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Jorge Sierra

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Relationship between Mineral N Content and N Mineralization Rate in Disturbed and Undisturbed Soil Samples Incubated Under Field and Laboratory Conditions

J. Sierra

Departamento de Suelos, Facultad de Agronomia, Universidad de Buenos Aires, Av. San Martin 4453, 1417 Buenos Aires, Argentina.
Present address: Station de Science du Sol, INRA Avignon, Domaine Saint Paul, BP 91, 84143 Montfavet, France.

Abstract

An investigation of *in situ* N mineralization, using undisturbed soil samples, indicated a negative relationship between the mineral N content [(NO_3+NH_4)-N] at the beginning of the experiment and the mineral N produced during it. This suggests that a maximum value of mineral N accumulation in intact soil cores could be calculated from the relationship between mineral N content and N mineralization rate. This value would be related to the size of the mineralizable N pool. If this hypothesis is true, the amount of mineralizable N could be estimated from *in situ* incubations and utilized in the modelling of N mineralization in the field.

The aim of this work was to verify this hypothesis. The relationship between the mineral N content and the N mineralization rate was analysed for *in situ* and laboratory incubations of disturbed and undisturbed soil samples. A negative relationship between the two variables was only obtained for the experiments carried out with undisturbed samples (in the field and laboratory incubations) when the soil moisture content was not limiting for N mineralization. Futhermore, in undisturbed samples, a negative relationship between mineralization rates of consecutive incubation periods was observed, i.e. the soil sample producing relatively more, during a given period, produced relatively less in the following period. This relationship suggests a feedback mechanism operating in N mineralization which would be related to a mineralization–immobilization process in soil microsites. Thus, the N mineralization pattern was more complex than that described by initial hypothesis. The possible consequence of this feedback mechanism on *in situ* N dynamics is discussed.

Keywords: feedback mechanism, *in situ* soil incubation, mineralizable N, soil conditions, soil microsites.

Introduction

Knowledge of the dynamics of N mineralization is necessary to determine the production of N available for plants, and to make reliable fertilizer recommendations from an economical, as well as from an ecological, point of view.

Several studies to evaluate N mineralization were based on laboratory or field incubations of disturbed soil samples (Stanford and Smith 1972; Smith *et al.* 1977; Westermann and Crothers 1980; Campbell *et al.* 1984). It is well known that the handling and the treatment of the sample influences the amount of N mineralized during these experiments (Macduff and White 1985; Seneviratne and

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Wild 1985). Some authors have shown that measurements of N mineralization made in undisturbed and disturbed soil samples are not in good agreement. Further, the dynamics of N mineralization is different in both cases (Raison *et al.* 1987; Cabrera and Kissel 1988*a*; Hadas *et al.* 1989). Due to the disruption of the aggregates, the use of disturbed samples allows additional organic N to become accessible to the microflora and can increase N mineralization (Craswell *et al.* 1970). The size of this additional N pool depends upon the degree of physical protection and, consequently, on the size and stability of the aggregates (Sollins *et al.* 1984; Nordmeyer and Richter 1985). These factors vary with agricultural practice, so that, even for very similar soils, the agreement between both measurements is poor (Zourarakis *et al.* 1987; Frazer *et al.* 1990).

A major problem for improving the modelling of soil N mineralization is the estimate of a pool (or pools) of mineralizable N (Broadbent 1986). To analyse experiments using disturbed soil samples, several workers have utilized exponential models by assuming one or more mineralizable N pools of discrete size (Stanford and Smith 1972; Molina *et al.* 1980; Deans *et al.* 1986; Cabrera and Kissel 1988*a*). However, serious errors in the estimation of these pools have been noted (Broadbent 1986; Cabrera and Kissel 1988*c*; Sierra 1990). Using undisturbed soil samples in laboratory incubations, Cabrera and Kissel (1988*a*) found that the N mineralizable pool and its decomposition rate constant, calculated by an exponential model, provided a reasonable estimate of the amount of N mineralized in the field. Thus, a better approach to understanding the N mineralization process may be obtained by using undisturbed soil samples.

In situ incubations of undisturbed soil samples can be helpful in order to validate the N mineralization models obtained from laboratory studies. However, only a few studies of *in situ* N mineralization in undisturbed samples have been reported. *In situ* incubations of undisturbed soil samples were made by Zourarakis *et al.* (1987) in Argentine Pampa. They found a negative relationship between the mineral N content [(NO_3+NH_4)-N] at the beginning of the experiment and the mineral N produced during it. That is, the sampling sites that had the higher initial mineral N content produced less mineral N during the experiment. This relationship varied depending upon soil water content, becoming weaker at low moisture values. This pattern of N mineralization could imply that the sampling sites with a relatively high mineral N content at the beginning of the experiment have less mineralizable N remaining in the soil, and thus that their mineralization rate is smaller during the period of incubation.

If this hypothesis is true, a maximum value of mineral N accumulation, related to the amount of mineralizable N, could be estimated from the relationship between mineral N content and mineralization rate, and utilized in the modelling of soil N mineralization. The present study was carried out to verify this

Table 1. Some properties of the soil used (surface horizon)

Plot	Organic nitrogen (%)	Organic carbon (%)	pH	Texture
A	0·20	2·3	6·3	loam
B	0·20	2·5	6·4	loam

hypothesis. More specifically, the objective of this work was to analyse, during field and laboratory experiments, the relationship between the mineral N content and the mineralization rate as a function of time, soil moisture content and soil disturbance.

