosure hypothesis* be a better model for the effect of food web interactions on biogeochemistry at low matric potentials?

One of the consequences of grazing in enclosures is that grazers and their forage and predators are confined to and isolated in limited pore spaces. This should result in shorter separation distances between microorganisms, microbivorous and predaceous nematodes and, thus, in greater microbivorous grazing and nematode predation efficiencies. In August, when soil moisture was low everywhere in the field, predaceous nematodes were 12 times more abundant than microbivores (Fig. 1B), suggesting efficient predation. We observed one of the highest respiration rates, microbial biomass C and predaceous nematode abundance in August. These data could have been the result of greater interactions between trophic levels in hydrologically isolated aggregates, i.e. enclosures. Predators may have taken advantage of close distances between themselves and their prey when resource availability, possibly due to rhizodeposition, was not limiting lower levels of production. Wardle et al. (1995) suggested that high predator abundances can be explained by active lower food web levels. However, caution must be used in inferring causal relationships from correlative data. Factors not considered or not found to be significant in a correlation analysis may have caused a significant correlation between two otherwise unrelated properties. For example, increased temperature or resource availability may also have contributed positively to predator abundance in August.

We had expected a low mineral N content in August 1997 because of tight coupling of N mineralization and immobilization predicted in enclosures. Diffusion of mineralized N out of enclosures is limited, making N readily available for microbial growth inside the enclosure. In a laboratory study (Savin et al., 2000), there was no net N mineralization in undisturbed soil cores during a 1 month incubation period at constant matric potential (−50 kPa) and temperature (22°C), representing environmental field conditions in August 1997. C resources in the field with a low C:N ratio may have allowed for mineralization of N, while, after incubation in the laboratory, degradation of recalcitrant compounds may have resulted in immobilization of mineralized N. Alternatively, in the field, moisture and temperature were not constant over time. When soil is rewetted, capillary water in macropores reconnects the enclosures allowing N and motile organisms to diffuse and migrate from previously isolated enclosures (Addiscott, 1977; Hattori, 1994). Due to time lags in the adjustments of biological activity, microbial activity may remain high for a period after rewetting, even when migration of nematodes reduces grazing pressure. At the cessation of grazing, microbial growth rates would remain in the logistic range until microbial biomass approached carrying capacity. This would be reflected in high C mineralization and an accumulation of mineral N in soil solution.

If the *enclosure hypothesis* is correct, spatial patterns of biological and abiotic soil properties should coincide at low moisture. At the local scale, our data suggest that, in August, microbivores controlled C mineralization. But the numbers of microbivores might have been controlled themselves by the large number of omnivores/predators, which in August were exceptionally high everywhere in the field (Fig. 3), suggesting substantial energy fluxes into the predator pool. At the field scale in August 1997, traditional statistical linear analyses suggest that C and N mineralization was controlled by microbivory, which in turn may have been controlled by predation. Additionally, the lack of apparent statistically significant trophic relationships in May 1998, mirrored by lack of coincidences in local patterns of soil respiration or accumulated mineral N and biotic soil properties, are inconsistent with the *exclusion hypothesis* (Fig. 4). Greater moisture content may have relaxed the small-scale spatial coupling imposed on the interactions among food web levels by entrapment in enclosures. The loss of coupling is also expressed in a lower predator-to-microbivore ratio than in August 1997 (Fig. 1B).

We conclude that the *exclusion hypothesis* does not explain field patterns of soil respiration or mineral N content. An alternative hypothesis suggests that food web interactions are more intense at low moisture as a result of enclosure of predators, grazers and microorganisms. Under dry soil conditions, soil respiration and mineral N content were related to microbivore and predator abundance.

Acknowledgements

We thank Jodi Lyons and Tom Weicht for their technical assistance in analyzing our soil samples. This work was carried out with financial assistance from the National Research Initiative Competitive Grants Program of the United States Department of Agriculture and the Rhode Island Agricultural Experiment Station.

References

Addiscott, T.M., 1977. A simple computer model for leaching in structured soils. Journal of Soil Science 28, 554–563.

