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Effects of nitrogen and intensive mixing on decomposition of ^{14}C-labelled maize (*Zea mays* L.) residue in soils of different land use types

Haiqing Chen [a,*], Norbert Billen [a], Karl Stahr [a], Yakov Kuzyakov [b]

[a] *Institute of Soil Science and Land Evaluation, University of Hohenheim, Emil-Wolff-Str. 27, D-70593 Stuttgart, Germany*
[b] *Department of Agroecosystem Research, University of Bayreuth, Universitätsstr. 30, D-95440 Bayreuth, Germany*

Abstract

This study investigated the effects of mineral-N addition and intensive mixing (analogous to disturbance by plowing) on decomposition of ^{14}C-labelled maize (*Zea mays* L.) residue and soil organic matter (SOM). Soils were collected from the upper 5 cm of three land use types at Edelweiler, Germany: plow tillage (PT), reduced tillage (RT), and grassland (GL). Soils were incubated for 112 days at 20 °C, with or without ^{14}C-labelled maize residue (4 g DM kg^{-1} soil), with or without nitrogen (100 mg N kg^{-1} soil as NH_4NO_3) and with or without intensive mixing.

The effect of mineral-N on maize residue decomposition differed depending on the stage of decomposition and land use type. Nitrogen accelerated residue decomposition rates in the first 5 days in RT and GL soils, but not in PT soil, and decreased residue decomposition rate in all three land use types after 11 days. At the end of the incubation, N suppressed $^{14}CO_2$ efflux in RT and PT soils, but not in GL soil. Mineral-N did not increase SOM decomposition independently on the land use types.

Intensive mixing stimulated decomposition of both plant residue and SOM in all three land use types. However, effects were smaller in GL soil than in RT or PT soil, presumably because stronger soil aggregates in GL would have been less affected by mixing and allowed greater protection of SOM and plant residue against decomposition.
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1. Introduction

Soil organic carbon (SOC) is the largest terrestrial C pool; it is nearly three times the size of the biotic pool and double the size of the atmospheric pool (Post et al., 1990; Lal, 2004). Therefore, understanding the key factors and processes controlling SOC storage and turnover is critical because any changes in SOC could significantly affect the level of CO_2 in the atmosphere.

Inputs of mineral-N to agricultural and natural ecosystems have increased as a consequence of anthropogenic activities (Vitousek et al., 1997; Moran et al., 2005). Whether increased N-inputs can enhance SOC sequestration became an issue of debate. Nitrogen additions usually result in an increase in soil organic matter (SOM) content due to greater plant production leading to higher C input into the soil (Townsend et al., 1996; Christensen and Johnston, 1997; Nadelhoffer et al., 1999). However, it is argued that N additions do not always increase C storage (Cao and Woodward, 1998; Paul et al., 2004), since increased N availability can also stimulate microbial decomposition and C turnover

* Corresponding author. Tel.: +49 711 45923668;
fax: +49 711 45923117.

(Körner and Arnone, 1992). These discrepant results observed in various studies can be explained by contrasting effects of N on decomposition of plant litter and SOM since their availability for microbial decomposition and C-to-N ratio are strongly different. Foereid et al. (2004) found N addition did not affect C mineralization of *Miscanthus* residue. Conde et al. (2005) reported increased maize (*Zea mays* L.) residue decomposition in saline alkaline soils by inorganic N addition. A positive effect of N on decomposition of plant residues in the initial decomposition phase and a negative effect in later stages have been reported in some studies (Fog, 1988; Berg and Matzner, 1997; Henriksen and Breland, 1999; Wang et al., 2004; Moran et al., 2005). Highly variable effects of addition of N on decomposition of SOM are described in various studies (Jenkinson and Rayne, 1985; Berg and Matzner, 1997; Henriksen and Breland, 1999; Saiya-Cork et al., 2002; Neff et al., 2002; Foereid et al., 2004). Therefore, such uncertainties in the relationships between mineral-N and decomposition of plant litter and SOM call for further investigations. Therefore, the first aim of our study was to help clarify the effects of mineral-N addition on the decomposition of plant residues and SOM.

Land use and management practices affect C dynamics and storage in soil. Many studies have indicated that the use of conservation tillage (e.g. reduced or no-tillage practices) and conversion of cropland to grassland increase SOC contents (reviewed by Smith et al., 2000; Six et al., 2002; West and Post, 2002). Tillage promotes SOM decomposition by disrupting soil aggregates and exposing physically protected organic material (Paustian et al., 2000; Six et al., 2000), as well as increasing aeration, which enhances C mineralization and CO_2 fluxes to the atmosphere (Elliott, 1986; Reicosky et al., 1995). Thus, soil disturbance caused by tillage practices is an important factor affecting the decomposition rates of SOM and litter. Some reports have shown that intensive disturbance of soil samples increased CO_2 production in traditional incubation experiments (Sorensen, 1979; Powlson, 1980; Hassink et al., 1993). Bell et al. (2003) reported that disturbance resulted in slight but no significant increase in mineralization of ^{14}C-labelled wheat (*Triticum aestivum* L.) straw. Alvarez et al. (1995) found that the intensity of ^{14}C-labelled wheat decomposition was similar in disturbed and undisturbed soil. Thus, the magnitude of the response of residue and SOM to disturbance remains unclear. So, the second aim of our study was to determine the effects of disturbance on the decomposition of plant residues and SOM in soils of contrasting land use types.

