Soil microbial activities in Luvisols and Anthrosols during 9 years of region-typical tillage and fertilisation practices in northern Germany

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Abstract. In order to evaluate soil functions of contemporary agricultural management practices, the adjustment of microbial biomass and C and N mineralisation capacities was monitored during 9 years following the implementation of conventional and reduced tillage, and mineral N and pig slurry fertilisation systems. Soil microbial biomass content and microbial activities decreased continuously from initial values. The decrease was slowed by slurry application, compared to either no or mineral N fertilisation, and both slurry and mineral N application stimulated soil microbial activities in the long-term. There were no significant differences in microbiological characteristics between conventional and reduced tillage for the 0 to 30 cm soil depth but microbial biomass and activity were highest from 0 to 15 cm depth under reduced tillage. Changes in several microbial properties became evident when analysing the whole experiment of 9 years and the soil unit is also of importance as shown by higher microbial activity level in Anthrosols in comparison to Luvisols.

Introduction

Soil organisms respond to agricultural management practices (Beare et al. 1997) and, in turn, soil microbial activities affect soil productivity by mobilising and immobilising plant nutrients (e.g., Richards (1987) and Schlesinger (1997)). In accordance with current national programmes for sustainable agricultural management practices (e.g., Beare (1997)), the Collaborative Research Centre 192 of the German Research Foundation aimed at optimising food production with reference to both the economical and ecological outcomes in northern Germany. The following systems and management practices were implemented: Conventional tillage was compared with reduced tillage which avoids the mixing of the soil and, thus, reestablishes the stratification of the natural habitats. Reduced tillage is considered favourable, reducing organic matter losses and aiding nutrient retention (Alvarez et al. 1998; Beare 1997). Several inorganic N fertilisation levels and slurry application were compared in order to evaluate fertiliser use efficiency (Stevenson and

Cole 1999) that enhance the nutritional status for both soil organisms and plants and, thus, may increase soil quality (McCarty and Meisinger 1997).

The effects of agricultural management practices on abiotic soil characteristics have been extensively investigated in Europe in long-term experiments, e.g., in Rothamested, U.K. (Glendining et al. 1996), Bad Lauchstädt, Germany (Leinweber et al. 1994) and Dehérain, France (Houot and Chaussod 1995). The results of such experiments were used to model elemental fluxes at a landscape level (Smith et al. 1997). Effects of long-term tillage and nitrogen fertilisation (Salinas-Garcia et al. (1997a, 1997b)) and of cropping systems (Breland and Eltun 1999) on soil C and N dynamics have been shown. However, the adjustment of biotic soil components to modern agricultural management practices during the initial phase, e.g., 3 to 10 years, is poorly understood. Kandeler et al. (1999) followed the effect of conventional, reduced and minimal tillage on substrate-induced respiration, N mineralisation and the extracellular protease, xylanase and alkaline phosphatase activities in an Austrian haplic Chernozem for 8 years under wheat, pea, barley, sorghum, sugar beet and maize. They found high activities from 0 to 10 cm soil depth under reduced and minimal tillage due to the accumulation of organic matter. The extracellular xylanase activity reacted most rapidly within the first year either because of the high enzyme production by the microbial biomass or the accumulation of the particulate organic matter. In contrast, the microbial biomass and the intracellular N mineralisation were found to respond after approximately 4 years, probably because of their specific turnover rate. Emmerling et al. (2001) studied the response of microbial biomass and activities to agricultural de-intensification over a 10-year period between 0 to 15 and 15 to 30 cm soil depth and observed effects after approximately 5 years in central and western Germany. Emmerling et al. concluded that attention should be given to crop rotation, intercrops and cover crops, conservation tillage practices and organic matter input since they affect soil microbiological properties.

This paper investigates how soil microbial biomass and C and N mineralisation capacities adjust over time to different crop, fertilisation and tillage systems. We particularly addressed if several microbial characteristics respond similarly to modern agricultural management practices and also environmental factors. The experiment has been implemented on two soil units and soil data were analysed to 30 cm depth since microbial properties may be changed in specific layers but not significantly in the whole soil profile or ploughing depth (Needelman et al. 1999). The soil microbial biomass and the C and N mineralisation capacities were selected as they represent a labile pool and the current C and N mineralisation capacity respectively. Their ratios were used to evaluate changing abilities of the soil to transform different compound classes.

Reduced Tillage Conventional tillage NIL NIL 120N NIL NIL 120N Luvisols 120N 120N+SL 120N+SL 120N 120N+SL 120N+SL +SL 240N 240N 240N 240N +SL NIL 120N 120N NIL NIL NIL Anthrosols 120N 120N+SL 120N+SL 120N 120N+SL 120N+SL +SL 240N 240N 240N 240N +SL

Figure 1. Thirty-six plots selected containing the 2 soil units, 2 tillage systems and 5 fertilisation systems. No fertilisation NIL. Slurry application + SL. Application of 120 kg mineral N plus slurry 120N + SL. Application of 120 kg mineral N ha $^{-1}$ a $^{-1}$ 120N. Application of 160 to 240 kg mineral N 240N.

