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Aspects of carbon and nitrogen cycling in soils of the Bornhöved Lake district
I. Microbial characteristics and emissions of carbon dioxide and nitrous oxide of arable and grassland soils

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Abstract. Soil microbial biomass content, organic carbon mineralization as well as arginine ammonification rates were estimated in samples from arable and grassland soils and carbon dioxide and nitrous oxide emission rates were measured *in situ* at four sites along a catena. Soil microbial biomass content increased in the order, maize monoculture < crop rotation < dry grassland < wet grassland. The two arable soils had similar rates of carbon mineralization in the laboratory at 22 °C (basal respiration) as well as *in situ* (carbon dioxide emission) at field temperature. Under crop rotation, maize monoculture and dry grassland, the arginine ammonification rate significantly correlated to the microbial biomass content. In contrast, the biomass-specific ammonification rate was low in wet grassland soil, as were *in situ* N_2O emission rates. Data from all sites together revealed no general relationship between microbial biomass content and C and N mineralization rates. In addition, there was no general relationship between the quantity of soil microbial biomass and the emission rates of the greenhouse gases CO_2 and N_2O. The maize monoculture induced a soil microbial community that was less efficient in using organic carbon compounds, as shown by the high metabolic quotient (respiration rate per unit of biomass). However, microbial biomass content was proportional to basal respiration rate in soils under crop rotation, dry and wet grassland. Arginine ammonification rate was related to microbial biomass content only in fertilized soils. Applications of high quantities of inorganic nitrogen and farmyard manure apparently increase *in situ* N_2O emission rates, particularly under crop rotation. The microbial biomass in the unfertilized wet grassland soil seems to be a sink for nitrogen.

Introduction

The soil microbial community is the driving force for nutrient transformations and thus plays a major role in soil fertility and the functioning of ecosystems (Smith & Paul 1992). Soil microbial biomass is the 'eye of the needle' through

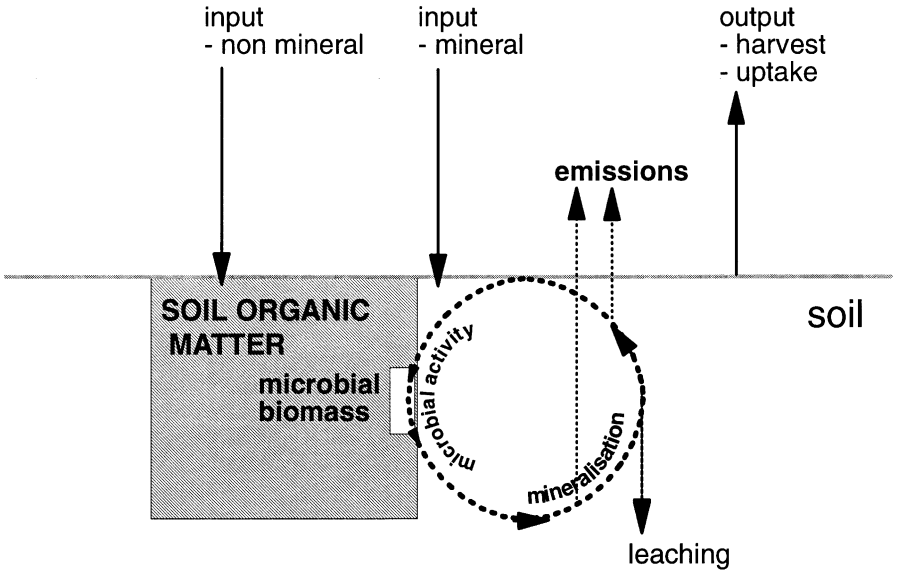

Figure 1. Aspects of carbon and nitrogen cycling in soils.

which all organic material that enters the soil must pass (Jenkinson 1977, in Martens 1995) and thus plays a major role in C and N cycling in soils. Microbial biomass is only a small fraction (1 to 3%) of the total organic C (Martens 1995) graphically presented in Figure 1. Yet microbial activity regulates C and N pools in soils and therefore ecosystem functioning as well as atmospheric chemistry through emission of CO_2 and N_2O (Figure 1).

Each microbiological estimate refers to different microbial components or transformation processes (summarized as microbiological properties). Different microbiological properties may only be correlated under specific soil properties and a certain structure of the microbial community. Relationships between different microbiological properties appear to provide ecological characteristics that are useful to understand microbial processes and elemental cycling in soils. Such an approach was developed by Anderson & Domsch (1990, 1993), who used the metabolic quotient, the ratio between microbial respiration rate and microbial biomass content, as an indicator for environmental conditions. In the present paper we used such ratios as indicators for C and N utilization efficiency of the microflora, and to evaluate transformation rates of elements and compounds by the microbial community.

The interdisciplinary project "Ecosystem Research in the Bornhöved Lake District" aims to analyze and model structures, dynamics and functioning of terrestrial (agricultural and forest) and aquatic ecosystems in a landscape unit. The investigated sites are representative of large parts of Northern Germany.

One of the remarkable features of the soils of the central area is the coarse, sandy texture in hill and slope positions, as well as the high contents of organic C in depressions. The systems under investigation covered four sites under different land use along a catena. Soil microbial biomass content, basal respiration rate to estimate of C mineralization, and arginine ammonification rate to indicate N mineralization were measured by laboratory experiments. Furthermore, *in situ* CO_2 and N_2O emission rate were estimated by field measurements. Due to high cost, *in situ* CO_2 measurements were concentrated on two arable fields with contrasting land use. The data were combined and their interdependency analyzed to draw interrelated conclusions about effects of land use and the significance of the microbial biomass content on C and N transformation and on the emission rate of greenhouse gases. We assumed that C and N mineralization, and formation of emitted CO_2 and N_2O, take place mainly in topsoil (A horizon).