In order to differentiate and quantify C decomposed from either added plant residues or SOM, ^{14}C-labelled maize residue was used in our study. Most of the studies have focused on the effects of either N or disturbance alone on decomposition of either plant residue or/and SOM in a single soil, however, N application and disturbance caused by tillage are common practices in cropland. Therefore, the objectives of this study were to quantify the effects of mineral-N addition and intensive mixing (analogous to disturbance by plowing) on the decomposition of ^{14}C-labelled maize residue and SOM in three soils with different management: plow tillage (PT), reduced tillage (RT) and grassland (GL) soils.

2. Materials and methods

2.1. Soils

Soil samples were collected from three land use types at Edelweiler (8°33′E, 48°32′N), located in the Black Forest (Baden-Württemberg, southwest Germany), with mean annual temperature 6–7 °C, mean annual precipitation 1000–1200 mm (655 m above sea level).

Nearby GL, RT and PT plots were carefully selected at this site, with similar soil type, stoniness and slope. The GL was established in 2000 on previous long-term (>10 years) plowed soil. The GL was used for hay or silage, and was cut four times per year. The RT was managed since 1991 in a crop rotation of maize for silage, winter wheat, and winter triticale (×*Triticosecale*). The tillage depth for maize was 10 cm and for winter wheat and winter triticale was 8 cm. The PT plot was cultivated for 5 years on previous 2-year clover (*Trifolium pretense*) grassland. At Edelweiler, farmers have the tradition to grow clover grass for 2 years after every 10 years of PT. The cropping systems in PT included oat (*Avena sativa* L.), summer barley (*Hordeum vulgare* L.), triticale and rapeseed (*Brassica napus* L.); the tillage depth was 15 cm. Soils developed from sandstone (Buntsandstein) and were classified as Dystric Cambisol (WRB, the World Reference Base of Soil Resources (FAO, 1998)) with 19.3% clay, 48.8% silt and 31.9% sand.

Soil was collected from the upper 5 cm of the three land use practices in November 2004. Soil samples were air dried, mixed and passed through a 2-mm sieve; plant residues >2 mm such as straw and roots were removed before chemical analyses (Table 1).

2.2. Incubation

The laboratory incubation was conducted in closed vessels for 112 days at 20 °C. Twenty grams of air-dried

Table 1
Selected characteristics of the soils (0–5 cm) collected from Edelweiler, Germany

Land use	Plow tillage	Reduced tillage	Grassland
pH (CaCl$_2$)	5.31 ± 0.10	4.60 ± 0.03	4.97 ± 0.07
SOC (%)	2.109 ± 0.004	2.618 ± 0.121	2.991 ± 0.107
Total N (%)	0.179 ± 0.002	0.224 ± 0.007	0.257 ± 0.009
C/N	11.8 ± 0.1	11.7 ± 0.3	11.6 ± 0.2
WHC (%)	30.00	34.14	28.75
C-microbial biomass (μg g^{-1})	372.0 ± 29.1	274.8 ± 30.5	564.9 ± 41.6
N-microbial biomass (μg g^{-1})	54.8 ± 0.6	40.6 ± 4.8	76.0 ± 4.5

Values are means ± S.D. ($n = 3$); SOC: soil organic carbon; WHC: water holding capacity.

soil were weighed into 250 ml glass vessels. The experiment was set up with 24 treatments in triplicates including four factors. The first factor was three land use types: PT, RT, and GL (described above). The second factor was ^{14}C-labelled maize residue addition. No residue (−residue) or 80 mg ^{14}C-labelled maize residue (+residue) ground with ball mill (specific activity 5970 DPM mg^{-1} C, 36.5% C and 2.9% N) was added to each of the three soils. The third factor was mineral-N application. No N (−N) or 2 mg N as NH$_4$NO$_3$ (100 mg N kg^{-1} soil) solution (+N) was applied to each of the three soils. The fourth factor was intensive mixing. No mixing (−mixing) or intensive mixing (+mixing) was performed in each of the three soils. ^{14}C-labelled maize residues were taken from the previous experiment, in which maize shoots were labelled three times during 29 days of growth (Werth and Kuzyakov, 2006). At the beginning, labelled maize residues were thoroughly mixed with the soil. The soil or soil–residue mixture was moistened to 80% of its water holding capacity (WHC) with deionized water or NH$_4$NO$_3$ solution. Small vials with 4 ml of 1.0 M NaOH were placed in the vessels for trapping CO$_2$. The traps were changed weekly till day 70 and biweekly to the end of incubation (14 sampling cases). Additional intensive mixing of the soil (analogous to disturbance by plowing) was performed by hand stirring for 0.5 min and was carried out on days 14, 43, 56 and 70. Additional triplicate blank vessels containing only the vials with NaOH served as controls to account for the CO$_2$ trapped from the air inside the vessels. Deionized water was added to the vessels as needed to keep soil moisture at 80% of WHC.

2.3. Analysis

The concentration of total C and N were determined in ball-milled soil samples by dry combustion with a LECO CN2000 analyzer. Since the soils were free of carbonates, total C was equivalent to SOC. Soil pH was measured using a 1:2.5 (w/v) soil to 0.01 M CaCl$_2$ ratio with a glass electrode. Soil texture was determined by the pipette method (Schlichting et al., 1995).