Materials and methods

Sites and soils

The experiment was carried out approximately 15 km west of Kiel in northern Germany (54°19′ N, 10°00′ E). The field was part of the experimental farm 'Hohenschulen' of the University of Kiel and located in a moraine landscape of loamy till with gentle slopes formed during the last glacial period. Before the experiment was started, the plot of approximately 3 ha was uniformly managed with a crop rotation (winter rapeseed, winter wheat and winter barley) and fertilisation practices typical for the region. The experiment started in autumn 1990 and the plot was divided over 3 fields with the 3 crops. Each field was again divided in 288 parcels containing 1 crop and 2 tillage systems with 6 fertiliser and 8 fungicide treatments, all in triplicate. From field no. 3, 36 typical plots of 36 m² each were selected containing the two soil units 'Luvisol' and 'Anthrosol' and also conventional tillage (CT) and reduced tillage (RT), each soil unit and tillage system in 18 replicates (Figure 1).

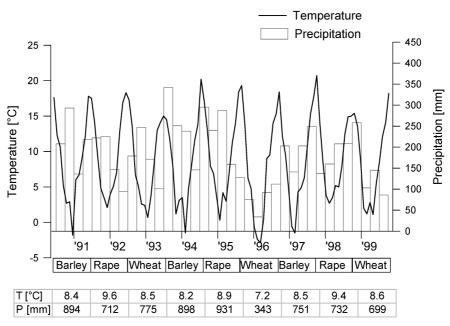


Figure 2. Temperature and precipitation at 'Hohenschulen' in northern Germany between 1991 and 1999; temperature averaged for each month and precipitation summarised for each 3 month period; below, temperature and precipitation correspondent to the growing season from August to July each year.

Hohenschulen has a moderate oceanic climate with a mean annual temperature of 8.3 °C and approximately 750 mm precipitation between 1974 and 1994 (Sieling 2000). The temperature and precipitation during the years of the investigation are shown in Figure 2. The long history of agricultural land use resulted in the development of two soil units 'Haplic Luvisol', partly eroded, and 'Cumulic Anthrosol', partly stagnic and/or gleyic, according to ISSS/ISRIC/FAO (1998). The Luvisols are sandy loams with clay migration, partly eroded and located mainly in upper slope position. In contrast, the Anthrosols have a similar texture but substantial amounts of humic material up to 80 cm soil depth and are located in a foot slope to valley position. The Luvisols can be distinguished from the Anthrosols by (i) dryness in summer and (ii) less wet conditions during spring. For more detailed information refer to Ziogas (1995).

The crop rotation 'winter rapeseed' (*Brassica napus* L.), 'winter wheat' (*Triticum aestivum* L.) and 'winter barley' (*Hordeum vulgare* L.) was repeated 3 times between 1990 and 1999 as illustrated in Figure 2 and is typical in northern Germany. At each harvest, straw was cut and remained in the field. Conventional tillage was ploughed to approximately 30 cm depth, and reduced tillage (RT) with a rotary cultivator at approximately 5 cm soil depth. Thus, the straw was distributed throughout the depth of 30 cm in CT or remained in the upper 5 cm in RT. All treatments received approximately 39 kg phosphorus, 100 to 116 kg potassium, 70 to 100 kg magnesium and 800 kg calcium ha⁻¹ year⁻¹. Five N fertilisation prac-

tices were selected (Figure 1): Control with no mineral N and no pig slurry application [Nil] with 8 replicates, 120 kg NH₄NO₃-N ha⁻¹ divided at equal rates and applied at three plant growth stages [120N] with 8 replicates, 160 to 240 kg NH₄NO₃-N ha⁻¹ divided at equal rates and applied at the same growth stages [240N] with 8 replicates, approximately 15 m³ pig slurry ha⁻¹ applied in spring and autumn [+ SL] with 4 replicates, and 120 kg NH₄NO₃-N ha⁻¹ plus the pig slurry [120N + SL] with 8 replicates. The slurry treatment represented a fertilisation of approximately 615 kg organic C, 145 kg N, 70 kg P, 54 kg K, 24 kg Mg and 68 kg Ca ha⁻¹ year⁻¹.

Soil was sampled from 0 to 15 and 15 to 30 cm depth from 2 replicated plots per treatment. The plots were sampled 26 times between 1991 and 1999 (Figure 4). Field-moist soil was sieved to pass a 5-mm screen (visible pieces of crop residues and roots were removed) and stored in a moist condition at 4 °C. Soil was stored at -21 °C when analyses could not be done within one month and later gently thawed in the fridge. The average bulk density was 1.29 and 1.41 Mg m⁻³ for the ploughed soil and 1.12 and 1.61 Mg m⁻³ for the soil under reduced tillage for the respective depths (Frey 1998).

Analyses

Throughout the experimental period, substrate-induced respiration 'SIR' and basal respiration 'BAS' were determined in the laboratory as microbial biomass estimate (Anderson and Domsch 1978) and current C mineralisation capacity (Anderson 1988) respectively. Before determining SIR and BAS, soil was preconditioned for at least 3 days at approximately 22 °C in the laboratory. Both were measured on the basis of the $\rm O_2$ uptake using a Sapromat respirometer (Fa. IBUK, Königsbronn, Germany). Microbial biomass was calculated using the conversion factor 29 mg C corresponding to 1 mg $\rm O_2$ h⁻¹, which is equivalent to 40.04 mg C for 1 ml $\rm CO_2$ h⁻¹ (Anderson and Domsch 1978) and for basal respiration a respiratory quotient of 1 was assumed. Soil moisture content corresponded to approximately 40 to 70% water-holding capacity. From 1994 onwards, microbial C and N was estimated using the fumigation-extraction method, 'FE-C' and 'FE-N' (Vance et al. 1987; Brookes et al. 1985).