Materials and Methods

Soil and Field Experiment Design

The experiment was carried out on a farm located at Salto (34° 32′ S., 60° 21′ W.) in the corn belt of the Argentine Pampa. The soil at this location is a Typic Argiudoll. Two plots of 10 ha, each cultivated with corn, were selected (designated A and B). The plots had very similar soil properties (Table 1) and management regime (7 years of continuous corn). At the beginning of the experiment, the soil matric potential was 30 and 1300 kPa for the two plots, respectively. The experiment was run from day 60 to day 116 after sowing. Three subplots were established in each plot (designated I, II and III) with 10 incubation sites per subplot (Fig. 1).

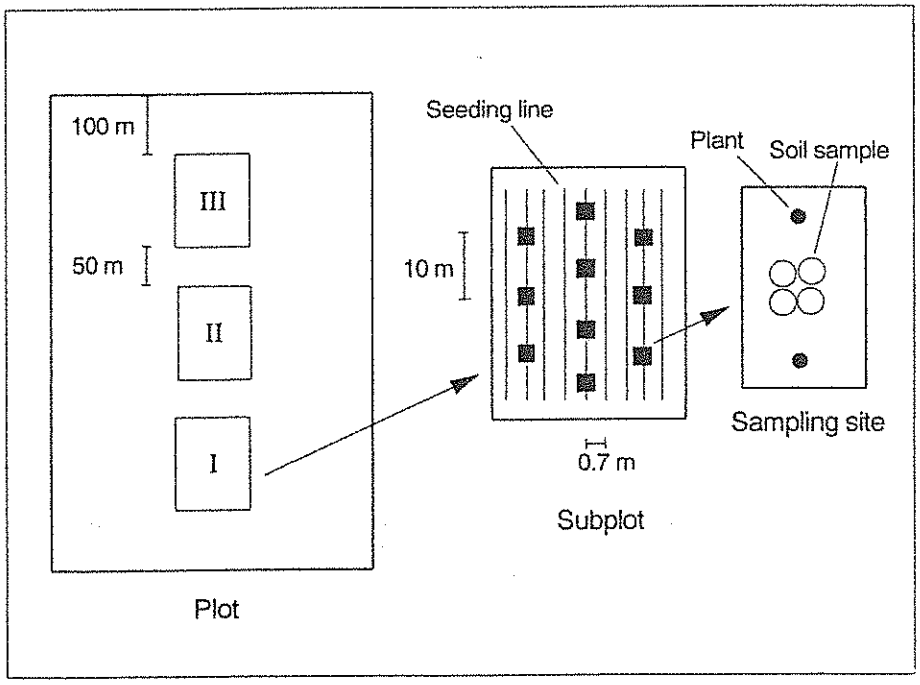

Fig. 1. Field experiment design.

Field Incubations of Undisturbed Soil Samples

Incubations were carried out in PVC cylinders 5 cm deep, 6·5 cm in external diameter and with a wall thickness of 0·2 cm. Each incubation site was located on the seeding line between plants. Four undisturbed soil samples were extracted at each sampling site between approximately 3 and 8 cm depth (Fig. 1). One of them was utilized to make initial measurements. The others were placed in polyethylene bags (20 μm thick) to prevent evaporation and then in plastic rectangular boxes measuring 8×6 cm. The boxes were reburied in the original holes and covered with soil. The boxes prevented the entrance of plant roots to the soil sample. Holes with diameter of about 0·1 cm were drilled in the boxes in order to facilitate aeration (Zourarakis *et al.* 1987).

One cylinder was selected at random from each site and was removed after 2, 4 and 8 weeks. For each sample, water content was determined gravimetrically, nitrate content colorimetrically by hydrazine reduction (Kampshake *et al.* 1967) and ammonium content by the phenol-nitroprusiate method (Kaplan 1965).

Laboratory Incubations of Undisturbed Soil Samples

At the end of the field experiment, 12 undisturbed samples were removed at random in Plot A. These samples were placed in polyethylene bags (20 μm thick) and returned to the laboratory for incubation. The undisturbed samples were leached before incubation with 500 mL of 0·01 M $CaCl_2$ and their water suction was adjusted under vacuum to 30 kPa. The nitrate and ammonium content in the leachate were determined as described above. After leaching, the soil samples were replaced in their bags and incubated for 10 days at 35°C. At the end of this period, the samples were leached and the nitrate and ammonium content determined. The samples were then incubated at 25°C for 10 more days. At the end of this period, the samples were leached again, the mineral N was determined and the samples were dried at 105°C in order to determine the mass of dried soil.

Laboratory Incubations of Disturbed Soil Samples

The 10 soil samples utilized to make initial measurements from plot A-subplot I were used. The samples were dried and sieved (<2 mm). Two 30 g subsamples from each sample were placed in plastic flasks and distilled water was added to obtain matric potentials of 30 and 1300 kPa, respectively. The flasks were covered with a polyethylene film (20 μm thick) and incubated for 10 days at 35°C and later for 10 days at 25°C. The nitrate and ammonium contents were measured after 0, 10 and 20 days of incubation.

The reason for the variation of incubation temperatures during these experiments is discussed below.

The Variables and Hypothesis in the Field Experiment

Two kinds of variables may be defined: site variables, S, and production variables, P. Site variables represent the mineral N content [(NO_3+NH_4)-N] at each sampling date (S_0, S_2, S_4, S_8). Production variables are obtained by taking the difference between two site variables. Thus, P_{0-2} is the mineral N produced between time zero and the second week, that is $P_{0-2} = S_2 - S_0$. Here we are concerned with net mineral N production because immobilization and denitrification may occur simultaneously with ammonification and nitrification (Nordmeyer and Richter 1985; Parkin 1987).