Microbial biomass C and N in soil samples before incubation was measured by the fumigation-extraction method (Vance et al., 1987). Each replicate was divided into two equivalent portions, one was fumigated for 24 h with ethanol-free chloroform and the other was the unfumigated control. Both fumigated and unfumigated soils, were shaken for 30 min with 0.5 M K$_2$SO$_4$ (1:4 soil:extraction ratio) and centrifuged and filtered. Extracts were analyzed for total organic C and total N on a Dimatoc-100 analyzer (Dimatec Co., Essen, Germany). A K_C value of 0.45 and a K_N of 0.54 were used to calculate the C and N content of the microbial biomass (Jorgensen, 1996; Joergensen and Mueller, 1996).

Total CO$_2$ absorbed in NaOH solution was measured by titration of an aliquot (1 ml) with 0.1 M HCl against phenolphthalein after addition of 0.5 M BaCl$_2$ solution (Black, 1965). The ^{14}C activity in CO$_2$ collected in NaOH solution was measured by adding 2 ml scintillation cocktail Rothiscint-22× (Roth Company, Germany) to 1 ml aliquot of NaOH after the decay of chemiluminescence. The ^{14}C counting efficiency was about 87% and the ^{14}C activity measurement error did not exceed 2%.

2.4. Calculations and statistical analysis

^{14}CO$_2$ efflux from maize residue was presented as percentages of initial input ^{14}C activity. To compare the unlabelled CO$_2$ efflux from soils having initially different SOC content, all results were presented as percentages of SOC. ^{14}CO$_2$ or CO$_2$ efflux rates were calculated as ^{14}CO$_2$ or CO$_2$ production divided by each sampling interval, and expressed as % of input ^{14}C per day or % of SOC per day. Percent change induced by

mineral-N addition (+N) or soil mixing was calculated as follow:

$$\text{Percent change} = \frac{((+\text{N (or + mixing) treat.)} - (-\text{N (or} - \text{mixing)treat.)})}{(-\text{N (or} - \text{mixing) treat.)}} \times 100 \tag{1}$$

The corresponding standard deviation (S.D.) was calculated as follow:

$$\text{S.D.} = \sqrt{\left(\frac{\text{S.D.}_A}{\bar{A} - \bar{B}}\right)^2 + \left(\frac{\text{S.D.}_B}{\bar{A} - \bar{B}}\right)^2} \tag{2}$$

where S.D._A and S.D._B are standard deviations of +N (or +mixing) treatments and the respective controls −N (or −mixing), respectively. $\bar{A}$ and $\bar{B}$ are the mean values of +N (or +mixing) treatments and the respective controls −N (or −mixing), respectively.

Standard errors (S.E.) were calculated as:

$$\text{S.E.} = \frac{\text{S.D.}}{\sqrt{n}} \tag{3}$$

Within each land use type, the effects of N and intensive mixing on decomposition of ^{14}C-labelled maize residue and SOM were subjected to one-way ANOVA to test for significant difference between the treatments and their respective controls separately. Differences were considered significant at 0.05 level. The SAS statistical package was used for the statistical analysis (SAS, 1998).

The statistic analysis of the effects of N, intensive mixing, land use types, incubation time and all interactions on cumulative ^{14}CO$_2$ efflux was performed with SPSS for windows, Version 11.0.

Table 2
Significance of nitrogen (N), mixing, land use types (L), decomposition period (T) and all interactions on cumulative ^{14}CO$_2$ efflux

Effect	d.f.	F	Sig.
T	13	29032	<0.001
L	2	1121	<0.001
Mixing	1	273	<0.001
N	1	714	<0.001
T × L	26	210	<0.001
T × mixing	13	2	0.045
L × mixing	2	40	<0.001
T × L × mixing	26	1	0.171
T × N	13	35	<0.001
L × N	2	392	<0.001
T × L × N	26	8	<0.001
N × mixing	1	31	<0.001
T × mixing × N	13	<1	0.914
L × mixing × N	2	15	<0.001
T × L × mixing × N	26	<1	0.998

3. Results

3.1. Effect of nitrogen

3.1.1. Effect of N on ^{14}C-labelled maize residue decomposition

The effects of N, land use types, and decomposition period on ^{14}CO$_2$ efflux from maize residue were all significant ($p < 0.05$, Table 2). There were significant interactions between N and decomposition period, between N and land use types, and between N and mixing. The three-ways interactions of N × decomposition period × land use types and N × land use types × mixing were also significant ($p < 0.05$, Table 2).

In the first phase of decomposition (1–5 days), addition of inorganic N significantly increased mean ^{14}CO$_2$ efflux rates in RT and GL land uses, but not in PT ($p < 0.05$, Fig. 1). In the subsequent 11–105 days, N
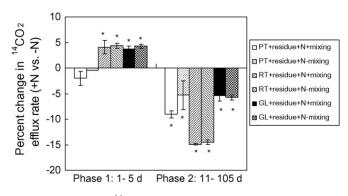

Fig. 1. Percent change in mean ^{14}CO$_2$ efflux rate (% of input ^{14}C per day) induced by mineral-N addition compared to the respective controls without N during two decomposition phases of maize residue (mean ± S.E.) (symbol (*) denotes values are significantly different ($p < 0.05$) from the respective controls without N; PT: plow tillage; RT: reduced tillage; GL: grassland; +residue: with residue; +N: with nitrogen; −N: without nitrogen; +mixing: with mixing; −mixing: without mixing).

addition significantly decreased mean $^{14}CO_2$ efflux rate in all three land use types ($p < 0.05$, Fig. 1). The magnitude of decrease caused by N addition tended to be stronger in RT than in PT and GL ($p < 0.05$, Fig. 1).

