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Soil organic carbon and microbial biomass after six years of reduced tillage under organic farming

Humusgehalt und mikrobielle Eigenschaften nach sechs Jahren reduzierter Bodenbearbeitung in der Ökologischen Landwirtschaft

University of Natural Resources and Applied Life Sciences
Department of Sustainable Agricultural Systems
Division of Organic Farming

FIBL, Research Institute of Organic Agriculture

Final Thesis by

Florian Gadermaier

Supervisor: Ao. Univ. Prof. DI Dr. agr. Jürgen Kurt Friedel

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Zusammenfassung

Konservierende, nicht wendende Bodenbearbeitung (Direktsaat und Reduzierte Bodenbearbeitung, RT) verringert die Bodenerosion und trägt dazu bei, Bodenfruchtbarkeit zu bewahren. Im Oberboden wird organischer Kohlenstoff (C_{org}) angereichert, die mikrobielle Biomasse und Aktivität steigen an. Es erfolgt eine Horizontdifferenzierung bei konservierender Bodenbearbeitung. Obwohl konservierende Bodenbearbeitung ökologische und ökonomische Vorteile bietet, ist die konventionelle Bodenbearbeitung mit Pflug (CT) nach wie vor Standard auf ökologisch wirtschaftenden Betrieben. In dieser Arbeit werden die Auswirkungen von Bodenbearbeitung (CT vs. RT), Düngung (Gülle vs. Mistkompost) und biologisch-dynamischen Präparaten (mit vs. ohne Präparate) auf die Bodenfruchtbarkeitsindikatoren C_{org}, mikrobielle Biomasse und mikrobielle Aktivität und Bodennährstoffgehalte von Phosphor (P) und Kalium (K) nach der ersten Fruchtfolgeperiode (sechs Jahre) untersucht und Nährstoffbilanzen berechnet. Der Langzeitversuch befindet sich auf tonigem Boden in Frick, Schweiz, die Summe der Jahresniederschläge beträgt im Mittel 1000 mm.

Bei RT stieg C_{org} in 0-10 cm Bodentiefe von 2,16 % (w/w) im Jahr 2002 auf 2,61 % im Jahr 2008 ($p<0.001$), keine Veränderungen gab es bei CT. C_{org} war in 10-20 cm Bodentiefe bei beiden Bodenbearbeitungsvarianten unverändert. Die Mengen an mikrobiellem Kohlenstoff und mikrobiellem Stickstoff lagen bei RT in 0-10 cm Bodentiefe um 37 % ($p<0.01$) beziehungsweise 35 % ($p<0.05$) über den Werten von CT, die mikrobielle Aktivität war um 57 % ($p<0.05$) erhöht. Die Gehalte an löslichem und pflanzenverfügbarem P waren bei RT in 0-10 cm Bodentiefe 75 % ($p<0.05$) und 27 % ($p<0.05$) höher als bei CT, die Unterschiede bei löslichem und pflanzenverfügbarem K betrugen + 40 % ($p<0.1$) und + 23 % ($p<0.05$). In 10-20 cm Bodentiefe lagen bei RT nur die Gehalten an mikrobiellem Kohlenstoff (+ 10 %, $p<0.05$) und die mikrobielle Aktivität (+ 17 %, $p<0.05$) über den Werten von CT. Die Düngung hatte keinen Einfluss auf die untersuchten Eigenschaften, der Einsatz von biologisch-dynamischen Präparaten erhöhte das Verhältnis von mikrobiellem Kohlenstoff zu mikrobiellem Stickstoff um 7 % ($p < 0.05$) in 0-10 cm Bodentiefe.

Die Nährstoffbilanzen für P waren in allen Varianten ausgeglichen, die Stickstoffbilanzen wiesen bei RT auf Grund höherer Erträge und Entzüge ein höheres Defizit und die Kaliumbilanzen einen niedrigeren Überschuss auf.

Die Ergebnisse zeigen, dass RT eine geeignet ist, um die Bodenfruchtbarkeit in der Ökologischen Landwirtschaft zu steigern. Die kombinierten Effekte von RT und einer ökologischen Bewirtschaftung mit einer abwechslungsreichen, auf Futterleguminosen basierenden Fruchtfolge und organischer Düngung bedürfen noch weiterer Forschung.