S_{ijk} is defined as the mineral N content in cylinder j at incubation site i on sampling date k; thus the net mineral N produced at incubation site i in the *(k, k+1)* period is

$$P_{i(k,k+1)} = S_{ij(k+1)} - S_{ij(k)+\epsilon_i}\,, \tag{1}$$

where ϵ_i is the error of estimation related to the variance of S_i (σ_s^2 within site): the greater the σ_s^2 within-site, the greater will be ϵ_i. It is usual in field experiments to assume that σ_s^2 within-site is less than the between sites variability (σ_s^2 between-sites). However, in the present work it is necessary to verify this in order to confirm the validity of the relationship between mineral N content and mineral N production. According to equation (1), if the mineral N content $S_{ij(k)}$ is overestimated, then the mineral N production $P_{i(k,k+1)}$ is underestimated. Thus, if σ_s^2 within-site and σ_s^2 between-sites have the same order of magnitude, a negative relationship between S and P variables exists due to error ϵ_i (Benyaklef 1977).

This was tested by comparing the negative correlation between S and P variables observed in this study (Table 2) and the negative correlation due to error ϵ_i (Benyaklef 1977).

Results and Discussion

N Mineralization Rates

The initial soil water content in the field experiment was 0·28±0·02 g g^{-1} (30 kPa) in plot A and 0·15±0·02 g g^{-1} (1300 kPa) in plot B. The coefficient of

variation within subplots was never higher than 7%. The moisture losses during the whole experiment were negligible.

Table 2. Linear correlation coefficients between the production *(P)*and site variables *(S)*

Variables	Plot A Subplot			Plot B Subplot		
	I	II	III	I	II	III
S_0/P_{0-2}	$-0{\cdot}81^{**}$	$-0{\cdot}56$	$-0{\cdot}23^{A}$	$-0{\cdot}07^{A}$	$-0{\cdot}40$	$-0{\cdot}29$
S_2/P_{2-4}	$-0{\cdot}78^{**}$	$-0{\cdot}87^{**}$	$-0{\cdot}44$	$-0{\cdot}51$	$-0{\cdot}61$	$-0{\cdot}64^{*}$
S_4/P_{4-8}	$-0{\cdot}73^{*}$	$0{\cdot}07^{A}$	$-0{\cdot}71^{*}$	$-0{\cdot}05^{A}$	$-0{\cdot}78^{*}$	$-0{\cdot}51$

$^{*}P < 0{\cdot}05$, $^{**}P < 0{\cdot}01$.
[A] The covariance was less than COV* (equation 2) (see text).

For field and laboratory experiments, the ammonium content ranged from 0·4 to 1·0 mg N kg^{-1}. These results are in good agreement with those obtained by Zourarakis *et al.* (1987) for similar soils. These values of ammonium content represent 3–8% of the total mineral N measured for each incubation period of each experiment. It is well known that the ammonification and nitrification rates depend on a large number of variables. Whilst the rate of nitrification may depend on the rate of ammonification (Gilmour 1984; Darrah *et al.* 1985; Macduff and White 1985), it is also a function of a number of other factors, which may affect ammonification to a greater or lesser extent than they affect nitrification (Gilmour 1984; Frazer *et al.* 1990). Gilmour (1984) reported that zero-order kinetics best described nitrification as a function of ammonium concentration, which ranged from 43 to 124 mg NH_4-N kg^{-1}. In a comprehensive study, Darrah *et al.* (1985, 1986) utilized a modified Michaelis–Menten equation to calculate the rate of nitrification from the ammonium concentration; the affinity constant used was 0·02 μ mol NH_4-N cm^{-3} of soil solution (approx. 0·06 mg NH_4-N kg^{-1}). With this value the maximum rate of nitrification was reached at 2–3 mg NH_4-N kg^{-1}. For several soils of the Argentine Pampa, Navarro *et al.* (1980) noted that the ammonium concentration for the maximum nitrification rate ranged from 5 to 10 mg NH_4-N kg^{-1}. Futhermore, they observed that the rate of nitrification was always higher than the rate of ammonification. In these well aerated soils, ammonium is oxidized to nitrate rapidly enough so that ammonium does not accumulate. Consequently, the rate limiting step of N mineralization is the ammonification rate. Similar results were reported by Stanford *et al.* (1973) and Macduff and White (1985).

Fig. 2 shows the time course data of mineral N content in both plots for the field experiment. After 8 weeks, the amount of mineral N produced was 22·9 mg N kg^{-1} in plot A and 11·7 mg N kg^{-1} in plot B. These values are equivalent to mineralization of approximately 1% and 0·5% of the initial organic nitrogen, respectively. There were significant differences in mineral N produced between plots, but they were not found between subplots. This was due mainly to the different water content of the plots, which was similar for all subplots within a given plot. The coefficient of variation (CV) of mineral N produced for each subplot and each period ranged from 20 to 30% in plot A and from 30 to 70% in plot B.