Application of inorganic N significantly increased $^{14}CO_2$ efflux in GL + residue + N − mixing in the first 7 days and in GL + residue + N + mixing in the first sampling, but showed no significant effects in GL later ($p < 0.05$, Fig. 2). After the initial increase, the cumulative $^{14}CO_2$ effluxes in RT with added N were significantly decreased from day 21 to the end of the incubation compared to the respective controls without N ($p < 0.05$, Fig. 2). Pronounced negative effects of N additions were observed in PT from day 36 to the end of the experiment ($p < 0.05$, Fig. 2). The magnitude of decrease of plant residue decomposition induced by N addition tended to rank in the order: RT > PT > GL, within each land use type, treatments with intensive mixing >treatments without intensive mixing, from day 21 to the end of the incubation. Clearly, the reduced $^{14}CO_2$ production with addition of N was associated with lower microbial biomass C and N contents.

3.1.2. Effect of N on soil organic matter decomposition

The effect of N on SOM decomposition was evaluated in the soil-only treatments (without plant residue addition).

In the first phase of decomposition (1–5 days), N addition significantly decreased CO_2 efflux rate in PT + N + mixing (−8.4%), but not in the other treatments ($p < 0.05$, Fig. 3). After day 10, mineral-N
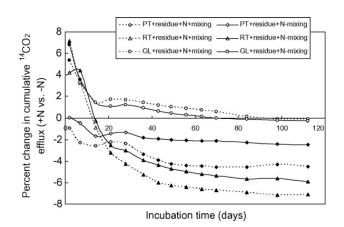

Fig. 3. Percent change in mean CO_2 efflux rate (% of soil organic C per day) induced by mineral-N addition compared to the respective controls without N (mean ± S.E.) during two decomposition phases of soil organic matter (symbol (*) denotes values are significantly different ($p < 0.05$) from the respective controls without N; PT: plow tillage; RT: reduced tillage; GL: grassland; +N: with nitrogen; −N: without nitrogen; +mixing: with mixing; −mixing: without mixing).

significantly suppressed CO_2 efflux rate only in RT + N − mixing (−9.4%) ($p < 0.05$, Fig. 3).

Mineral-N did not significantly affect cumulative CO_2 emission in GL throughout the incubation, except for the negative effect in GL + N + mixing in the second sampling ($p < 0.05$, Fig. 4). No significant effect of N addition was observed in PT during the incubation, except the negative effect in PT + N + mixing in the first three samplings ($p < 0.05$, Fig. 4). After the stimulation in the first sampling date, mineral-N exhibited no significant effects on cumulative CO_2 production in RT + N + mixing in the rest of the incubation ($p < 0.05$, Fig. 4). However, cumulative CO_2 efflux was significantly suppressed by N addition in RT + N − mixing from day 36 to day 112 ($p < 0.05$, Fig. 4).

Fig. 2. N addition induced percent change in cumulative $^{14}CO_2$ efflux (% of input ^{14}C) compared to the respective controls without N (the full dark symbol denotes values are significantly different ($p < 0.05$) from the respective controls without N; PT: plow tillage; RT: reduced tillage; GL: grassland; +residue: with residue; +N: with nitrogen; −N: without nitrogen; +mixing: with mixing; −mixing: without mixing).
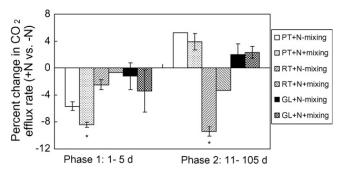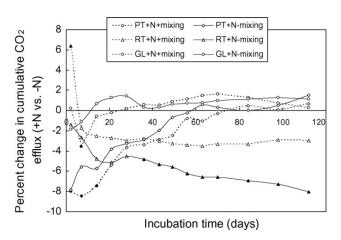

Fig. 4. N addition induced percent change in cumulative CO_2 efflux (% of soil organic C) compared to the respective controls without N (the full dark symbol denotes values are significantly different ($p < 0.05$) from the respective controls without N; PT: plow tillage; RT: reduced tillage; GL: grassland; +N: with nitrogen; −N: without nitrogen; +mixing: with mixing; −mixing: without mixing).
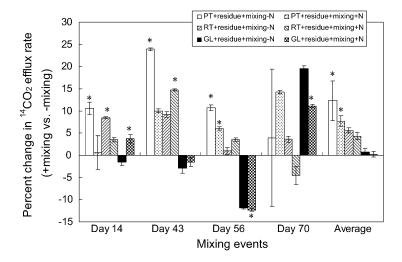
Fig. 5. Percent change in mean $^{14}CO_2$ efflux rate (% of input ^{14}C per day) induced by mixing events compared to the respective controls without mixing (mean ± S.E.) (symbol (*) denotes values are significantly different ($p < 0.05$) from the respective controls without mixing; PT: plow tillage; RT: reduced tillage; GL: grassland; +residue: with residue; +mixing: with mixing; −mixing: without mixing; +N: with nitrogen; −N: without nitrogen).