The β -glucosidase activity 'GLU' (an 'extracellular' enzyme), and the arginine ammonification 'ARG' ('intracellular' since being related to active organisms), were analysed as indicators of the current C polymer degradation and N mineralisation capacities (Dilly 1997) respectively. The estimates may not be applicable to *in situ* conditions (Mary and Recous 1994). In these enzymatic assays, the phenol released from salicine and NH $_4^+$ released after arginine addition was estimated after 3 hours at 37 and 30 °C respectively (Hoffmann and Dedeken 1965; Alef and Kleiner 1986).

The microbial metabolic quotient $q\mathrm{CO}_2$ was calculated by dividing basal respiration by SIR-derived microbial C. The microbial quotient ($C_{\mathrm{mic}}/C_{\mathrm{org}}$ ratio) was calculated on the basis of SIR-derived microbial C and soil organic C content. The ratio between SIR and FE-C was considered as an indicator of the proportion of

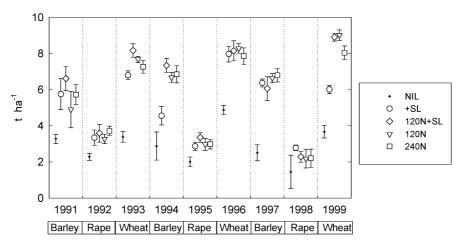


Figure 3. Grain yield of rapeseed, wheat and barley rotation under different fertilisation systems. No fertilisation NIL. Slurry application + SL. Application of 120 kg mineral N plus slurry 120N + SL. Application of 120 kg mineral N ha $^{-1}$ a $^{-1}$ 120N. Application of 160 to 240 kg mineral N 240N.

active microorganisms or the metabolic-responsive biomass (Dilly and Munch 1998). Finally, the ratios between (i) BAS and ARG, (ii) SIR and ARG and (iii) GLU and ARG were considered as indicators for the microbial C-to-N degradation capacity since BAS refers to the current C mineralisation potential based on endogenous soil C compounds, SIR to the current C mineralisation potential when available C is not restricting microbial metabolism, GLU to the enzymatic C polymer degradation potential and ARG to the current N mineralisation in the presence of N substrate.

Temperature, precipitation, soil water content, soil organic C content, pH value determined in 0.01 M CaCl₂ solution and straw production were considered as abiotic factors controlling the biotic characteristics. The temperature and precipitation data were taken from the database (http://www.rz.uni-kiel.de:8000/home/db/public/datenbank.html) and aggregated for 30 days before sampling. Antilogarithmic soil pH values were calculated as factor 'H⁺ concentration'. Since the amount of straw remaining in the field was estimated only a few times during the experiment and was then highly correlated with grain yield for rapeseed, wheat and barley (data not shown), grain yield was used for testing the effect of 'plant residues remaining on the field' on microbiological components. The grain yields during the years with reference to the fertilisation systems are given in Figure 3.

Statistics

Statistical analyses were performed using SigmaStat (Jandel Scientific, Erkrath, Germany). Simple linear regression analyses were considered to estimate parameters controlling soil microbiological characteristics. Spearman rank correlations were used to evaluate the interrelationships between microbiological characteristics

since normality test failed and constant variance test has not been passed in most cases (P < 0.05).

Results and discussion

Data range

Over the 9-year period, the mean microbial biomass content estimated with SIR was $329 \pm 6~\mu g$ C g^{-1} soil (\pm confidence limits at P < 0.05), the mean microbial respiration rate was $0.48 \pm 0.01~\mu g$ CO₂-C g^{-1} soil h^{-1} , mean arginine ammonification was $1.95 \pm 0.04~\mu g$ NH₄-N g^{-1} soil h^{-1} , and mean β -glucosidase activity was $93.5 \pm 1.2~\mu g$ phenol g^{-1} soil $3h^{-1}$. These values were respectively 13, 16, 44 and 34% higher than those found in an Arensols under crop rotation at the nearby Bornhöved Lake district which contained more sand (83%) and had lower pH value (Dilly et al. 1997; Dilly and Munch 1998). The average microbial biomass value in these loamy agricultural soils in northern Germany was lower than those under similar conditions. Joergensen (1995) and Emmerling et al. (2001) reported values of 345 μg C g^{-1} soil and 443 μg C g^{-1} soil in arable soils in central and western Germany respectively.

Changes in microbial biomass, basal respiration, arginine-ammonification and β -glucosidase activity

Microbial biomass, arginine ammonification and β -glucosidase activity significantly decreased over the 9-year investigation period (Figure 4, Table 1). According to the linear regression line (Normality test and Constant Variance Test were passed), the microbial biomass decreased by 21% from approximately 470 to 370 mg C_{mic} l⁻¹ soil. Such a decline was not expected since similar farming practices and environmental factors were likely present before the investigation began and, in addition, straw remained in the field to sustain soil fertility. The declining trends may be attributable to the constancy of farming practices, i.e., the regular application of mineral nitrogen and slurry either alone or in combination. We hypothesise that the practices before 1991 were more diverse. In addition, climatic changes with higher intra-annual variability of temperature and water regime may have contributed to the declining trend (Mamilov and Dilly 2002). The mean annual temperature was approximately 0.4 °C higher between 1990 and 1999 in comparison to the period from 1974 to 1994 while the annual precipitation stayed at 750 mm. Beside the fertilisation regime, the biomass and activity reduction might be induced by the modern tillage system (Ananyeva et al. 1999). Only basal respiration did not show such a significant decrease (p = 0.099)