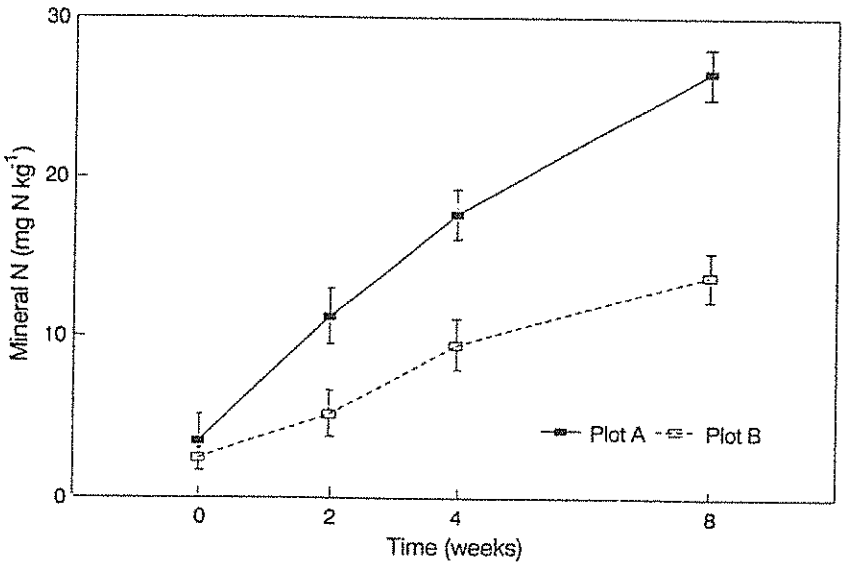

Fig. 2. Time-course data for mineral N produced during the field experiment. The vertical bars indicate the standard error. The soil water content was 0·28 g g^{-1} (30 kPa) in plot A and 0·15 g g^{-1} (1300 kPa) in plot B.

The rate of N mineralization in this study is about twice that reported for forest soils for *in situ* measurements with similar soil conditions (Raison *et al.* 1987; Poovarodom *et al.* 1988; Frazer *et al.* 1990), but is of the same order of magnitude as those obtained in agricultural soils (Cabrera and Kissel 1988*b*; Hadas *et al.* 1989). The high degree of spatial variability in N mineralization observed in each subplot was not surprising and similar variability for *in situ* studies was found by other authors for forest as well as for agricultural soils (Raison *et al.* 1987; Zourarakis *et al.* 1987; Poovarodom *et al.* 1988; Frazer *et al.* 1990). It is known that several factors may induce this high degree of spatial variability in N mineralization (Frazer *et al.* 1990; Bramley and White 1991*a*, 1991*b*; Mazzarino *et al.* 1991). For a soil similar to the one used in this study, Hashimoto *et al.* (1988) noted that the spatial variation in the light fraction of soil organic matter (Sollins *et al.* 1984) was able to explain 50% of the variation of N mineralization measured for field incubations of undisturbed samples.

A significant amount of mineral N was produced at 1300 kPa (plot B, Fig. 2). This result is in agreement with those reported by Frazer *et al.* (1990) and Mazzarino *et al.* (1991). At this soil matric potential, the spatial variability of N mineralization was higher than at a matric potential of 30 kPa. This variability cannot be attributed to soil moisture variation because this was small. Bramley and White (1990) found significant nitrifying activity at matric suction as great as 10^5 kPa, however, a large variation in activity at each soil moisture content was also observed. They noted that soil moisture stress was not as critical in regulating nitrifier activity as was soil pH, and they concluded that the nitrifier population is tolerant to changes in soil moisture because it is adapted to seasonal soil moisture variations. Frazer *et al.* (1990) proposed that soil microaggregates

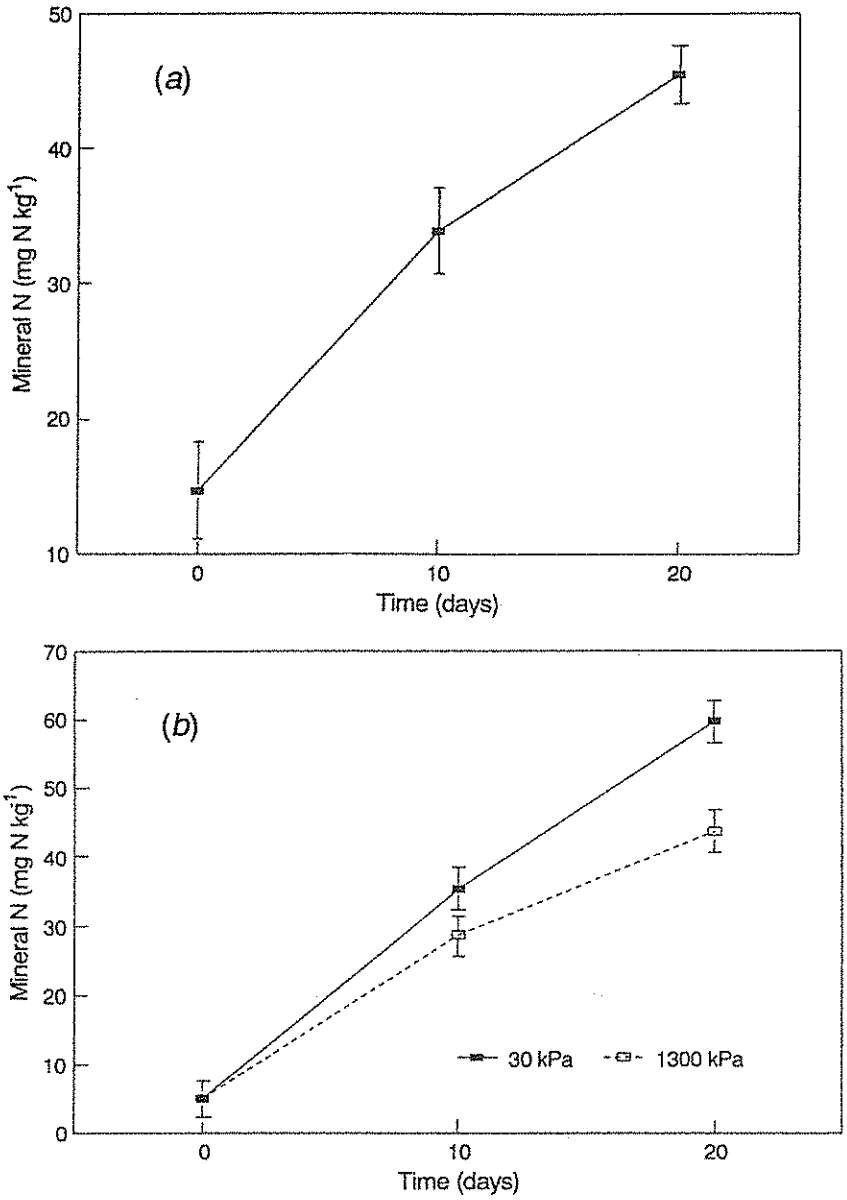

Fig. 3. Time-course for mineral N produced during the laboratory experiments: (*a*) laboratory incubations of undisturbed soil samples and (*b*) laboratory incubations of disturbed soil samples at two different soil moisture levels. The vertical bars indicate the standard error.