3.2. Effect of intensive mixing

3.2.1. Effect of intensive mixing on decomposition of ^{14}C-labelled maize residue

Incubated soil samples were intensively mixed on 4 dates, on day 14, 43, 56 and 70. The effect of intensive mixing on mean $^{14}CO_2$ efflux rates between two subsequent mixing events is shown in Fig. 5. The first mixing significantly increased mean $^{14}CO_2$ efflux rates in PT + residue + mixing − N (+10.6%), RT + residue + mixing − N (+8.5%), and GL + residue + mixing + N (+3.7%), compared to their respective controls without mixing ($p < 0.05$, Fig. 5). The second mixing exhibited positive effects in PT + residue + mixing − N (+23.9%) and RT + residue + mixing + N (+14.7%) ($p < 0.05$, Fig. 5). The third mixing induced significant increase in $^{14}CO_2$ production rate in PT ($p < 0.05$, Fig. 5). The fourth mixing resulted in a positive effect in GL + residue + mixing + N (+11.1%) ($p < 0.05$, Fig. 5). When calculating the average $^{14}CO_2$ efflux rates over the four mixing events, there was a significant positive increase only in PT ($p < 0.05$, Fig. 5). Clearly, the response of land use type with higher C and N contents to mixing was smaller.

3.2.2. Effect of intensive mixing on decomposition of SOM

The effect of intensive mixing on SOM decomposition was assessed in the soil-only treatments (without plant residue addition).

Fig. 6 shows the effect of intensive mixing on the mean CO_2 efflux rate between two subsequent mixing events. The first mixing significantly increased mean

CO_2 efflux rate in PT and GL + mixing + N ($p < 0.05$, Fig. 6). The second mixing significantly stimulated SOM mineralization rate in PT + mixing + N (+5.9%) and RT + mixing + N (+14.5%) ($p < 0.05$, Fig. 6). The significant increase in mean CO_2 efflux rate caused by the third mixing also occurred in PT + mixing + N and RT + mixing + N ($p < 0.05$, Fig. 6). The fourth mixing did not increase mean CO_2 efflux rate significantly in any treatment ($p < 0.05$, Fig. 6). When taking all mixing events into account, significant increases were detected in PT + mixing + N (+7.7%) and RT + mixing + N (+15.3%), and no significant effects occurred in GL ($p < 0.05$, Fig. 6).
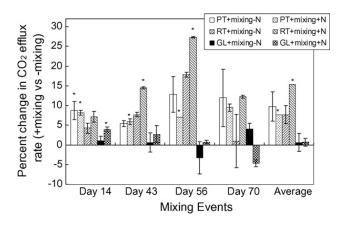

Fig. 6. Percent change in mean CO_2 efflux rate (% of soil organic C per day) induced by mixing events compared to the respective controls without mixing (mean ± S.E.) (symbol (*) denotes values are significantly different ($p < 0.05$) from the respective controls without mixing; PT: plow tillage; RT: reduced tillage; GL: grassland; +mixing: with mixing; −mixing: without mixing; +N: with nitrogen; −N: without nitrogen).

4. Discussion

4.1. Effect of N on decomposition of ^{14}C-labelled maize residue

The effects of mineral-N on maize residue decomposition differed depending on the stage of decomposition and land use types.

Addition of mineral-N increased maize residue decomposition rates in RT and GL in the first phase (1–5 days) (Fig. 1). Added mineral-N increased the decomposition rate of residue by satisfying N requirements of microorganisms (Jenkinson and Rayne, 1985). However, the positive effects disappeared after 5 days, suggesting that water-soluble compounds had been largely degraded.

Mineral-N decreased residue decomposition rate in all the treatments in the second phase (11–105 days) (Fig. 1), which was consistent with several reports (Fog, 1988; Henriksen and Breland, 1999; Carreiro et al., 2000; Saiya-Cork et al., 2002; Wang et al., 2004; Moran et al., 2005). The mechanisms of the suppressive effects of N in the late stage are not fully understood, but some were partly proven and suggested. Fog (1988) suggested that ammonia suppressed the production of enzymes required for degradation of lignin and other recalcitrant compounds. Carreiro et al. (2000) found that chronically applied aqueous NH_4NO_3 reduced the activity of ligninolytic enzyme produced by white-rot fungi. Ågren et al. (2001) showed that mineral-N inputs increased decomposer efficiency, but led to more rapid formation of recalcitrant material. The most likely reason why decomposer efficiency increases with increasing N availability is that there is a shift in the decomposer community composition towards organisms that are more efficient but have a greater N requirement (Ågren et al., 2001). Similarly, Moran et al. (2005) found that significantly higher residue-C amount was transformed into humin-C with mineral-N input suggesting that mineral-N enhances residue decomposition and favors SOM formation.

Addition of N decreased cumulative $^{14}CO_2$ efflux in RT and PT but not in GL in the late stage (Fig. 2). This was probably connected with higher microbial biomass and SOC and total N contents in GL (Table 1).

At the end of the incubation, mean cumulative $^{14}CO_2$ production in treatments with N addition was suppressed as compared to treatments without N addition by 3.5 and 6.5% in PT and RT, respectively (Fig. 2). A decrease in residue mineralization could suggest that more residue-C is sequestrated in soil or was incorporated into microbial biomass. Although, the decreases were small, however, as N fertilizer application may occur regularly in the field, the small differences in the residue decomposition caused by addition of N could add up significant differences in long-term storage of SOM (Ågren et al., 2001). However, long-term field studies are necessary to elucidate this question.

4.2. Effect of N on decomposition of SOM

Addition of mineral-N decreased or caused no change in SOM decomposition in the three land use types at Edelweiler (Figs. 3 and 4). Our results are consistent with a number of studies. With addition of $(NH_4)_2SO_4$ or NH_4NO_3, decreased or unchanged soil respiration was observed in laboratory incubated soils (Henriksen and Breland, 1999; Foereid et al., 2004; Moran et al., 2005). Field studies have also shown that N application depressed or did not influence soil respiration (Bowden et al., 2004; Burton et al., 2004; Micks et al., 2004).