Abstract

No-tillage (NT) and reduced tillage (RT) systems are well-known management tools for preventing soil erosion and conserving soil fertility. NT and RT cause a stratification of soil organic carbon (C_{org}) and microbial properties in the soil profile. NT and RT may improve the environmental and economic performance of organic farming but they are still not common practice among organic farmers. This paper presents the effects of tillage (RT vs. conventional tillage, CT), fertilization (slurry vs. manure compost) and biodynamic preparations (with vs. without) on soil fertility indicators such as C_{org}, microbial biomass and microbial activity, soil nutrients (available fractions of P and K) and nutrient budgets in an organic farming system during the first crop rotation period (6 years) of a long-term experiment on a clayey soil in a temperate climate. Under RT, C_{org} in the 0-10 cm soil layer increased from 2.19% to 2.61% (w/w) ($p < 0.001$) from 2002 to 2008 whereas it remained constant under CT. In both tillage treatments C_{org} remained constant in 10-20 cm soil depth. Microbial biomass C and N were increased by 37% and 35% respectively under RT in 0-10 cm soil depth. Microbial activity (dehydrogenase activity = DHA) was even increased by 57%. Soluble and plant-available phosphorus were 72% ($p < 0.05$) and 27% ($p < 0.05$) higher in 0-10 cm under RT when compared to CT, soluble potassium was 40% ($p < 0.1$) and plant available potassium 23% ($p < 0.05$) higher under RT in 0-10 cm soil depth. Soil microbial biomass C and DHA in 10-20 cm were also higher under RT (+ 10% and + 17% respectively). There were no differences in soil microbial biomass N and contents of plant available nutrients in the 10-20 cm soil depth layer. Fertilization showed no effects, biodynamic preparations increased the C_{mic}-N_{mic} ratio by 7% ($p < 0.05$) in 0-10 cm soil depth. Nutrient budgets for P were balanced in all treatments; the N budget showed a higher deficit and the K budget a lower surplus under RT compared to CT due to higher yields under RT. Thus we conclude that RT is a suitable method for increasing soil fertility in organic farming systems. The combined effects of RT and an organic farming system with a diverse, ley-based crop rotation and organic fertilization merit further assessment.

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1. Introduction

Soil erosion and other forms of soil degradation are major problems facing agriculture today. Soils are not renewable over a human time scale. Most arable soils are prone to degradation, mainly caused by soil mismanagement. The degradation processes are more dependent on "how" rather than on "what" crops are grown (Lal, 2009), highlighting the importance of sustainable soil and crop management.

No-tillage (NT) and reduced tillage (RT) systems are well-known management tools for preventing soil erosion and conserving soil fertility (Pekrun and Claupein, 1998). A positive effect on soil organic carbon (C_{org}) contents in the superficial soil layer has frequently been reported (Pekrun and Claupein, 1998; Rasmussen, 1999; Kladivko, 2001; Kay and VandenBygaart, 2002; Alvarez, 2005; Koch and Stockfisch, 2006), whereas effects on C_{org} in the whole profile are still a matter of controversy (Baker *et al.*, 2007). NT and RT cause a stratification of C_{org} and microbial properties in the soil profile (Kandeler *et al.*, 1999; Kladivko, 2001; Kay and VandenBygaart, 2002; Peigné *et al.*, 2007). The intensity of tillage operations in RT and the amount and management of above-ground crop residues affect the degree of stratification. Total N, organic N, mineralizable N, P and K follow the same pattern with a concentration in the surface layer and no change or a decrease below (Peigné *et al.*, 2007).

Organic farming practices are reported to have a positive impact on air, soil, ground and surface water and biodiversity (FAO, 2003). Through concentration on building and maintaining soil fertility – mainly through multicropping systems and crop rotation, cover crops, organic fertilizers and minimum tillage – the organic matter content builds up and increases the soil's capacity to circulate nutrients, air and water (FAO, 2003). Crop production in organic farming relies and depends on nutrient transformation processes in the soil (Fließbach *et al.*, 2007). Soil quality is thus an important factor in organic farming and C_{org} is a lynchpin in this system. C_{org} in the topsoil is driven by interacting influences of climate, topography, soil type and aspects of crop management such as fertilization, tillage and crop rotation (Peigné *et al.*, 2007). Conversion of natural land to crop production and tillage generally leads to a loss of C_{org} (Scheffer, 2002).

C_{org} is reported to remain constant in an organic farming system including ley-based crop rotations and application of organic fertilizers, while it decreased under conventional farming with mineral fertilization (Fließbach *et al.*, 2007). Munro *et al.* (2002) found organically managed topsoils to contain a higher percentage of organic matter, total N and available P when compared to their conventional counterparts at 14 paired sites in England. Drinkwater *et al.* (1998) argue that the higher quality of added organic matter in organic farming leads to an accumulation of C_{org}. The microbial communities are key regulators of SOM dynamics and nutrient availability (Six *et al.*, 2006). Soil microbial biomass and activity, both indicators for biological soil fertility, are enhanced by organic farming (Fließbach and Mäder, 2000; Six *et al.*, 2006; Fließbach *et al.*, 2007). To ensure both short-term productivity and long-term sustainability, achieving a balance between inputs and outputs of nutrients is critical, especially as the use of imported materials to build soil fertility is restricted under organic farming (Watson *et al.*, 2002). Suitable crop rotations containing legumes produce surpluses in the N budgets of organic farms. P and K budgets show both surpluses and deficits, depending on the farm type and the import of nutrients (Watson *et al.*, 2002; Berry *et al.*, 2003).