may retain appreciable moisture after gravimetric drainage ended. The moisture in these microsites may be sufficient for ammonification and nitrification to continue during the dry season. Thus, spatial variation in soil structure (i.e. amount of microaggregates) might be important to induce a high variability in N mineralization in undisturbed samples of dry soil.

The results of laboratory experiments are presented in Fig. 3. A comparison of N mineralization rates at 30 kPa shows that for the disturbed soil samples the N mineralized was 1·8 times higher than that for the undisturbed samples. These differences are in good agreement with the results obtained by Nordmeyer and Richter (1985) and Cabrera and Kissel (1988*a*). At 30 kPa, the coefficient of variation of the rate of N mineralization was smaller in disturbed samples (16% at 35°C, 13% at 25°C) than in undisturbed samples (39% at 35°C, 51% at 25°C). In undisturbed soil samples, N mineralization may be a spatially variable process, presumably due to variable physical protection of the mineralizable N pool (Sollins *et al.* 1984). Futhermore, the rupture of anaerobic soil microsites in disturbed soil samples induces a more homogeneous soil aeration (Parkin 1987). Similar results have been reported by Parkin *et al.* (1987) for denitrification; they found that potential denitrification showed a smaller degree of spatial variability and was less skewed than denitrification in undisturbed soil samples.

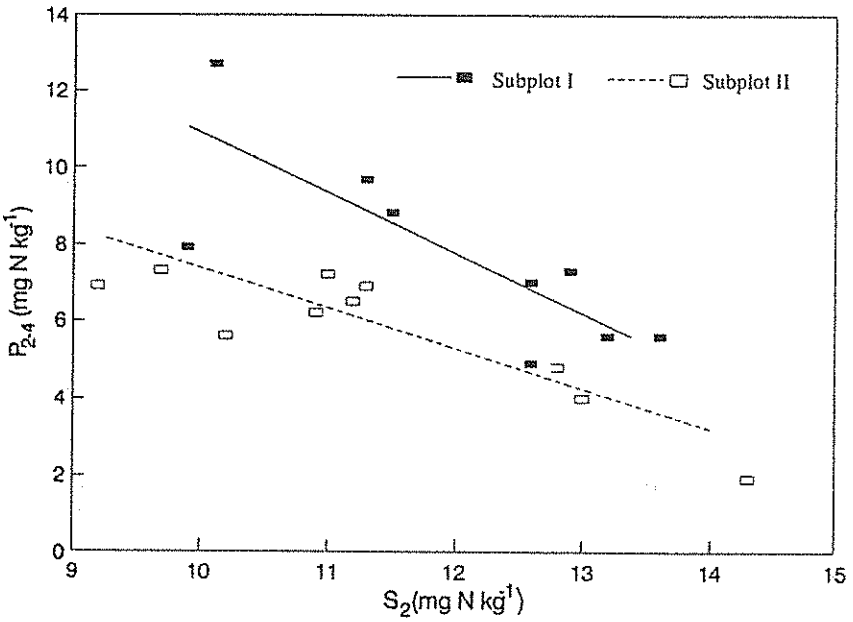

Fig. 4. Relationship between mineral N content and mineralization rate in plot A (field experiment). S_2 is the mineral N content after two weeks of incubation, P_{2-4} is the mineral N produced during the 2–4 weeks period (see Table 2 for levels of significance of each correlation).

Relationship between Mineral N Content and Mineralization Rate

In order to analyse the relationship between S and P variables, linear correlations between both variables were calculated. Table 2 gives the coefficients obtained for the field experiment. In plot A, negative correlation coefficients, significant in most cases, were obtained. For plot B, negative coefficients were again obtained but were only significant in two cases. These results were similar to those found

in a previous experiment with monthly *in situ* incubations (Zourarakis *et al.* 1987). In Fig. 4, the relationship between S_2 and P_{2-4} is shown for subplots I and II of plot A. It can be seen that the relationship between S and P variables was not the same in each subplot. When all 30 sites of each plot were considered, significant coefficients were only obtained for plot A (S_0/P_{0-2}, $r = -0{\cdot}59$; S_2/P_{2-4}, $r = -0{\cdot}42$; S_4/P_{4-8}, $r = -0{\cdot}66$, $P < 0{\cdot}05$).