Addiscott, T.M., 1983. Kinetics and temperature relationships of mineralization and nitrification in Rothamsted soils with differing histories. Soil Science 34, 343–353.

Amador, J.A., Wang, Y., Savin, M.C., Görres, J.H., 2000. Fine-scale spatial variability of physical and biological soil properties in Kingston, Rhode Island. Geoderma 98, 83–94.

Anderson, R.V., Coleman, D.C., Cole, C.V., Elliott, E.T., 1981. Effect of the nematodes *Acrobeloides* sp. and *Mesodiplogaster lheritieri* on substrate utilization and nitrogen and phosphorous mineralization in soil. Ecology 62, 549–555.

Anderson, R.V., Gould, W.D., Woods, L.E., Cambardella, C., Ingham,

R.E., Coleman, D.C., 1983. Organic and inorganic nitrogenous losses by microbivorous nematodes in soil. Oikos 40, 75–80.

Ayoub, S.M., 1980. Plant Nematology, An Agricultural Training Aid. NemaAid Publications, Sacramento, CA.

Bowden, R.D., Newkirk, K.M., Rullo, G.M., 1998. Carbon dioxide and methane fluxes by a forest soil under laboratory-controlled moisture and temperature conditions. Soil Biology & Biochemistry 30, 1591–1597.

Brookes, P.C., Landman, A., Pruden, G., Jenkinson, D.S., 1985. Chloroform fumigation and the release of soil nitrogen: a rapid direct extraction method to measure microbial biomass nitrogen in soil. Soil Biology & Biochemistry 17, 837–842.

Burgess, T.M., Webster, R., 1980. Optimal interpolation and isarithmic mapping of soil properties. I. The semivariogram and punctual kriging. Journal of Soil Science 31, 315–331.

Cabrera, M.L., Beare, M.H., 1993. Alkaline persulfate oxidation for determining total nitrogen in microbial biomass extracts. Soil Science Society of America Journal 57, 1007–1012.

Cambardella, C.A., Moorman, T.B., Novak, J.M., Parkin, T.B., Karlen, D.L., Turco, R.F., Konopka, A.E., 1994. Field-scale variability of soil properties in central Iowa soils. Soil Science Society of America Journal 58, 1501–1511.

Caveness, F.E., Jensen, H.J., 1955. Modification of the centrifugal-flotation technique for the isolation and concentration of nematodes and their eggs from soil and plant tissue. Proceedings of the Helminthology Society 22, 87–89.

Coleman, D.C., Cole, C.V., Anderson, R.V., Blaha, M., Campion, M.K., Clarholm, M., Elliott, E.T., Hunt, H.W., Shaefer, B., Sinclair, J., 1977. An analysis of rhizosphere–saprophage interactions in terrestrial ecosystems. In: Lohm U., Persson, T. (Eds.), Soil Organisms as Components of Ecosystems. Proceedings of the 6th International Colloquium of Soil Zoology, Ecological Bulletin, vol. 25, Stockholm, pp. 299–309.

Darbyshire, J.F., 1976. Effect of water suctions on the growth in soil of the ciliate *Colpoda steinii* and the bacterium *Azotobacter chroococcum*. Journal of Soil Science 27, 369–376.

Edwards, N.T., 1975. Effects of temperature and moisture on carbon dioxide evolution in a mixed deciduous forest floor. Soil Science Society of America Proceedings 39, 361–365.

Elliott, E.T., Anderson, R.V., Coleman, D.C., Cole, C.V., 1980. Habitable pore space and microbial trophic interactions. Oikos 35, 327–335.

Ettema, C.E., Coleman, D.C., Vellidis, G., Lowrance, R., Rathbun, S.L., 1998. Spatiotemporal distributions of bacterivorous nematodes and soil resources in a restored riparian wetland. Ecology 79, 2721–2734.

Görres, J.H., Savin, M.C., Amador, J.A., 1997. Dynamics of carbon and nitrogen mineralization, microbial biomass, and nematode abundance within and outside the burrow walls of anecic earthworms (*Lumbricus terrestris*). Soil Science 162, 666–671.