The trend in microbiological characteristics suggests that microbial biomass, and the N degradation capacity declined during the 9 years after the establishment of the field trial. This is in contrast to the current C mineralisation capacity. Concur-

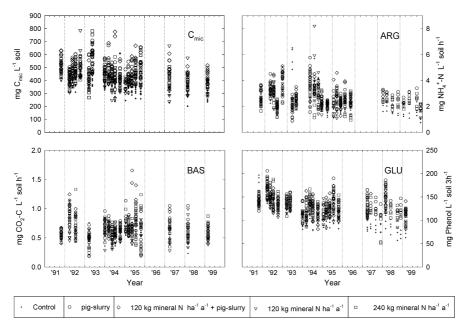


Figure 4. Temporal variation of microbial biomass (C_{mic}), arginine ammonification (ARG), basal respiration (BAS) and β -glucosidase activity (GLU) in agricultural systems at 'Hohenschulen' in northern Germany.

Table 1. Spearman rank order correlation coefficients (R) and probability (P) showing the trend of microbiological characteristics in agricultural soils during 9 years following the establishment region-typical tillage and fertilisation systems

	$C_{\rm mic}$	BAS	ARG	GLU	$q\mathrm{CO}_2$	BAS/ARG	SIR/ARG	GLU/ARG	SIR/FE
R	-0.31	-0.07	-0.31	-0.41	+ 0.12	+ 0.03	-0.08	-0.09	-0.26
P	< 0.001	0.099	< 0.001	< 0.001	0.002	0.569	0.023	0.015	0.046

Abbreviations: Microbial biomass C_{mic} . Basal respiration BAS.Substrate-induced respiration SIR. Arginine ammonification ARG. β -glucosidase activity GLU.

rently, organic C content declined from 19 to 17 mg $C_{\rm org}$ g^{-1} soil, the metabolically-responsive biomass decreased (Table 1).

Separating the data into 3 sets from 1991 to 1993, from 1994 to 1996 and from 1997 to 1999, each period referring to one crop rotation cycle of barley, rapeseed and wheat (Elsner 1994; Bode 1998; Frahm 2000), microbial biomass content and arginine ammonification rates decreased significantly (Table 2). The decrease in β -glucosidase activity was significant from the first to the second period. Basal respiration declined significantly from the second to the third period. Thus, microbial biomass and current N mineralisation decreased steadily. The C polymer degradation decreased more rapidly than the current C mineralisation capacity. Concurrently, organic matter content declined and soil pH value increased from 6.50 to 6.64 during the 9-year period (Table 3).

Table 2. Medians of microbiological characteristics in agricultural soils for three periods following the establishment of regional-typical agricultural systems; different letters indicate significant differences based on Kruskal-Wallis One Way Analysis of Variance on Ranks (P < 0.05)

	$C_{\rm mic}$	BAS	ARG	GLU	$q\mathrm{CO}_2$	BAS/ARG	SIR/ARG	GLU/ARG
1991–1993	458a	0.589ab	2.72a	143a	1.74b	0.33a	11a	75a
1994-1996	414b	0.638a	2.39b	119b	2.17a	0.30b	10a	70b
1997-1999	381c	0.575b	2.25c	118b	2.13a	0.36a	10a	72ab

Abbreviations: Microbial biomass C_{mic} [μg C g^{-1} soil]. Basal respiration BAS [μg CO₂-C g^{-1} soil h^{-1}]. Substrate-induced respiration SIR [μg CO₂-C g^{-1} soil h^{-1}]. Arginine ammonification ARG [μg NH₄+N g^{-1} soil h^{-1}]. β -glucosidase activity GLU [mg Phenol g^{-1} soil $3h^{-1}$].

Table 3. Median of temperature (T), soil water content (WC) and pH value for three periods following the establishment of regional-typical agricultural systems; different letters indicate significant differences based on Kruskal-Wallis One Way Analysis of Variance on Ranks (P < 0.05)

	T [°C]	WC [mg g^{-1} dry soil]	pH [CaCl ₂]
1991–1993	12.4a	21.3c	6.50c
1994-1996	9.6b	22.1b	6.55b
1997–1999	8.6b	24.1a	6.64a

The change in microbial characteristics was greatest for microbial biomass, basal respiration and arginine ammonification between 3 to 6 years and for β -glucosidase activity between 6 to 9 years. This may be related to the microbial adjustment to the management practices (Kandeler et al. 1999).

When data were separated with respect to the fertilisation system (Table 4), microbial biomass also declined for all management practices: the fertilisation treatments do not sustain the initial microbial biomass level. In contrast, the basal respiration decreased significantly only for the non-fertilised treatment suggesting that soil carbon is depleted in soils cultivated without fertilisation. Arginine ammonification decreased for the NIL and 120N + SL, and β -glucosidase activity for all treatments except the sole slurry application indicating that N mineralisation may be reduced in the absence of N fertilisation or with a small amount of mineral N and slurry fertilisation. The C polymer degradation potential may be stabilised by adding C sources.

In contrast to microbiological characteristics themselves, which are related to soil volume, the metabolic quotient that relates the C release to C incorporated in the biomass significantly and steadily increased (Table 1), particularly from the first to the second period (Table 2). This suggests that the soil microbiota conserved less C in the biomass as the experiment proceeded.