While a decrease or no change in SOM decomposition in response to N addition may be a common finding, the mechanisms have not been clearly identified. Neff et al. (2002) thought that soils are highly complex media with a diversity of substrates that vary in both the energy required for their breakdown and in their total N content. As a result, addition of N to soil could increase decomposition rates of some SOM fractions while simultaneously decreasing rates for other fractions. Different results in the literature may be due to varied and compensatory response of SOM fractions to N additions. With respect to the suppressive effect of N on SOM decomposition, some mechanisms have been suggested. N additions decreased microbial biomass, microbial activity and diversity (Thirukkumaran and Parkinson, 2000; Bowden et al., 2004; Compton et al., 2004). Frey et al. (2004) reported that active fungal biomass was lower in the fertilized compared to control plots, while active bacterial biomass was not greatly affected by N additions, resulting in significantly lower fungal: bacterial biomass ratios in the N-treated plots. This shift in microbial community composition was accompanied by a significant reduction in the activity of phenol oxidase, a lignin-degrading enzyme produced by white-rot fungi. Saiya-Cork et al. (2002) observed that N amendment decreased phenol oxidase activity. Decreases in soil pH due to nitrification caused by N addition could also influence microbial communities (Fog, 1988; Kuzyakov et al., 2000; Bowden et al., 2004; Compton et al., 2004).

Thus, improved understanding of different fractions of SOC and the underlying mechanisms controlling the direction and magnitude to N availability is highly needed for accurate prediction of SOM decomposition. However, our results clearly showed that N addition did not stimulate decomposition of SOM in all three soils from different land use types.

4.3. Effect of intensive soil mixing

Our results clearly showed trends (4 of 12 observations significant) that intensive mixing accelerated decomposition rates of maize residue and native SOM in the three land use types (Figs. 5 and 6), which were in agreement with other reports (Sorensen, 1979; Powlson, 1980; Hassink et al., 1993; Bell et al., 2003).

Intensive mixing was carried out by hand stirring which was intended to increase aeration, destroy aggregates and therefore, prevent stabilization of plant residue within the aggregates. Therefore, under mixing treatments, residue and SOM were more susceptible to microbial decomposition than in the controls without mixing (Elliott, 1986; Reicosky et al., 1997; Tebrügge and Düring, 1999; Six et al., 2000). In contrast, in treatments without mixing, soil aggregation and aggregate stability would have likely been improved, residue and SOC would have been protected in the aggregates and the decomposition would have been slowed down (Beare et al., 1994; Franzluebbers et al., 1995; Six et al., 2000; Six et al., 2002). The accumulation of higher SOC and total N contents in GL and RT than in PT was also a manifestation of less disturbance in the field.

When taking all four mixing events into account, the extent of stimulating effects on both maize residue and SOM decomposition rate was much smaller in GL than in RT and PT, indicating that aggregates were more stable in GL, disturbance may not have disrupted aggregates in the mixing treatments, and stronger aggregates in GL allowed better protection of SOM and plant residue against decomposition.

Our results indicated that if grassland were converted to cropland, in which soil were frequently mixed, plant residue and SOM mineralization would be accelerated. Many studies have shown that adoption of conservation tillage and switching from cropland to grassland minimize SOM losses, maintain and even increase SOM stocks (Lal, 1997; Bayer et al., 2000; West and Post, 2002; Franzluebbers, 2005). However, caution should also be taken with regard to the adoption of conservation tillage to increase C sequestration, as Baker et al. (2007) reported that evidence that conservation tillage promotes C sequestration is not compelling.

5. Conclusions

The effects of mineral-N on decomposition of SOM were different from the effects on decomposition of plant residues. N additions stimulated residue-C mineralization in RT and GL in the first 5 days, but thereafter decreased maize residue decomposition in all three land use types. At the end of the incubation, N additions significantly suppressed maize residue decomposition in RT and PT. Mineral-N additions decreased or caused no change in SOM decomposition in RT, PT, and GL.

Intensive soil mixing increased decomposition rates of both plant residue and SOM in all three land use types. The magnitude of increase was smaller in GL than in RT or PT, indicating that soil aggregates in GL were more stable and had greater protective capacity against disturbance. Therefore, conversion from grassland to cropland would increase decomposition rates of residue and SOM, not only because of lower C input in cropland, but also because of low aggregate stability and protection of organic matter.

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References

Ågren, G.I., Bosatta, E., Magill, A.H., 2001. Combining theory and experiment to understand effect of inorganic nitrogen on litter decomposition. Oecologia 128, 94–98.

Alvarez, R., Santanatoglia, O.J., Garcia, R., 1995. In situ decomposition of ^{14}C-labelled wheat as affected by soil disturbance. Agrochimica 39, 343–348.

Baker, J.M., Ochsner, T.E., Venterea, R.T., Griffis, T.J., 2007. Tillage and soil carbon sequestration. What do we really know? Agric. Ecosyst. Environ. 118, 1–5.

Bayer, C., Mielniczuk, J., Amado, T.J.C., Martin-Neto, L., Fernandes, S.V., 2000. Organic matter storage in a sandy clay loam Acrisol affected by tillage and cropping systems in southern Brazil. Soil Till. Res. 54, 101–109.