Material and methods

Sites and soils

The research site is located 30 km south of Kiel in Schleswig-Holstein, Northern Germany (59°97′ N, 35°81′ E; Figure 2). The landscape, formed during the Pleistocene consists of morainic hills and lakes. The climate is influenced by the North Sea and the Baltic Sea. Long-term (1951 to 1980) mean annual rainfall is 697 mm and average annual air temperature is 8.1 °C according to the local meteorological stations Eutin and Plön. For 1992 and 1993, the course of temperature, precipitation (at the meteorological station in Ruhwinkel) and water tension for the field and grassland topsoils are presented in Figure 3.

Along a transect from a kames hill to the lake Belau, a west to east running agricultural catena was established with a sequence of arable fields and grasslands (Figure 2): A field under crop rotation (*Avena sativa* L. in 1990, *Beta vulgaris* L. in 1991, *Secale cereale* L. in 1992, *Zea mays* L. in 1993), regularly fertilized with organic manure and mineral N is called "field crop rotation"; a field under maize (*Zea mays* L.) monoculture regularly fertilized, mainly with cattle slurry and mineral N is called "field maize monoculture". The sloping dry grassland (Lolio-Cynosuretum typicum) regularly was fertilized with mineral N. The wet grassland at the bottom of the catena (Ranunculo-Alopecuretum geniculati, partly dominated by *Alopecurus pratensis* L.) received no fertilizer. The soils of the first three sites were predominantly sandy, the wet grassland soil had a high organic matter content. Further soil properties and the N fertilization rates between 1990 and 1993 are

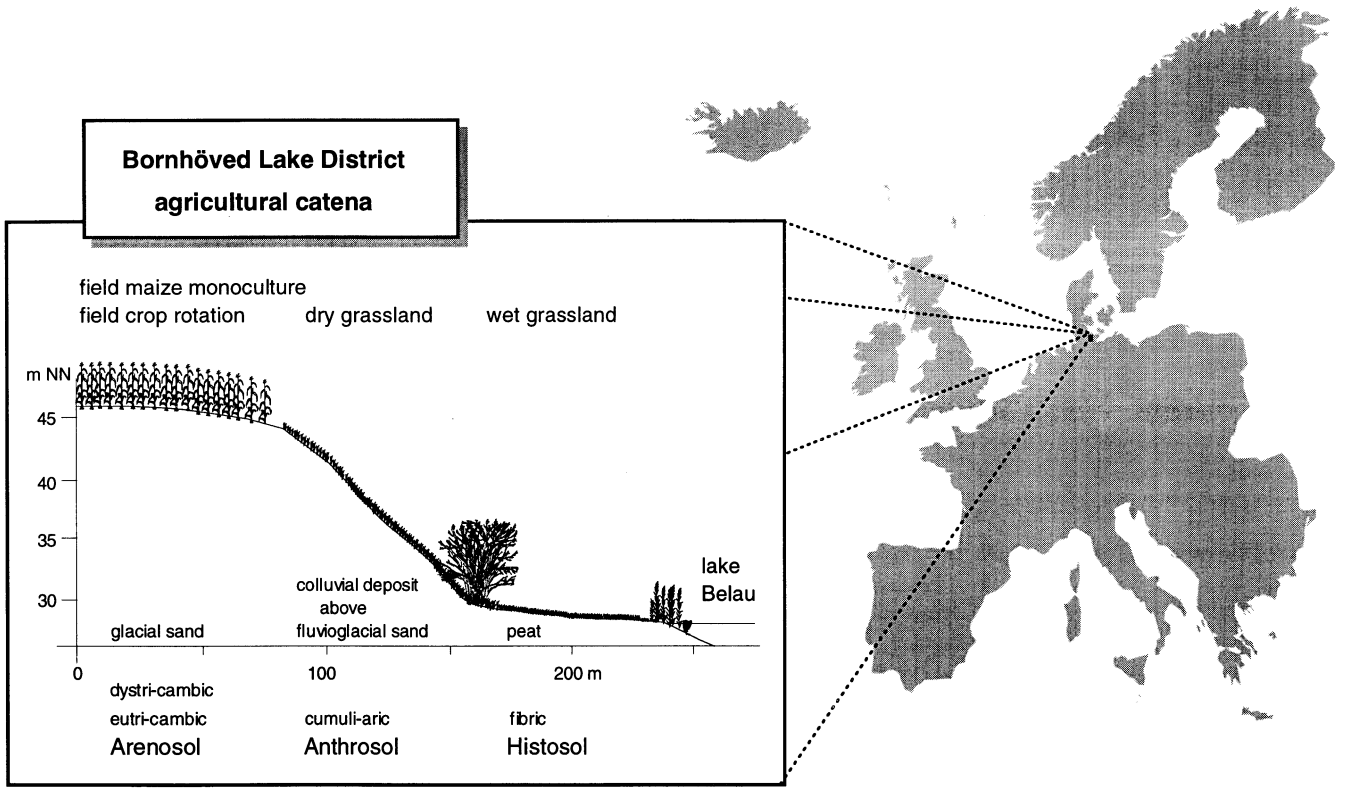

Figure 2. Location of the research site.