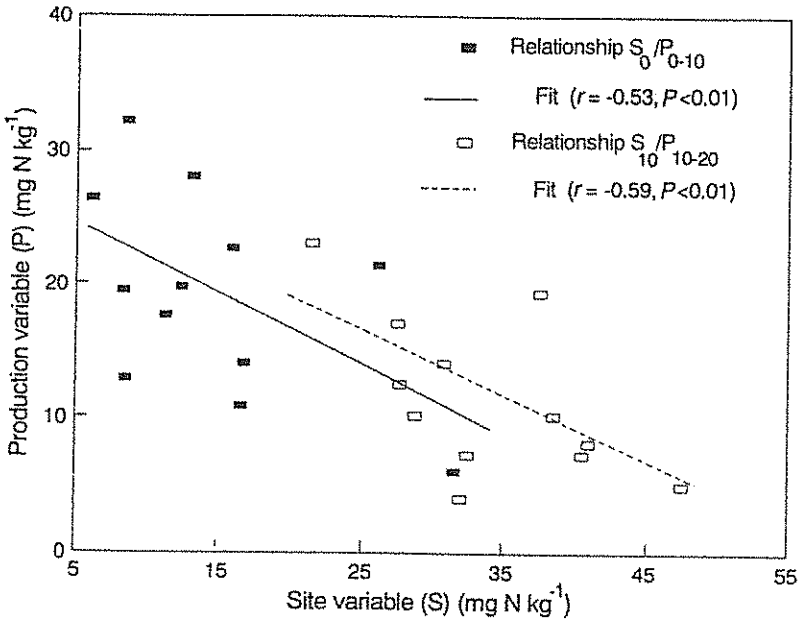

Fig. 5. Relationships between mineral N content and mineralization rate for the laboratory experiment using undisturbed soil samples. S_0 and S_{10} are the mineral N content at the beginning and after 10 days of incubation, respectively. P_{0-10} and P_{10-20} are the mineral N produced during the 0–10 days period and 10–20 days period, respectively.

For the laboratory experiment with disturbed samples, significant relationships between S and P variables were not found. In the other experiment using undisturbed samples, a weak correlation between both variables was observed (Fig. 5).

Bramley and White (1991*a*, 1991*b*) used geostatistical methods to analyse spatial variability in soil mineral N and nitrifying activity and found that these properties were spatially dependent over a short range. These authors also reported that the variability of biological soil properties had a temporal as well as a spatial component. These results raise the question as to whether the variance within site is less than the variance between sites in all of the experiments.

The analysis of the relationship between S and P variables implies an analysis of correlation between the intercept (S) and the slope (P) for a given period (see Fig. 2). For example, for the first 2-week period, S_0 is the intercept and P_{0-2} is the slope of the time course data for this period of measurement. If all 10 sites of each subplot are pooled (σ_s^2 within site = σ_s^2 between sites), the

covariance between both variables (COV^*) can be established (Benyaklef 1977):

$$\mathrm{COV}^*(S_0\, P_{0\text{–}2}) = -(k\sigma_r^2)/\sum(k-\bar{k})^2\,, \qquad (2)$$

where k is the sampling date (abscissa value in Fig. 2), $\bar{k}$ is the mean of k for a given period (i.e. for the 0–2 week period, $\bar{k}=1$), and $\sigma^2{}_r$ is the residual variance of the straight line. Equation (2) implies that if the time course data of each subplot are pooled, then a negative relationship between the S and P variables exists. The biological significance of this negative relationship, observed in the present work, depends on whether the covariance calculated by equation (2) (COV^*) was less than the covariance between S and P calculated from individual data of S_i and P_i in each subplot. This second covariance corresponds to the coefficients presented in Table 2.

The COV^* was computed for each subplot and period and compared with the covariance calculated from individual data. The results of this analysis showed that COV^* was small and less than the covariance from individual data except in four cases (see Table 2). This result implies that the relationship between S and P variables represents a true effect and is not an artifact due to sampling error.

The negative relationship between S and P variables observed for the field experiment is in accordance with the results obtained by Zourarakis *et al.* (1987). That is, the sampling sites that had the higher mineral N content produced less mineral N in the following incubation period. In the present study, the S-P relationship was observed during each period of field incubation and was particularly clear in subplot I of plot A which showed significant correlation in all three periods (Table 2).

Fig. 6 shows a scheme that represents the time course of mineral N content under the hypothesis of a maximum value in mineral N accumulation related to the amount of mineralizable N in the soil. The scheme describes the existence of a negative relationship between S and P variables in each period; that is, the sampling site with the lower mineral N content always has the higher rate of N mineralization. This is related to the amount of N needed to reach the maximum value (amount of mineralizable N remaining in the soil at the end of each incubation period). This hypothesis is supported by the conclusions that

(i) the mineralizable N pool is defined as an organic N pool of discrete size and the rate of mineralization is determined by the magnitude of this pool and its decomposition rate constant (Stanford and Smith 1972; Marion and Black 1987; Cabrera and Kissel 1988*a*);

(ii) the homogeneity found in N mineralization in disturbed soil samples suggests that the amount of mineralizable N has a small spatial variability (Stanford and Smith 1972; Zourarakis *et al.* 1987). This N pool may be assumed to be approximately the same for the soil samples of a given subplot.

If soil conditions (i.e. soil temperature) change during the experiment, as for the third period in Fig. 6, the decomposition rate will change with the same magnitude for all of the soil samples (Stanford *et al.* 1973; Marion and Black 1987). This implies that the negative relationship between S and P variables