Beare, M.H., Hendrix, P.F., Coleman, D.C., 1994. Water-stable aggregates and organic matter fractions in conventional-tillage and no-tillage soils. Soil Sci. Soc. Am. J. 58, 777–786.

Bell, J.M., Smith, J.L., Bailey, V.L., Bolton, H., 2003. Priming effect and C storage in semi-arid no-till spring crop rotations. Biol. Fertil. Soils 37, 237–244.

Berg, B., Matzner, E., 1997. Effect of N deposition on decomposition of plant litter and soil organic matter in forest systems. Environ. Rev. 5, 1–25.

Black, C.A., 1965. Methods of Soil Analysis. American Society of Agronomy, Inc., Madison, Wisconsin, USA.

Bowden, R.D., Davidson, E., Savage, K., Arabia, C., Steudler, P., 2004. Chronic nitrogen additions reduce total soil respiration and microbial respiration in temperate forest soils at the Harvard Forest. For. Ecol. Manage. 196, 43–56.

Burton, A.J., Pregitzer, K.S., Crawford, J.N., Zogg, G.P., Zak, D.R., 2004. Simulated chronic NO_3^- deposition reduces soil respiration in northern hardwood forests. Global Change Biol. 10, 1080–1091.

Cao, M., Woodward, F.I., 1998. Dynamic responses of terrestrial ecosystem carbon cycling to global climate change. Nature 393, 249–252.

Carreiro, M.M., Sinsabaugh, R.L., Repert, D.A., Parkhurst, D.F., 2000. Microbial enzyme shifts explain litter decay responses to stimulated nitrogen deposition. Ecology 81, 2359–2365.

Christensen, B.T., Johnston, A.E., 1997. Soil organic matter and soil quality-lessons learned from long-term experiments at Askov and Rothamsted. In: Gregorich, E.G., Carter, M.R. (Eds.), Soil Quality for Crop Production and Ecosystem Health. Elsevier, Amsterdam, NI, pp. 399–430.

Compton, J., Watrud, L.S., Porteus, L.A., DeGrood, S., 2004. Response of soil microbial biomass and community composition to chronic nitrogen additions at Harvard forest. For. Ecol. Manage. 196, 143–158.

Conde, E., Cardenas, M., Ponce-Mendoza, A., Luna-Guido, M.L., Cruz-Mondragón, C., Dendooven, L., 2005. The impacts of inorganic nitrogen application on mineralization of ^{14}C-labelled maize and glucose, and on priming effect in saline alkaline soil. Soil Biol. Biochem. 37, 681–691.

Elliott, E.T., 1986. Aggregate structure and carbon, nitrogen, and phosphorus in native and cultivated soils. Soil Sci. Soc. Am. J. 50, 627–633.

FAO, 1998. World reference base for soil resources. Would Soil Resources Report No. 84. Food and Agricultural Organization of the United Nations, Rome.

Foereid, B., de Neergaard, A., HØgh-Jensen, H., 2004. Turnover of organic matter in a *Miscanthus* field: effect of time in *Miscanthus* cultivation and inorganic nitrogen supply. Soil Biol. Biochem. 36, 1075–1085.

Fog, K., 1988. The effects of added nitrogen on the rate of decomposition of organic matter. Biol. Rev. 63, 433–462.

Franzluebbers, A.J., 2005. Soil organic carbon sequestration and agricultural greenhouse gas emissions in the southeastern USA. Soil Till. Res. 83, 120–147.

Franzluebbers, A.J., Hons, F.M., Zuberer, D.A., 1995. Tillage and crop effects on seasonal dynamics of soil CO_2 evolution, water content, temperature, and bulk density. Appl. Soil Ecol. 2, 95–109.

Frey, S.D., Knorr, M., Parent, J.L., Simpson, R.T., 2004. Chronic nitrogen enrichment affects the structure and function of the soil microbial community in temperate hardwood and pine forests. For. Ecol. Manage. 196, 159–171.

Hassink, J., Bouwman, L.A., Zwart, K.B., Bloem, J., Brussaard, L., 1993. Relationship between soil texture, physical protection of soil organic matter, soil biota, and C and N mineralization in grassland soils. Geoderma 57, 105–128.

Henriksen, T.M., Breland, T.A., 1999. Nitrogen availability effects on carbon mineralization, fungal and bacterial growth, and enzyme activities during decomposition of wheat straw in soil. Soil Biol. Biochem. 31, 1121–1134.

Jenkinson, D.S., Rayne, J.H., 1985. Interactions between fertilizer nitrogen and soil nitrogen-the so-called 'priming' effect. J. Soil Sci. 36, 425–444.

Jorgensen, R.G., 1996. The fumigation-extraction method to estimate soil microbial biomass: calibration of the k_{EC} value. Soil Biol. Biochem. 28, 25–31.

Joergensen, R.G., Mueller, T., 1996. The fumigation-extraction method to estimate soil microbial biomass: calibration of the K_{EN} value. Soil Biol. Biochem. 28, 33–37.

Körner, C., Arnone, J.A., 1992. Responses to elevated carbon dioxide in artificial tropical ecosystems. Science 257, 1672–1675.

Kuzyakov, Y., Friedel, J.K., Stahr, K., 2000. Review of mechanisms and quantification of priming effects. Soil Biol. Biochem. 32, 1485–1498.