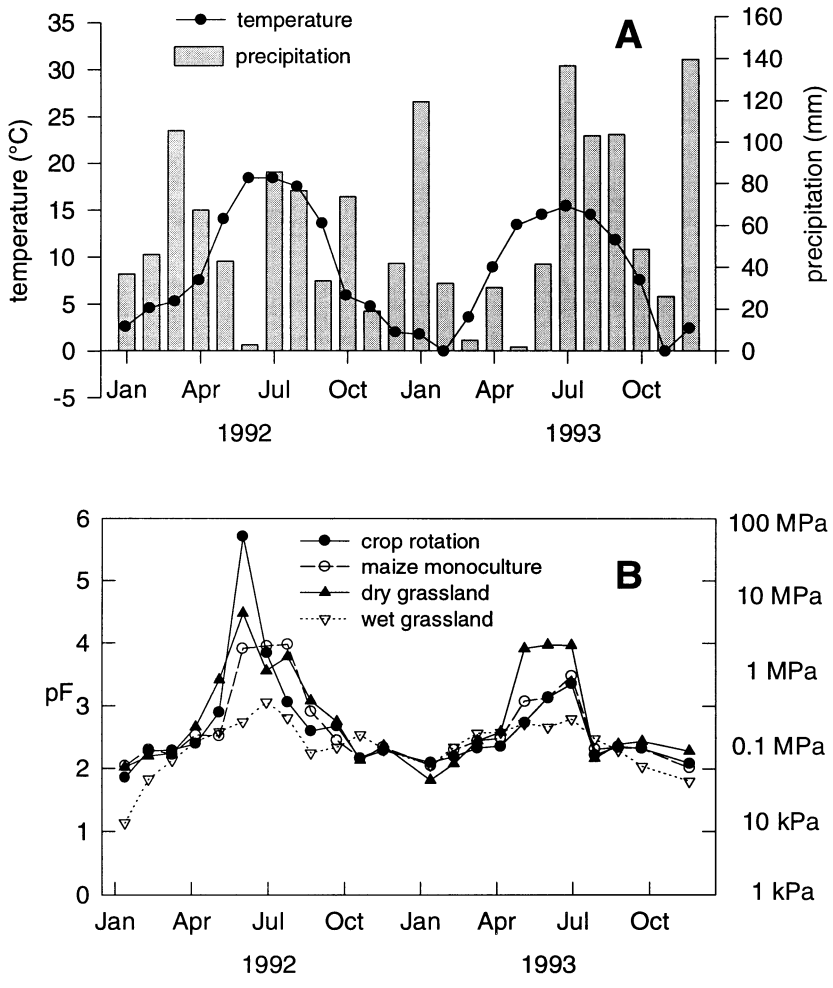

Figure 3. Temperature and precipitation (A) and matric tension (derived by combining gravimetric water contents and pF-WC-curves) and in field and grassland topsoils (B) of the Bornhöved Lake district in 1992 and 1993.

presented in Table 1. According to FAO (1988), soils were classified as eutri-cambic Arenosol (field crop rotation), dystri-cambic Arenosol (field maize monoculture), cumuli-aric Anthrosol (dry grassland) and fibric Histosol with a highly decomposed, dark grey to black topsoil (wet grassland).

Sampling

To estimate microbial biomass content and activity rates, bulk soils were sampled monthly from July 1992 to December 1993. At each site, about fifty

Table 1. Main properties of the topsoils (data achieved in spring 1992) and N fertilization rates between 1990 and 1993 at the agricultural catena in the Bornhöved Lake district, Northern Germany.

	Horizon	Depth (cm)	ph (H_2O)	C_{org} mg g^{-1} dry soil	N_{org} mg g^{-1} dry soil	C/N	Bulk density (g cm^{-3})	Organic + mineral N fertilization (kg ha^{-1}) 1990	1991	1992	1993
Field crop rotation	Ap	0–20	6.4	12.8	1.32	9.7	1.3	135[a)]+15	146[a)]+100	0+143	146[a)]+22
Field maize monoculture	Ap	0–20	5.4	12.1	1.01	11.9	1.3	144[b)]+95	343[c)]+123	156[b)]+109	291[d)]+109
Dry grassland	Ap	0–10	6.4	16.5	1.53	10.8	1.3	25[e)]+88	0+116	0+97	+78
Wet grassland	Ap	0–20	5.8	75.7	6.45	11.7	0.6	–	–	–	–

[a)] 30 (in 1990), 32.5 (in 1991), 31.5 (in 1993) t organic manure ha^{-1}; 4.5 kg N_t t^{-1} organic manure
[b)] 37 (in 1990) and 40 (in 1992) m^3 cattle slurry ha^{-1}; 3.9 kg N_t m^{-3} cattle slurry
[c)] 33 t organic manure ha^{-1} and 50 m^3 cattle slurry ha^{-1}
[d)] 30 t organic manure ha^{-1} and 40 m^3 cattle slurry ha^{-1}
[e)] 1.5 + 4 t organic manure ha^{-1}; 4.5 kg N_t t^{-1} organic manure

cores were taken with a Puerckhauer drill and mixed together. The samples were gently sieved and stored at 4 °C for at maximum of four weeks until analysis. The fraction <2 mm was used. To allow comparison with laboratory data, *in situ* CO_2 emission rates were determined at selected times in the absence of vegetation and when the soil moisture content was more than 40% of the water-holding capacity. CO_2 measurements were carried out in 1990 and 1992 using eight continuous-flow-boxes (area = 200 cm^2, volume = 2800 cm^3) equipped with Pt100 temperature sensors in parallel. Concentrations of CO_2 and Pt100 signals of each box were analysed for at least three times per hour. *In situ* N_2O emission rates were measured on 2 consecutive days every two weeks from July 1992 to December 1993 using six closed tubes (area = 707 cm^2, mean volume = ca. 16000 cm^3) and sampling after maximum enrichment of 1.5 hours.

To enhance the observations for statistical procedures, to draw general conclusions and to determine system-specific values, values of microbial biomass and activities as well as *in situ* N_2O emission rates were summarized for three investigation periods: II/92 (July to December 1992), I/93 (January to July 1993), II/93 (July to December 1993). The three intervals of six months were selected to highlight temporal variations particularly for the highly variable N_2O emissions. *In situ* CO_2 emission data were integrated for each day and related to the average temperature measured over the day at 10 cm soil depth.