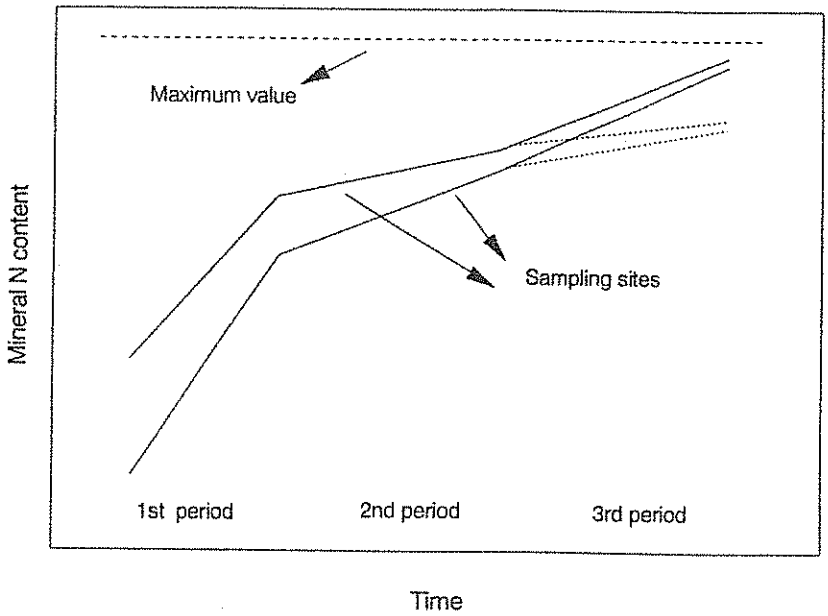

Fig. 6. Scheme representing the hypothesis of a maximum value in the mineral N accumulation process in intact soil cores. In the third period, two different situations are represented: a more (solid lines) and a less (dashed lines) favourable soil condition for N mineralization.

Table 3. Linear correlation coefficients between production variables *(P)*

Variables	Plot A				Plot B			
	Subplot			All plot	Subplot			All plot
	I	II	III		I	II	III	
P_{0-2}/P_{2-4}	−0·53	−0·85**	−0·40	−0·47**	−0·66*	−0·47	−0·56	−0·49**
P_{2-4}/P_{1-4}	−0·70*	−0·06	−0·80**	−0·61**	0·21	−0·73*	−0·23	−0·40
P_{0-2}/P_{2-8}	−0·51	−0·75*	−0·45	−0·50**	−0·56	0·05	−0·38	−0·38
P_{0-4}/P_{4-8}	−0·52	−0·14	−0·77**	−0·60**	0·01	−0·74*	−0·34	−0·21

*$P < 0·05$, **$P < 0·01$.

is independent of the magnitude of the mineralization rate (Fig. 6), which is consistent with the results obtained for each incubation period (Table 2, Fig. 2).

This hypothesis also implies that the sampling sites having the greater mineralization rate for a given period will have the greater rate of mineralization for the consecutive period. As discussed above, this is independent of the magnitude of the mineralization rate. Thus, the scheme in Fig. 6 predicts a positive relationship between mineralization rates in consecutive periods. In order to determine whether this is true, linear correlations between P variables of consecutive periods were calculated. Table 3 shows that the results were contrary to those predicted by Fig. 6. Consequently, a site producing relatively more during a given period, produced relatively less during the following incubation period. This negative relationship between P variables implies that the N mineralization pattern was more complex than that described in Fig. 6. A negative relationship between mineralization rates of consecutive incubation periods may reflect a

biological feedback mechanism operating in N mineralization. The laboratory experiment using undisturbed soil samples was carried out to verify the presence of this feedback mechanism.

Using undisturbed samples in a laboratory incubation study at constant soil temperature, Nordmeyer and Richter (1985) and Cabrera and Kissel (1988*a*) observed that N mineralization was a linear function of time. For the reasons discussed above, a linear accumulation of mineral N is a source of bias in the relationships studied in this work. To avoid this perturbation in the analysis of the results, the incubation temperature in the laboratory experiments was not constant but was changed during incubation (35°C for the first period and 25°C for the second period). This induced a nonlinear accumulation of mineral N (Fig. 3). It was assumed that the variation in incubation temperature affects the decomposition rate of each soil sample in a similar fashion (Marion and Black 1987) and that the relationship between P variables was not affected. This assumption is consistent with the results of the field incubations, during which the rate of N mineralization in plot A varied significantly throughout the study (0·55, 0·46 and 0·30 mg N kg^{-1} day^{-1} for the 0–2, 2–4 and 4–8 weeks period, respectively) and the negative correlation between P variables was observed in all of the experiment (Table 3). The results found in the laboratory incubation of undisturbed samples confirm the existence of a negative relationship between rates of N mineralization of consecutive periods (Fig. 7).

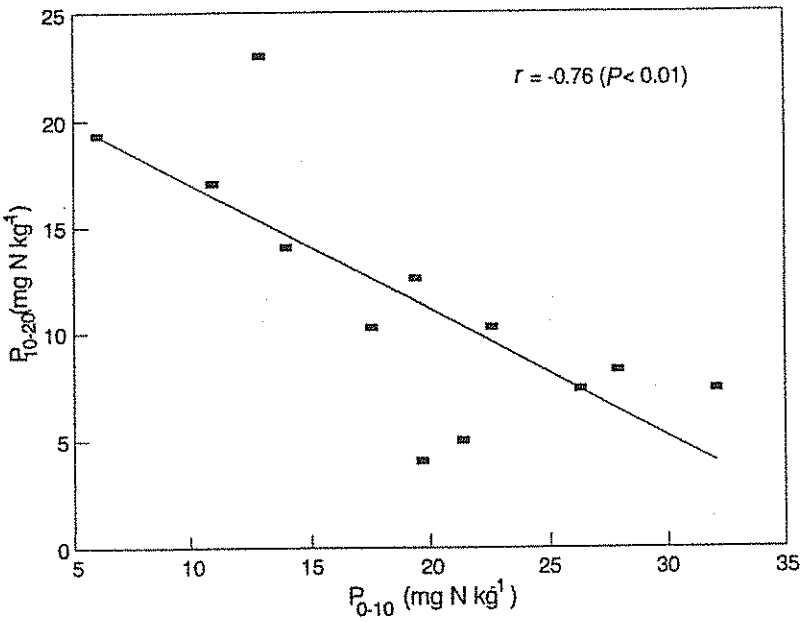

Fig. 7. Relationship between mineralization rates of consecutive incubation periods for the laboratory incubation of undisturbed samples (see Fig. 5 for nomenclature).