The ratios relating C and N degradation capacities (SIR/ARG and GLU/ARG) decreased continuously during the 9-year investigation period. For the 3 periods, the BAS/ARG ratio initially decreased followed by a later increase. Concurrently, the lowest GLU/ARG ratio was determined for the second period. The $q\mathrm{CO}_2$ and the ratios relating C and N degradation capacities suggest that mineral N fertilisa-

Table 4. Trends during the 9-year analysis of the microbiological characteristics in agricultural soils subjected to different fertilisation systems (P < 0.05); Positive +. Negative -. No change o

	C_{mic}	BAS	ARG	GLU	$q\mathrm{CO}_2$	BAS/ARG	SIR/ARG	GLU/ARG
NIL	_	(-)	(-)	_	O	(o)	(o)	0
+ SL	_	O	(o)	(o)	(o)	O	(o)	(o)
120N + SL	_	(o)	_	-	(o)	(o)	0	0
120N	(-)	O	(o)	-	+	(o)	-*	_
240N	(-)	o	(o)	-	O	(o)	(o)	(o)

^{*} p=0.069 The signs in brackets indicate that the Normality Test and/or Constant Variance Test failed. Abbreviations: Microbial biomass $C_{\rm mic}$. Basal respiration BAS. Arginine ammonification ARG. β -glucosidase activity GLU. Substrate-induced respiration SIR. No fertilisation NIL. Slurry application + SL. 120 kg mineral N per ha and year plus slurry 120N + SL. 120 kg N per ha and year 120N. 240 kg N per ha and year 240N.

tion favoured microbial communities that are inefficient in C-use and stimulated N liberation in relation to C mineralisation.

Seasonal changes and climatic factors controlling microbiological characteristics

Seasonal variation in climatic factors was found to affect microbial biomass, microbial activities and ratios between microbiological characteristics (Table 5). The correlation coefficients, however, were generally low indicating that the extent of seasonal dynamics in climatic factors were not well reflected in microbial biomass and C and N degradation capacities. Kaiser and Heinemeyer (1993) observed greater changes in soil microbial biomass (estimated by SIR on the basis of CO₂ evolution) of approximately 30% in agricultural loamy-silt soils in northern Germany. They found the highest values under barley in early summer for 0 to 10 cm soil depth. Here, changes in microbial biomass at 0 to 30 cm soil depth were small. Microbial biomass content in arable, grassland and forest soils worldwide has been reported to have maximum values either in spring and autumn, summer and autumn, summer, autumn or according to the root development and lowest values either in winter and early spring or during rain seasons (Friedel and Lützow von 1998). The ratio between maximum und minimum values ranged between 1.17 and 6.67 under different land use indicating that microbial growth and activities respond to the emergent environmental factors. No apparent seasonal dynamics were observed in our soils.

Soil microbial activities weakly correlated with climatic factors as they were estimated under optimised rather than under *in situ* conditions. Figure 4 demonstrates that seasonal variations concurred occasionally only with environmental factors. High temperature favoured microbial biomass and β -glucosidase activity, active microorganisms within the communities (SIR/FE ratio) and the prevalence of C over N mineralisation potential as indicated by the BAS/ARG ratio. The temperature before the samplings was higher during the first part of investigations (Table 3), and, thus, contributed to the declining trend observed for microbial biomass during

Table 5. Spearman rank order correlation coefficients between soil microbiological characteristics and environmental factors during the 9-year analysis of agricultural soils (P < 0.05)

	$C_{\rm mic}$	BAS	ARG	GLU	$q\mathrm{CO}_2$	SIR/FE				
							ARG	ARG	FE	ARG
T [°C]	0.22	-0.08	-0.11	0.22	-0.18	0.29	0.20		0.35	
WC [mg g ⁻¹ dry soil]	-0.14			-0.24					-0.35	
Precipitation [mm]		0.15	0.17	-0.24			0.17		0.25	0.16
Anti-log pH	0.16	0.03	0.15	0.35	-0.12					0.08
Plant residues [Yield]	0.25	-0.12	0.12		-0.23			0.12		

Abbreviations: Microbial C derived by fumigation-extraction FE. Other see Table 4.

the 9 years (Tables 1 and 4). However, the basal respiration, arginine ammonification and $q\mathrm{CO}_2$ were even higher during periods with low temperature. High $q\mathrm{CO}_2$ occurred in the study of Dilly et al. (2001) also at times with low temperature indicating that the metabolic potential in the microbial biomass is higher under winter conditions.

Soil water content seems to have a negative impact on several microbial functions although precipitation seems to have stimulated several microbiological functions (Table 5). The data reveals that high $\rm H^+$ concentration occurred with high microbial biomass and activity values and low $\rm qCO_2$ values. The observation of the $\rm qCO_2$ concurs to calculations of Stork and Dilly (1998) for numerous plots in a beech forest but is at variance with the study of Anderson and Domsch (1993), who hypothesised that low pH values (high $\rm H^+$ concentrations) induced high $\rm qCO_2$ values (low C use efficiency) in a wide range of soils. Here, high microbial colonisation was related to high C use efficiency and $\rm H^+$ concentration which may be the site-specific response of soil microbial communities (Dilly and Munch 1998).