Methods

Soil microbial biomass content was estimated by the fumigation-extraction-method (Vance et al. 1987) applying a conversion factor k_{EC} of 0.38 (k_{EC} = 1/2.64 = 0.38, where microbial biomass (C_{mic}) = (lysable) microbial C/k_{EC}). For a more detailed description see Dilly & Munch (1995). Basal respiration rate was determined at 22 °C with the apparatus described by Heinemeyer et al. (1989). Soil samples from crop rotation, maize monoculture and from the dry grassland were analyzed at a water content of 11 to 20% (g g^{-1} dry soil), equivalent to about 40 to 70% water-holding capacity (undisturbed soil). Soil samples from the wet grassland were analyzed at field water content. Samples were preconditioned for at least 3 days in the laboratory. The mean respiration rate after 15 to 24 h was calculated. Microbial metabolic quotient, qCO_2, was determined by dividing basal respiration [mg CO_2-C l^{-1} dry soil h^{-1}] by microbial C [g C_{mic} l^{-1} dry soil]. Arginine ammonification rate was measured according to Alef & Kleiner (1986) with minor modifications as described by Dilly & Munch (1995). Equipment for the *in situ* measurements included a continuous-flow-inverted-box system with infra-red gas analyser for the determination of CO_2 evolution (further information in Kutsch and

Kappen, this issue) and a 'closed-soil-cover'-system (Hutchinson & Mosier 1981) for N_2O emission. N_2O was analyzed according to Heinemeyer et al. (1991). Corresponding mineral N data (sum of 2 M KCl-extractable NH_4^+ and NO_3^-; shown in Figure 4) were measured in controls of ammonification method, NO_3^- determined with Cu reduction method according to Scharpf & Wehrmann (1976) with minor modifications. The results were expressed on soil volume or area basis.

Statistics

Each laboratory analysis (microbial biomass contents, basal respiration and arginine ammonification rates) was done in triplicate. *In situ* CO_2 data and the temperature signals were collected at least 3 times per hour per box. *In situ* N_2O emission rates were quantified in six replicates per sampling day. Since the assumption of normality test and equal variance was not fulfilled for the data, the nonparametric Kruskal-Wallis One Way Analysis of Variance on Ranks (all pairwise multiple comparison procedures; Student-Newman-Keuls Method or Dunn's Method) was applied to identify differences.

Results and discussion

Under suitable environmental conditions the extent of the turnover of organic compounds is mainly controlled by the quantity and activity of the microbial biomass (Martens 1995) and the community structure (Munch 1989). The mean microbial biomass content in the topsoils varied between 109 and 1230 mg C_{mic} l^{-1} soil (Figure 5) and significantly increased in the order, field maize monoculture $<$ field crop rotation $<$ dry grassland $<$ wet grassland. At a given site, microbial biomass content did not change between the three sampling periods. Martens (1995) reported that microbial biomass values between 0.2 and 1.0 mg C_{mic} g^{-1} soil are often found in agricultural soils. When expressed with the same units our microbial biomass values are lower than those reported by Martens (1995) under maize monoculture, slightly lower under crop rotation, in the same range under dry grassland, and higher under wet grassland. In contrast, after multiplying biomass values in Figure 5 with the thickness of soil horizon (Table 1) biomass data were within the range reported for different vegetation and soil types by Smith & Paul (1992). As soils differed in bulk density (Table 1), and in order to be enable to compare the habitats of the microbial community, laboratory values need to be related to volume or area for ecological comparisons. These results indicate that the driving force for biochemical transformations varied between the