Sollins *et al.* (1984) observed that the mineral N released from the heavy fraction of organic matter can be immobilized by microorganisms decomposing the light fraction, this immobilization being regulated by the C:N ratio of the

light fraction. The feedback mechanism observed in the present study may be an effect of this N mineralization/immobilization pattern. In those sites that had higher mineral N production, there would be an exhaustion of the labile N that was proportionally higher and, consequently, the mineralization rate would be relatively less in the following period; labile N renewal or the colonization by microorganisms of new microsites with organic N available would allow relatively more production in the following period. This N mineralization pattern may coexist with an immobilization process of the released N which contributes to microbial growth and promotes labile N renewal or the colonization of other microsites.

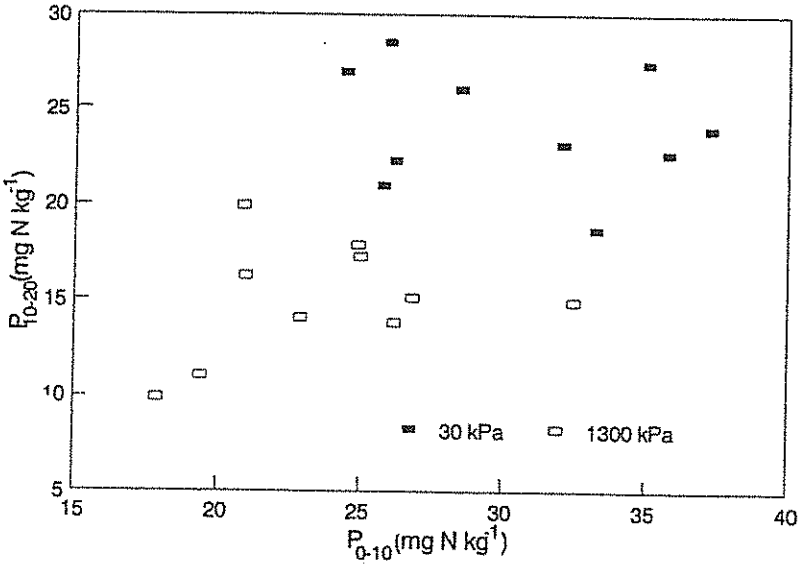

Fig. 8. Relationship between mineralization rates for the laboratory incubation of disturbed samples at two different soil moisture values (see Fig. 5 for nomenclature).

In disturbed samples, the number of accessible microsites with labile N is higher than in undisturbed samples (Craswell *et al.* 1970). Therefore, the exhaustion of labile N in some microsites would not have a great influence on the mineralization rate in the following period; this N mineralization pattern could explain the lack of significant correlation between N mineralization rates in disturbed soil samples (Fig. 8). Kilbertus (1980) observed a large degree of spatial variability in the distribution of microorganisms in the soil microaggregates. This spatial variability was associated with the presence of organic matter and with variation in pore size. Thus, soil aggregate disruption affects the amount of organic N available for soil biomass as well as the natural habitat of the microorganisms.

The hypothesis of a feedback mechanism, operating on N mineralization, suggests that the accumulation of mineral N is governed by the mineralizable N content in the accessible microsites, and may be modified by environmental variables. If the water content is limiting for N mineralization, as is the case for plot B in the field experiment, then the exhaustion of labile N is related to water availability in the accessible microsites. Under such soil conditions, the principal

factor governing N mineralization is not the N availability in the accessible microsites but the water content. Consequently, there is a poor correlation between P variables (Table 3).

As discussed above, the results of this study could not be described by the scheme shown in Fig. 6. However, there is some evidence that this scheme could be useful if longer incubation periods were utilized. An analysis of the S_0-P_{0-8} and S_2-P_{2-8} relationship, including all 30 sites of plot A, found significant negative correlations (S_0–P_{0-8}, $r = -0{\cdot}67$; S_2–P_{2-8}, $r = -0{\cdot}61$, $P < 0{\cdot}05$). According to this, the spatial variability was always less in S_8 (CV = 4–5%) than in S_0 (CV = 25–28%); that is, the spatial variability of mineral N content diminished with time as predicted by Fig. 6. Thus, on the basis of the results of this study, two process appear to be involved:

(i) a feedback mechanism operating in each period of incubation which determines the apparent or net mineralization rate as a result of a mineralization/immobilization process in the soil microsites;
(ii) a more long term process where the general pattern of N mineralization and the amount of mineral N released is governed by the soil mineralizable N.

Several authors (Stanford and Smith 1972; Molina *et al.* 1980; Deans *et al.* 1986; Cabrera and Kissel 1988*a*) have analysed N mineralization under varying soil conditions by assuming first-order kinetics for decomposition and one, two or more mineralizable N pools, each of them characterized by a particular rate constant. The feedback mechanism revealed in this study suggests that the net mineralization rate could result from interactions between fractions of organic N. Then, in short-term experiments, serious errors of estimation can be involved by assuming an independent rate constant for each N fraction (Sierra 1990).

The study of the feedback mechanism can be useful for understanding the interactive process between N fractions and improving the present models for N mineralization. The feedback and the long-term mechanisms are regulated by environmental variables. The literature is full of studies dealing with the effects of a whole range of environmental variables on N mineralization, but not on a feedback mechanism operating in N mineralization. Further work is necessary to analyse how environmental factors affect the feedback mechanism in a long-term incubation period. For this work, it seems advantageous to use undisturbed soil samples in laboratory incubations. This will allow the simulation of several soil conditions and will avoid a large experimental effort in the field.

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