Görres, J.H., Dichiaro, M.J., Lyons, J.B., Amador, J.A., 1998. Spatial and temporal patterns of soil biological activity in a forest and an old field. Soil Biology & Biochemistry 30, 219–230.

Görres, J.H., Savin, M.C., Neher, D.A., Weicht, T.R., Amador, J.A., 1999. Grazing in a porous environment: 1. The effect of pore structure on C and N mineralization. Plant and Soil 212, 75–83.

Griffiths, B.S., 1994. Soil nutrient flow. In: Darbyshire, J.F. (Ed.). Soil Protozoa, CAB International, Oxford, pp. 65–91.

Griffiths, B.S., Young, I.M., Caul, S., 1995. Nematode and protozoan dynamics on decomposing barley leaves incubated at different matric potentials. Pedobiologia 39, 454–461.

Groffman, P.M., Tiedje, J.M., 1988. Denitrification hysteresis during wetting and drying cycles in soil. Soil Science Society of America Journal 52, 1626–1629.

Grundmann, G.L., Renault, P., Rosso, L., Bardin, R., 1995. Differential effects of soil water content and temperature on nitrification and aeration. Soil Science Society of America Journal 59, 1342–1349.

Hassink, J., Bouwman, L.A., Zwart, K.B., Brussaard, L., 1993. Relationship between habitable pore space, soil biota and mineralization rates in grassland soils. Soil Biology & Biochemistry 25, 47–55.

Hattori, T., 1994. Soil microenvironment. In: Darbyshire, J.F. (Ed.). Soil Protozoa, CAB International, Oxford, pp. 43–64.

Hillel, D., 1971. Soil and Water: Physical Principles and Processes. Academic Press, New York.

Hunt, H.W., Coleman, D.C., Ingham, E.R., Ingham, R.E., Elliott, E.T., Moore, J.C., Rose, S.L., Reid, C.P.P., Morley, C.R., 1987. The detrital food web in a short-grass prairie. Biology and Fertility of Soils 3, 57–68.

Ingham, R.E., Trofymow, J.A., Ingham, E.R., Coleman, D.C., 1985. Interactions of bacteria, fungi, and their nematode grazers: effect on nutrient cycling and plant growth. Ecological Monographs 55, 119–140.

Isaaks, E.H., Srivastava, R.M., 1989. An Introduction to Applied Geostatistics. Oxford University Press, New York.

Jones, F.G.W., Larbey, D.W., Parott, D.M., 1969. The influence of soil structure and moisture on nematodes, especially *Xiphinema*, *Longidorus*, *Trichodorus* and *Heterodera* spp. Soil Biology & Biochemistry 1, 153–165.

Keeney, D.R., Nelson, D.W., 1982. Nitrogen-inorganic forms. In: Page, A.L., Miller, R.H., Keeney, D.R. (Eds.). Methods of Soil Analysis, Part 2, ASA, SSSA, Madison, WI, pp. 643–698.

Mary, B., Recous, S., Darwis, D., Robin, D., 1996. Interactions between decomposition of plant residues and nitrogen cycling in soil. Plant and Soil 181, 71–82.

Myrold, D.D., 1998. Transformations of nitrogen. In: Sylvia, D.M., Fuhrmann, J.J., Hartel, P.G., Zuberer, D.A. (Eds.). Principles and Applications of Soil Microbiology, Prentice Hall, Upper Saddle River, NJ, pp. 259–294.

Neher, D.A., Campbell, C.L., 1994. Nematode communities and microbial biomass in soils with annual and perennial crops. Applied Soil Ecology 1, 17–28.

Neher, D.A., Weicht, T.R., Savin, M., Görres, J.H., Amador, J.A., 1999. Trophic interactions in a porous environment 2. Nematode community structure. Plant and Soil 212, 85–99.

Norman, J.M., Gracia, R., Verma, S.B., 1992. Soil surface CO_2 fluxes and carbon budget of a grassland. Journal of Geophysical Research 97, 18845–18853.

Rao, P.S.C., Wagenet, R.J., 1985. Spatial variability of pesticides in field soils: methods for data analysis and consequences. Weed Science 33, 18–24.