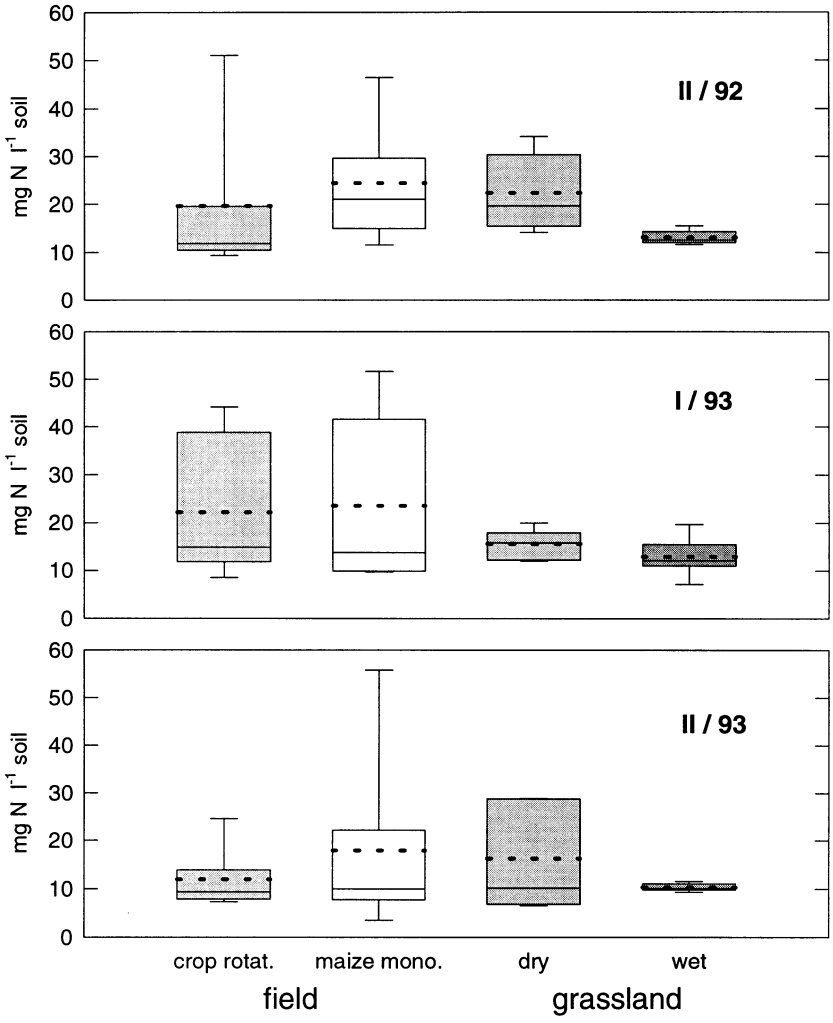

Figure 4. Mineral N (sum of NH_4^+ and NO_3^-) in field and grassland topsoils of the Bornhöved Lake district (for the sampling periods II/92, n = 18; I/93, n = 18; II/93, n = 15). Boxes encompass 25% and 75% quartiles, the central and the broken line represent the median and the mean, and bars extend to the 95% confidence limits.

agricultural soils being higher under grassland than under arable and lowest under monoculture.

Basal respiration estimates the intensity for C mineralization in soils and is controlled by endogenous C availability. It was lowest in the field samples, higher in dry grassland and highest in wet grassland soil for all three periods (Figure 6). In spite of significant differences in microbial biomass data, there

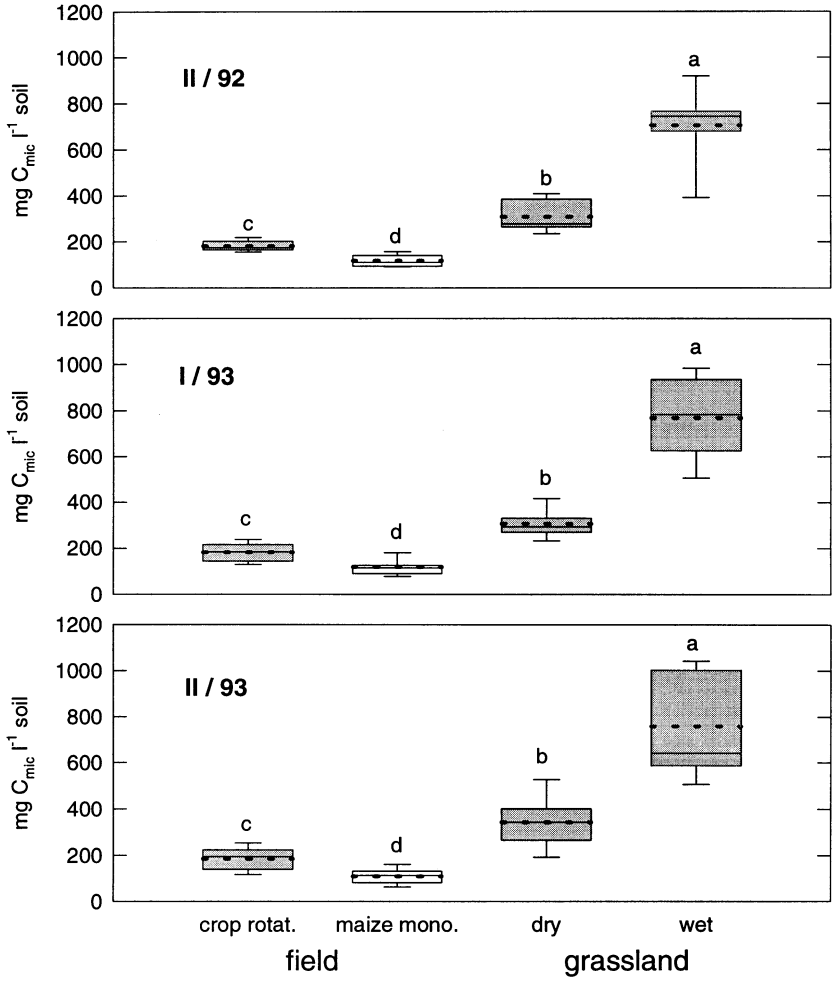

Figure 5. Microbial biomass content in field and grassland topsoils of the Bornhöved Lake district (for the sampling periods II/92, $n = 18$; I/93, $n = 18$; II/93, $n = 15$; different letters indicate significant differences applied the Student-Newman-Keuls Method, $p < 0.05$). Boxes encompass 25% and 75% quartiles, the central and the broken line represent the median and the mean, and bars extend to the 95% confidence limits.

was no significant difference in basal respiration between both arable soils, indicating that the C mineralization rates were similar in these soils. This conclusion from laboratory experiments was generally confirmed by field studies: For both arables, the *in situ* CO_2 emission rate from soil surface related to temperature was estimated in 1990 and 1992 at times with no vegetation and when soil respiration was not restricted by water deficiency (Figure 7). The regression equation for soil respiration against temperature