Lal, R., 1997. Residue management, conservation tillage and soil restoration for mitigating greenhouse effect by CO_2-enrichment. Soil Till. Res. 43, 81–107.

Lal, R., 2004. Soil carbon sequestration to mitigate climate change. Geoderma 123, 1–22.

Micks, P., Aber, J.D., Boone, R.D., Davidson, E.A., 2004. Short-term soil respiration and nitrogen immobilization response to nitrogen applications in control and nitrogen-enriched temperate forests. For. Ecol. Manage. 196, 57–70.

Moran, K.K., Six, J., Horwath, W.R., van Kessel, C., 2005. Role of mineral-nitrogen in residue decomposition and stable soil organic matter formation. Soil Sci. Soc. Am. J. 69, 1730–1736.

Nadelhoffer, K.J., Emmett, B.A., Gundersen, P., Kjonaas, O.J., Koopmans, C.J., Schleppi, P., Tietema, A., Wright, R.F., 1999. Nitrogen deposition makes a minor contribution to carbon sequestration in temperate forests. Nature 398, 145–148.

Neff, J.C., Townsend, A.R., Glexner, G., Lehman, S.J., Turnbull, J., Bowman, W.D., 2002. Variable effects of nitrogen additions on the stability and turnover of soil carbon. Nature (London) 419, 915–917.

Paul, E.A., Collins, H.P., Paustian, K., Elliott, E.T., Frey, S., Juma, N., Janzen, H., Campbell, C.A., Zentner, R.P., Lafond, G.P., Moulin, A.P., 2004. Management effects on the dynamics and storage rates of organic matter in long-term crop rotations. Can. J. Soil Sci. 84, 49–61.

Paustian, K., Six, J., Elliott, E.T., Hunt, H.W., 2000. Management options for reducing CO_2 emissions from agricultural soils. Biogeochemistry 48, 147–163.

Post, W.M., Peng, T.H., Emanuel, W.R., King, A.W., Dale, V.H., DeAngelis, D.L., 1990. The global carbon cycle. Am. Sci. 78, 310–326.

Powlson, D.S., 1980. The effects of grinding on microbial and non-microbial organic matter in soil. J. Soil Sci. 31, 77–85.

Reicosky, D.C., Dugas, W.A., Torbert, H.A., 1997. Tillage-induced soil carbon dioxide loss from different cropping systems. Soil Till. Res. 41, 105–118.

Reicosky, D.C., Kemper, W.D., Langdale, G.W., Douglas, C.L., Rasmussen, P.E., 1995. Soil organic matter changes resulting from tillage and biomass production. J. Soil Water Conserv. 50, 253–261.

Saiya-Cork, K.R., Sinsabaugh, R.L., Zak, D.R., 2002. The effects of long term nitrogen deposition on extracellular enzyme activity in an *Acer saccharum* forest soil. Soil Biol. Biochem. 34, 1309–1315.

SAS, 1998. SAS System for Windows, Version 6.12. SAS Institute Inc., Cary, NC.

Schlichting, E., Blume, H.P., Stahr, K., 1995. Bodenkundiches Praktikum, second ed. Blackwell, Berlin.

Six, J., Elliott, E.T., Paustian, K., 2000. Soil macroaggregate turnover and microaggragate formation: a mechanism for C sequestration under no-tillage agriculture. Soil Biol. Biochem. 32, 2099–2103.

Six, J., Feller, C., Denef, K., Ogle, S.M., Sa, J.C.D., Albrecht, A., 2002. Soil organic matter, biota, and aggregation in temperate and tropical soils-effects of no-tillage. Agronomie 22, 755–775.

Smith, W.N., Desjardins, R.L., Pattey, E., 2000. The net flux of carbon from agricultural soils in Canada 1970–2010. Global Change Biol. 6, 557–568.

Sorensen, L.H., 1979. Decomposition of straw in soil after stepwise repeated addition. Soil Biol. Biochem. 11, 23–29.

Tebrügge, F., Düring, R.A., 1999. Reducing tillage intensity—a review of results from a long-term study in Germany. Soil Till. Res. 53, 15–28.

Thirukkumaran, C.M., Parkinson, D., 2000. Microbial respiration, biomass, metabolic quotient and litter decomposition in a lodgepole pine forest floor amended with nitrogen and phosphorous fertilizers. Soil Biol. Biochem. 32, 59–66.

Townsend, A.R., Braswell, B.H., Holland, E.A., Penner, J.E., 1996. Spatial and temporal patterns in terrestrial carbon storage due to deposition of fossil fuel nitrogen. Ecol. Appl. 6, 806–814.

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

Vitousek, P.M., Aber, J.D., Howarth, R.W., Likens, G.E., Matson, P.A., Schindler, D.W., Schlesinger, W.H., Tilman, D.G., 1997. Human alteration of the global nitrogen cycle: sources and consequences. Ecol. Appl. 7, 737–750.

Wang, W.J., Baldock, J.A., Dalal, R.C., Moody, P.W., 2004. Decomposition dynamics of plant materials in relation to nitrogen availability and biochemistry determined by NMR and wet-chemical analysis. Soil Biol. Biochem. 36, 2045–2058.

Werth, M., Kuzyakov, Y., 2006. Assimilate partitioning affects ^{13}C fractionation of recently assimilated carbon in maize. Plant Soil 284, 319–333.

West, T.O., Post, W.M., 2002. Soil carbon sequestration by tillage and crop rotation: a global data analysis. Soil Sci. Soc. Am. J. 66, 1930–1946.