Large quantities of plant residues increased microbial biomass contents (Table 5). This concurs with studies of Pfender et al. (1996) proposing that microbial biomass can be regarded as 'biomass of decomposing communities' in plant residues. In contrast, basal respiration was lower in the presence of large quantities of plant residues. Microbial communities that are C-use efficient seem to have dominated the microbial biomass, which is indicated by the negative correlation between plant residues and qCO_2 values.

Long- and short-term effect of fertilisation

Fertilisation increased grain (Sieling 2000) and straw yields and both mineral N and slurry fertilisation increased microbial biomass. When considering the whole investigation period (Figure 5), microbial biomass increased in the order, NIL < 120N < 240N < + SL, 120N + SL. Slurry application predominantly stimulated biomass production. In contrast, arginine ammonification was not significantly modified by the treatments and may even be repressed by slurry application. Thus, slurry application reduced the arginine ammonification through the presence of available C (Alef and Kleiner 1986) and increased N immobilisation capacity (Dilly

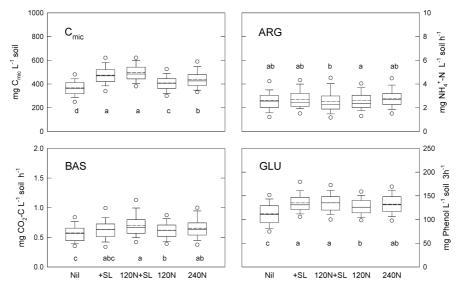


Figure 5. Effect of mineral N fertilisation [kg ha⁻¹ year⁻¹] and slurry application (SL) on microbial biomass ($C_{\rm mic}$), arginine ammonification (ARG), basal respiration (BAS) and β-glucosidase activity (GLU); different letters indicate significant differences using Kruskal-Wallis One Way Analysis of Variance on Ranks, P < 0.05; boxes enclose 25% and 75% quartiles, the central and the broken lines represent the median and the mean respectively, bars extend to the 90% confidence limits and circles show the 5% and 95% percentiles. Abbreviations: see Figure 1.

1997). Fertilisation also increased basal respiration. The 120N + SL treatment stimulated this current C mineralisation capacity more than the 120N treatment. All fertilisers, but predominately the slurry application, increased the β -glucosidase activity and, thus, this C polymer degradation capacity.

Slurry application increased microbial biomass content already during the first year (Figure 6). In contrast, Fließbach and Mäder (1997) found no significant differences between manure and mineral fertilisers in conventional systems after 18 years under more continental conditions in Switzerland, with samplings in spring and autumn. In our study, mineral N application did not significantly favour microbial biomass during the first year or the first 3 years (Elsner 1994). In addition, arginine ammonification and basal respiration were only slightly modified by fertilisation during the first years. A significant trend could only be shown as the experiment proceeded presumably since mineral N fertilisation favoured microbial growth and activity through the production of greater amounts of plant residues and root exudates.

Elsner (1994) reported that β -glucosidase activity was little changed in the first 3-year period and concluded that this enzyme activity may not be sensitive to management. The evaluation over the complete experiment (Figure 5), however, showed that this C polymer degradation capacity was significantly modified in the long term.

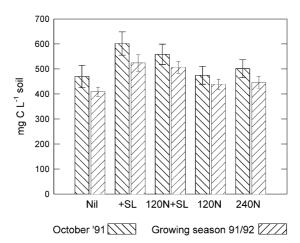


Figure 6. Short-term effect of mineral N fertilisation [kg ha⁻¹ year⁻¹] and slurry application (SL) on microbial C; bars extend to the 95% confidence limits. Abbreviations: see Figure 1.

Crop

Soil microbial biomass and activities were significantly affected by the crop (Figure 7). Soil below rapeseed showed the lowest microbial biomass content, which corresponds to results of Kaiser and Heinemeyer (1993) for sugar beet. Thus, both rapeseed and sugar beet, termed to be humus-degrading crops or 'hoe-crops' (crops under chop tillage), reduced microbial colonisation of the soil.

Highest microbial biomass content was seen under wheat, and highest arginine ammonification under barley. In contrast, soil basal respiration and β -glucosidase activity was stimulated under rapeseed. Rapeseed also stimulated feeding activity of soil fauna (Bode 1998). The microbiological ratios (Figure 8) indicate that soil microbial communities under rapeseed were less efficient in C-use and exhibited a lower C-to-N-degradation capacity. Higher root density was estimated under rapeseed in comparison to cereals in Hohenschulen (Max pers. comm.) which may have stimulated microbial activities through root exudation and favourable root litter quality (Grayston et al. 1996; Trinsoutrot et al. 2000). In contrast, soil microbial communities under barley were more efficient in C-use and showed the highest C-to-N-degradation capacity.