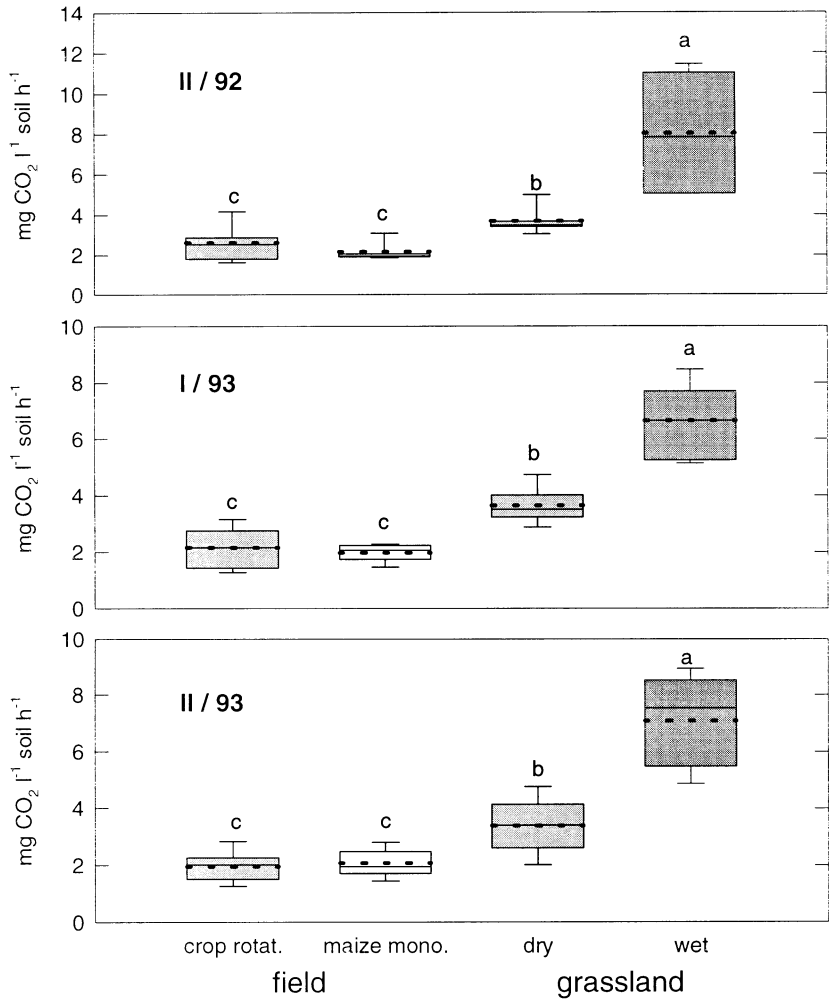

Figure 6. Basal respiration of field and grassland topsoils of the Bornhöved Lake district (for the sampling periods II/92, $n = 18$; I/93, $n = 18$; II/93, $n = 15$; different letters indicate significant differences applied Dunn's Method, $p < 0.05$). Boxes encompass 25% and 75% quartiles, the central and the broken line represent the median and the mean, and bars extend to the 95% confidence limits.

under crop rotation and under maize monoculture did not differ significantly (analysis of covariance, $p < 0.05$). The same result was obtained when 1992 data were excluded. Consequently, field and laboratory experiments indicate similar C mineralization rates for both arable soils.

The relatively high ratio of basal respiration or *in situ* CO_2 emission rate to microbial biomass content under maize monoculture indicates that

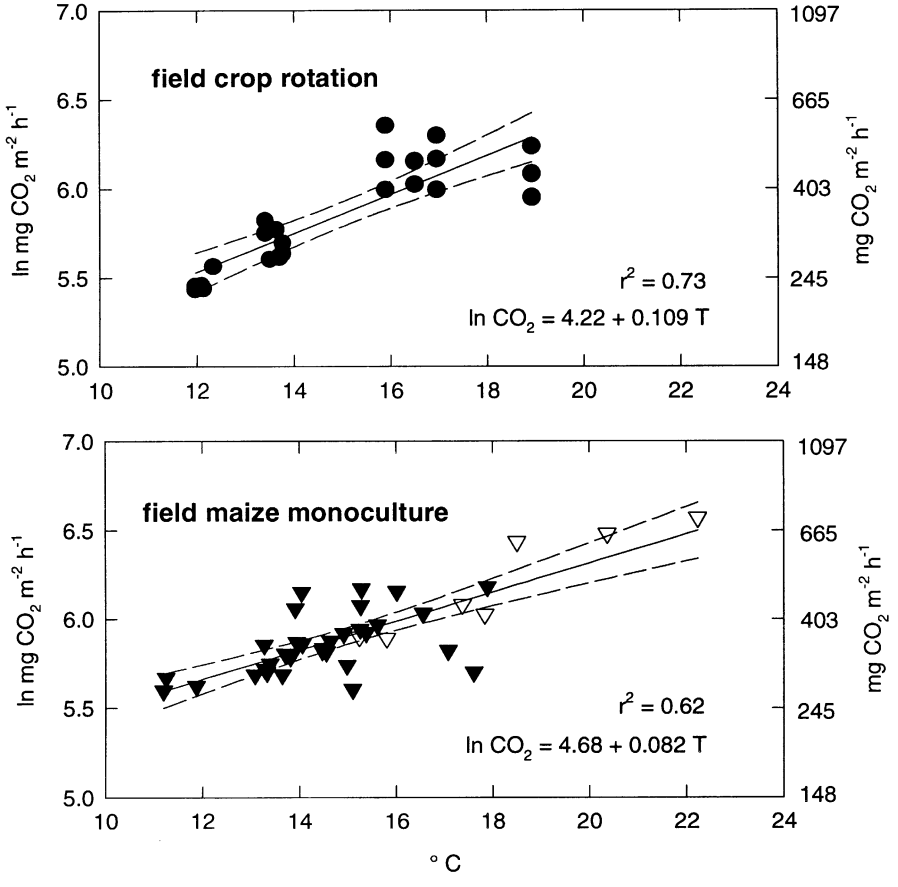

Figure 7. Mean daily *in situ* CO_2 emissions from field crop rotation and field maize monoculture soil – with no vegetation – related to the mean daily temperature (moisture more than 40% water-holding capacity) of the Bornhöved Lake district (field crop rotation: May to September 1990, n = 23; field maize monoculture: May to September 1990 and 1992, n = 39).

the microbial community under monoculture is less efficient in C conservation in comparison to that under crop rotation, dry grassland and wet grassland (Figure 8). This result confirms the earlier observation by Anderson & Domsch (1990), who showed that monoculture reduces the energetic utilization efficiency of the microbial community.