Robertson, G.P., Huston, M.A., Evans, F.C., Tiedje, J.M., 1988. Spatial variability in a successional plant community: patterns of nitrogen availability. Ecology 69, 1517–1524.

Robertson, G.P., Freckman, D.W., 1995. The spatial distribution of nematode trophic groups across a cultivated ecosystem. Ecology 76, 1425–1432.

Savin, M.C., Görres, J.H., Neher, D., Amador, J.A., 2000. Uncoupling of carbon and nitrogen mineralization: role of microbivorous nematodes, submitted for publication.

Schimel, J.P., Jackson, L.E., Firestone, M.K., 1989. Spatial and temporal effects on plant–microbial competition for inorganic nitrogen in a California annual grassland. Soil Biology & Biochemistry 21, 1059–1066.

Schnürer, J., Clarholm, M., Boström, S., Rosswall, T., 1986. Effects of soil microorganisms and nematodes: a field experiment. Microbial Ecology 12, 217–230.

Sierra, J., 1997. Temperature and soil moisture dependence of N mineralization in intact soil cores. Soil Biology & Biochemistry 29, 1557–1563.

Smith, J.L., 1994. Cycling of nitrogen through microbial activity. In: Hatfield, J.L., Stewart, B.A. (Eds.). Soil Biology: Effects on Soil Quality, Lewis Publishers, Boca Raton, FL, pp. 91–120.

Sohlenius, B., 1980. Abundance, biomass, and contribution to energy flow by soil nematodes in terrestrial ecosystems. Oikos 34, 186–194.

Stanford, G., Frere, M.H., Schwaninger, D.H., 1973. Temperature coefficient of soil nitrogen mineralization. Soil Science 115, 321–323.

Thorne, G., 1961. Principles of Nematology. McGraw-Hill, New York.

Traunspurger, W., Bergtold, M., Goedkoop, W., 1997. The effect of

nematodes on bacterial activity and abundance in freshwater sediment. Oecologia 112, 118–122.

Vance, E.D., Brookes, P.C., Jenkinson, D.S., 1987. An extraction method for measuring soil microbial biomass C. Soil Biology & Biochemistry 19, 703–707.

Verhoef, H.A., Brussaard, L., 1990. Decomposition and nitrogen mineralization in natural and agroecosystems: the contribution of soil animals. Biogeochemistry 11, 175–211.

Wagai, R., Brye, K.R., Gower, S.T., Norman, J.M., Bundy, L.G., 1998. Land use and environmental factors influencing soil surface CO_2 flux and microbial biomass in natural and managed ecosystems in southern Wisconsin. Soil Biology & Biochemistry 30, 1501–1509.

Wallace, H.R., 1958. Movement of eelworms: I. The influence of pore size and moisture content of the soil on the migration of the larvae of the beet eelworm, *Heterodera schachterii*, Schmidt. Annals of Applied Biology 46, 74–85.

Wardle, D.A., Yeates, G.W., 1993. The dual importance of competition and predation as regulatory forces in terrestrial ecosystems: evidence from decomposer food webs. Oecologia 93, 303–306.

Wardle, D.A., Yeates, G.W., Watson, R.N., Nicholson, K.S., 1995. The detritus food-web and the diversity of soil fauna as indicators of disturbance regimes in agro-ecosystems. Plant and Soil 170, 35–43.

Wildung, R.E., Garland, T.R., Buschbom, R.L., 1975. The interdependent effects of soil temperature and water content on soil respiration rate and plant root decomposition in arid grassland soils. Soil Biology & Biochemistry 7, 373–378.

Woods, L.E., Cole, C.V., Elliot, E.T., Anderson, R.V., Coleman, D.C., 1982. Nitrogen transformations in soil as affected by bacterial–microfaunal interactions. Soil Biology & Biochemistry 14, 93–98.

Yeates, G.W., Bongers, T., de Goede, R.G.M., Freckman, D.W., Georgieva, S.S., 1993. Feeding habits in soil nematode families and genera — an outline for soil ecologists. Journal of Nematology 25, 315–331.