With reference to the individual years (Figure 3), the soil under rapeseed gave highest $C_{\rm mic}$, arginine ammonification, β -glucosidase activity and SIR/ARG ratio in 1992 in the year with highest average temperature (Figure 2), highest qCO $_2$ and BAS/ARG ratio in 1995 with lowest temperature, lowest GLU/ARG ratio in 1995 and lowest basal respiration in 1998. The SIR/ARG and BAS/ARG ratios indicate the higher ability to mineralise C relative to N in 1992 and in 1995 respectively. In contrast, the GLU/ARG ratio showed lowest ability to degrade C polymers relative to mineralise N in 1995. Concurrent with low basal respiration, lowest yield and straw production occurred for rapeseed in 1998 (Sieling 2000). Under wheat, the

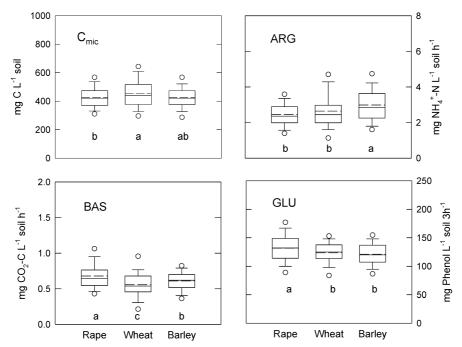


Figure 7. Crop-dependent modification in soil microbial biomass ($C_{\rm mic}$), arginine ammonification (ARG), basal respiration (BAS) and β-glucosidase activity (GLU); different letters indicate significant differences when applying Kruskal-Wallis One Way Analysis of Variance on Ranks, P < 0.05; boxes encompass 25% and 75% quartiles, the central and the broken lines represent the median and the mean respectively, bars extend to the 90% confidence limits and circles show the 5% and 95% percentiles.

highest arginine ammonification and lowest basal respiration and $q\mathrm{CO}_2$ occurred in 1993, β -glucosidase activity decreased steadily, lowest $\mathrm{C}_{\mathrm{mic}}$ values and highest BAS/ARG ratio were found in 1999, SIR/ARG values decreased from 1993 to 1996 and then increased again. The soil under barley showed lower arginine ammonification and higher $q\mathrm{CO}_2$ and BAS/ARG ratio in 1994 in comparison to 1997. Surprisingly, the cold and dry year 1996 (Figure 2) seems not to have had a negative impact on either wheat yield or microbiological characteristics and, thus, has not limited plant growth or interrelated microbial processes.

Conventional vs. reduced tillage

Many investigations showed stimulated microbial activities under RT (e.g., Beare et al. (1997) and Stenberg et al. (2000)). Similar effects were found for 0 to 15 cm soil depth in Figure 9. In detail, microbial biomass and respiration were highest under RT from 0 to 15 cm soil depth. In CT systems, the microbiological characteristics were higher in the lower soil layer from 15 to 30 cm soil depth indicating that ploughing buries substrates for soil microorganisms. Arginine ammonification and β -glucosidase activity, however, did not show highest values in 0 to 15 cm

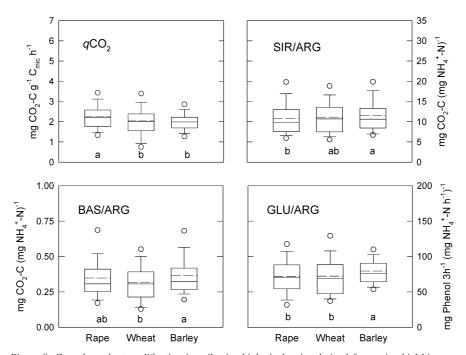


Figure 8. Crop-dependent modification in soil microbiological ratios derived from microbial biomass ($C_{\rm mic}$), arginine ammonification (ARG), basal respiration (BAS) and β -glucosidase activity (GLU) in agricultural systems at 'Hohenschulen' in northern Germany; different letters indicate significant differences when applying Kruskal-Wallis One Way Analysis of Variance on Ranks, P < 0.05; boxes encompass 25% and 75% quartiles, the central and the broken lines represent the median and the mean respectively, bars extend to the 90% confidence limits and circles show the 5th and 95th percentiles.

under RT indicating that the current N mineralisation potential and the C polymer degradation were relatively reduced probably, in conjunction with higher production and degradation of the enzymes (Kandeler et al. 1999). Furthermore, the presence of available C reduced arginine ammonification most likely due to higher microbial N requirement (Dilly 1997).

In the RT systems, the high microbial biomass and activities were associated with higher aggregate stability only at 0–15 cm, and particularly when organic fertilisers had been used (Victorino 1996). In accord with this observation, the lower tillage depth improved aggregate stability in studies of Stenberg et al. (2000). Thus, microbial activities physically stabilise soil structure at the surface reducing erosion potential and runoff. In contrast, the comparison between CT and RT at the ploughing depth of 30 cm showed no significant effects on microbial biomass (based on SIR), basal respiration, arginine ammonification and β -glucosidase activity (data not shown). This concurred with previous calculations for selected data from this experiment by Bode (1998) and Seese and Blume (1999).

It might be concluded that straw distribution in the plough layer in CT and residues remaining close to the surface in RT affected microbiological characteristics

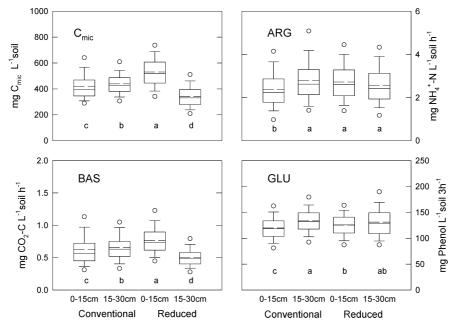


Figure 9. Soil microbial biomass ($C_{\rm mic}$), arginine ammonification (ARG), basal respiration (BAS) and β -glucosidase activity (GLU) under conventional and reduced tillage at two soil depths; different letters indicate significant differences when applying Kruskal-Wallis One Way Analysis of Variance on Ranks, P < 0.05; boxes encompass 25% and 75% quartiles, the central and the broken lines represent the median and the mean respectively, bars extend to the 90% confidence limits and circles show the 5% and 95% percentiles.

and, thus, microbial activities similarly over 30 cm soil depth. More active microorganisms in the total microbial biomass may have occurred in deeper soil layers in ploughed CT (Alvarez et al. 1998). Under RT, arginine ammonification showed higher values at P < 0.067 and a lower BAS/ARG ratio at P < 0.073. This suggests a small increase of N mobilisation in the presence of the readily available arginine and a lower C-to-N-mineralisation capacity.