Under wet grassland, where microbial biomass values were high, arginine ammonification rates were relatively low and similar to those under maize monoculture. Arginine ammonification rates were higher under crop rotation and highest in the dry grassland soil. The ratio between the arginine ammonification rate and microbial biomass content (mg NH_4^+-N g^{-1} C_{mic} h^{-1}) was not significantly different among field crop rotation, field maize monoculture

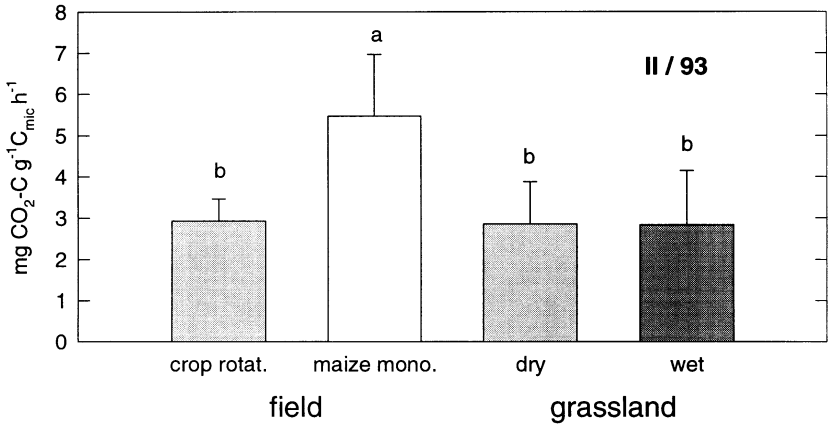

Figure 8. Metabolic quotient in field and grassland topsoils of the Bornhöved Lake district (for the sampling period II/93, $n = 15$; different letters indicate significant differences applied Dunn's Method, $p < 0.05$). Mean and standard deviation.

and dry grassland, but was less in wet grassland (Figure 9). In contrast to wet grassland, the three other sites were regularly fertilized with organic and mineral N (Table 1). Most, if not all heterotrophic bacteria release NH_4^+ when using nitrogen-rich C sources (Alef & Kleiner 1986). Dashman & Stotzky (1986) showed in studies with *Agrobacterium radiobacter* that arginine may be used as a carbon source in the presence of NH_4NO_3 and as a source of N if glucose is present. Low rates of arginine ammonification were also observed in the presence of easily degradable organic compounds (Forster et al. 1993). The low arginine ammonification rates in wet grassland soil, when measured as release of NH_4^+ (done here), may therefore indicate a low N status or a high N demand and efficiency of the microbial community.

N_2O is produced during denitrification, nitrification and dissimilatory formation of ammonia (Umarov 1990). Numerous genera of bacteria are able to denitrify (Umarov 1990) but they differ widely in specific N_2O production rate (Munch 1989). Fungi too may play a role in denitrification (Wainwright 1992). During nitrification, autotrophic nitrifiers and heterotrophic organisms are capable of producing N_2O. We assumed that N_2O emission rate is related to the quantity of microbial biomass (Drury et al. 1991). However, the comparison of the four sites demonstrated that the *in situ* N_2O emission rate (Figure 10) was not generally linked to the amount of microbial biomass. The wet grassland soil had significantly lower N_2O emission rates than the arable fields in spite of higher microbial biomass content, higher amount of substrate (Corg content) and seasonally low oxygen supply due to a high water table (low matric tension, see Figure 3). The low N_2O emission rates

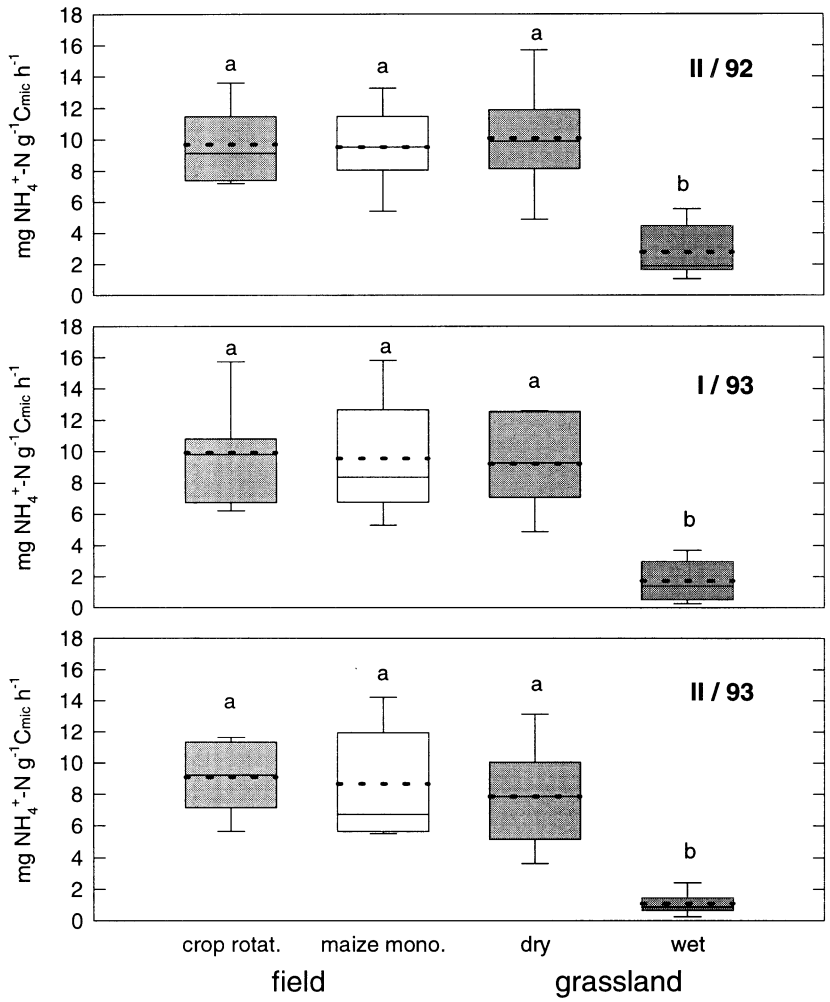

Figure 9. Arginine ammonification related to microbial biomass of field and grassland topsoils of the Bornhöved Lake district (for the sampling periods II/92, $n = 18$; I/93, $n = 18$; II/93, $n = 15$; different letters indicate significant differences applied Student-Newman-Keuls Method, $p < 0.05$). Boxes encompass 25% and 75% quartiles, the central and the broken line represent the median and the mean, and bars extend to the 95% confidence limits.

from wet grassland may be caused by diffusion barriers within the soil, further reduction of the nitrous compounds to N2 and the lower availability of N as reflected by the higher N efficiency of the microbial community. Comparing the three other sites, the highest N_2O emission rates were recorded in field crop rotation, particularly for the first period of 1993. Similar values were obtained for dry grassland in the sampling period II/93. *In situ* N_2O emission

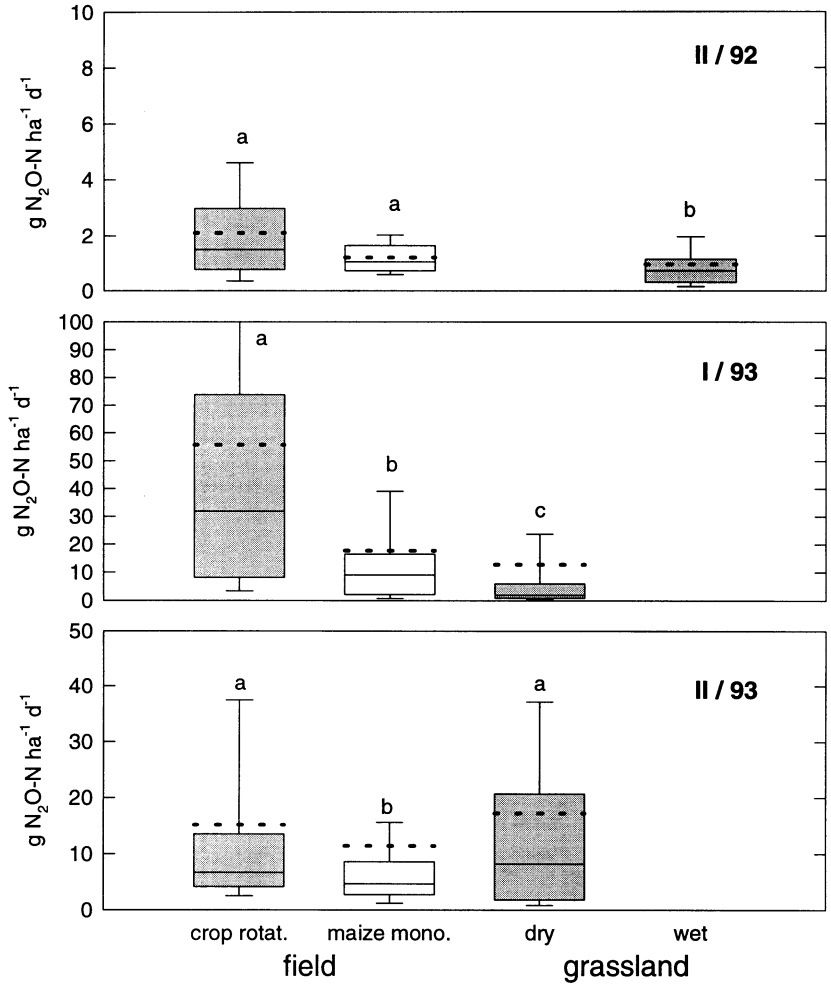

Figure 10. *In situ* N_2O emissions from field and grassland soils of the Bornhöved Lake district (for the sampling periods II/92, n = 95; I/93, n = 186; II/93, n = 173; different letters indicate significant differences applied Dunn's Method, $p < 0.05$). Boxes encompass 25% and 75% quartiles, the central and the broken line represent the median and the mean, and bars extend to the 95% confidence limits.

rate correlated to the microbial biomass content if only maize monoculture and crop rotation were considered. Indeed, Groffman & Tiedje (1989) as well as Drury et al. (1991) explained annual N denitrification and background denitrification by changes in microbial biomass content. However, the N_2O emission rates, the microbial respiration rates and the microbial biomass contents may not correspond to each other during the experiment (Fine et al. 1986; Kaiser 1994). The high N_2O emission rates in the field crop rotation

may have been caused by the high availability of N and the addition of organic manure (Mahimairaja et al. 1995).

Conclusions

Land use affected soil microbial biomass content and activity rates. In particular, microbial biomass content increased in the order, maize monoculture < crop rotation < grassland. Furthermore, microbial biomass content and C mineralization rate were correlated under crop-rotation and grassland, showing that the amount of microorganisms is proportional to C mineralization. By contrast, the metabolic quotient was significantly higher under maize monoculture, indicating that the microbial community is less efficient in C utilization. The N release was dependent on N status and the addition of easily degradable organic C compounds but not on the amount of microbial biomass. Conclusions concerning distinct C and N mineralization processes in the four agricultural soils were supported by combining laboratory and *in situ* data.

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