For the 3 barley-rapeseed-wheat cycles, microbial biomass content and β -glucosidase activity increased significantly in RT system during the first period from 1991 to 1993 (data not shown). The BAS/ARG ratio decreased under RT suggesting a stimulated N mineralisation in relation to C. No significant modifications were determined between 1994 and 1996 and, therefore, the increase in biomass and the modified microbial metabolism seems not to persist over 3 years. Between 1997 and 1999, only basal respiration and $q\text{CO}_2$ were enhanced in RT suggesting that the soil microbiota showed higher current C mineralisation capacity and inefficiency in C-use, probably as the result of some differences between CT and RT during the first period. The main beneficial effect of RT vs. CT seems, therefore, to improve soil properties close to the surface (Frede et al. 1994).

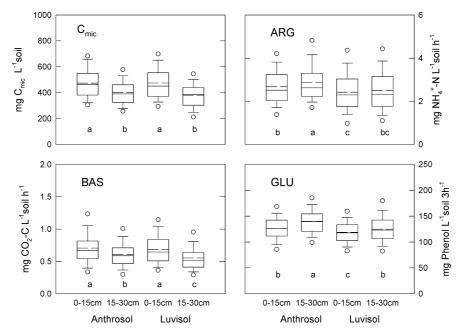


Figure 10. Soil-type dependent modification in soil microbial biomass ($C_{\rm mic}$), arginine ammonification (ARG), basal respiration (BAS) and β -glucosidase activity (GLU); different letters indicate significant differences when applying Kruskal-Wallis One Way Analysis of Variance on Ranks, P < 0.05; boxes encompass 25% and 75% quartiles, the central and the broken lines represent the median and the mean respectively, bars extend to the 90% confidence limits and circles show the 5% and 95% percentiles.

Soil unit

Microbial biomass contents were higher from 0 to 15 cm soil depth than from 15 to 30 cm in both Anthrosols and Luvisols (Figure 10). Basal respiration responded similarly, however, lowest values occurred from 15 to 30 cm soil depth in Luvisols indicating a greater decline in current C mineralisation capacity in this soil type. In contrast, highest arginine ammonification and β -glucosidase activity was determined for 15 to 30 cm in Anthrosols, and lowest arginine ammonification and β -glucosidase activity from 0 to 15 cm in Luvisols.

For 0 to 30 cm soil depth, microbial activities (basal respiration, arginine ammonification, β -glucosidase activity and qCO₂) were higher in Anthrosols then in Luvisols (data not shown). This can partially be attributed to greater organic matter (P < 0.054), total N and P contents and less clay in these well-aerated Anthrosols (Elsner 1994). Since the Anthrosols were cumulic and Luvisols partly eroded, the differences between the two soil types are more apparent when considering 100 cm soil depth. The microbial biomass estimated by SIR did not show this trend. However, the data available from FE (which were determined from 1995 onwards) also gave higher microbial C and N content for Anthrosols (data not shown). In addition, microbial C/N ratio was reduced probably following the higher microbial ac-

tivities. During the first 3 years, the Anthrosols showed a higher organic C content but did not exhibit more favourable conditions for the soil organisms than the Luvisols (Elsner 1994; Bode 1998). Significant effects became evident only when analysing the whole experiment of 9 years.

Conclusions

Over the 9-year experimental period with constant agricultural management and fertilisation treatments, the basal respiration, a measure of current C mineralisation capacity, was not significantly modified. In contrast, soil microbial biomass content and capacities for intracellular N mineralisation and extracellular C polymer degradation declined. This concurred with the decline of organic C and the increase in pH value. The decline was not expected since traditional farming practices were likely similar to the tested treatments. We must conclude that the trends may be attributed to current agricultural management practices, e.g., less favourable practices and increased liming, and probably also to environmental changes, e.g., increase in temperature by 0.4 °C. The current management with plant residues remaining on the field seemed not to stabilise the initial $C_{\rm org}$ and microbial levels.

The ratios between the microbial properties showed trends towards a less metabolically responsive microbial biomass and a decreasing relative capacity of the soil to mineralise C in comparison to N. Simultaneous slurry and mineral N application increased most microbiological characteristics in comparison to no or only mineral N fertilisation. However, the current N mineralisation capacity as determined using arginine ammonification was reduced by the slurry application. Without N fertilisation, the lower amount of plant residues remaining on the field seems to decrease soil microbiological activities. Microbial biomass content and metabolic responsive biomass was positively correlated to temperature, while basal respiration and $q\mathrm{CO}_2$ were negatively correlated.

Wheat and barley were found to favour microbial biomass, and rapeseed to stimulate microbial activities. Reduced tillage promoted microbial activities close to the soil surface but did not significantly modify the microbiological characteristics and, thus, elemental cycling for 0 to 30 cm soil depth. Specific nutritional and environmental factors existing in Anthrosols stimulated soil microorganisms in comparison to those in Luvisols.

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