The International Federation of Organic Agriculture Movements (IFOAM, 2006) recommends that organic farmers "take measures to prevent erosion, compaction, salinisation and other forms of soil degradation". Loss of topsoil should be minimized "through minimal tillage, contour ploughing, crop selection, maintenance of soil plant cover and other management practises that conserve soil".

Conservation tillage (NT and RT) may improve the environmental and economic performance of organic farming but is still not very common among organic farmers (Peigné *et al.*, 2007). There are major concerns about the adoption of conservation tillage. Increased weed pressure under conservation tillage as a result of mechanical weed control techniques not adapted to high levels of crop residues on the surface would appear to be the main problem. Topsoil compaction – especially during the first years of transition – and limited availability of N mainly at the beginning of the growing season also impede conversion to conservation tillage. Well-drained clays, stable loams and calcareous soils combined with moderate precipitation are favourable conditions for conservation tillage under organic farming conditions. Suitable crop rotations with a high weed-suppressing capacity include a ley phase, cover crops and intercropping (Peigné *et al.*, 2007).

The incorporation of the ley is a critical point under RT (Kainz *et al.*, 2005; Peigné *et al.*, 2007). Only a few experiments have investigated RT under organic farming conditions. Severe weed competition (Pekrun *et al.*, 2003; Kainz *et al.*, 2005; Schmidt *et al.*, 2006) and technical difficulties while incorporating grass-clover sods (Kainz *et al.*, 2005) led to the conclusion that an occasional use of the mouldboard plough is inevitable to overcome weed pressure under RT in organic farming (Hampl, 2005). Schulz *et al*. (2008) consistently found similar yields in CT and RT, when at least shallow turning of the soil was carried out. The stratification of C_{org}, soil nutrients, and microbial properties with RT in organic or conventional farming systems all developed in a similar way. Under organic farming, RT changed the allocation of C_{org} within the topsoil but did not enhance C_{org} over the whole investigated soil profile (Schulz *et al.*, 2008). Emmerling (2007) reported an increase in C_{org} in 0-25 cm soil depth where C_{org} was enhanced in the superficial layer (0-15 cm) and decreased in the layer below (15-25 cm). Microbial biomass and microbial activity in these soils were stratified under RT; there was an overall increase in microbial properties but the content of plant-available P in the investigated soil layer did not change (Emmerling, 2007).

Conservation tillage (NT and RT) in organic farming has not yet been successfully adapted and further research is required (Peigné *et al.*, 2007) into the adaptation of conservation tillage to different soils and climatic conditions, the development of suitable crop rotations and management practices to promote weed control and new strategies to remove and incorporate leys in a conservation tillage system. The influence of different fertilization strategies in stocked and stockless organic farming systems on N-mineralization and thus on reduced N supply under conservation tillage is not yet clearly understood.

In our experiment we studied the implementation of RT in an organic farming system with livestock. Two fertilization strategies, the effects of biodynamic preparations and their interactions with soil tillage were investigated. The first results of the conversion period from CT to RT showed an increase in C_{org}, microbial biomass and microbial activity in the superficial soil layer over the first 3 years. Average yields of cereals and sunflowers under RT were 93% of those obtained under CT (Berner *et al.*, 2008). Yields of fodder crops such as grass clover and silage maize were higher under RT, despite a considerably higher weed infestation of silage maize under RT (Krauss *et al.*, accepted). This paper presents the effects of tillage, organic fertilization strategies and biodynamic preparations on soil fertility indicators such as C_{org}, microbial biomass, microbial activity, soil nutrients and nutrient budgets after the first six-year crop rotation period of a long-term experiment on a clay soil in a temperate climate.

2. Material and methods

2.1 Field experiment

In autumn 2002 a factorized field experiment was established in Frick, Switzerland (47° 30' N, 8° 01' E) involving the factors tillage, fertilization and biodynamic preparations. A detailed description of the experiment is given by Berner *et al.* (2008).

CT uses a mouldboard plough operating at 15 cm depth. A chisel plough (15 cm) was used in the RT system and grass-clover in the RT system was superficially incorporated with a stubble cleaner running at 5 cm depth. Seedbed preparation was performed by a rotary harrow in both tillage systems (Table 1).

Inputs of organic matter (Table 2) were higher in the manure compost than in the slurry system due to the use of straw for animal bedding. The experimental farm where the experiment is located operates at a stocking density of 1.8 livestock units (LU) ha^{-1}. The farm has 19 ha of grassland and pastures and 13 ha of arable land. Mainly fodder crops are grown and additional fodder for swine breeding is purchased. Fertilization of our experiment was planned at a stocking density of 1.4 LU ha^{-1}. Differences in fertilization levels of N, P and K were due to different proportions of the excreted elements in solid and liquid organic manure types in the stable system and to N losses during manure storage. We aimed to achieve identical fertilization levels for phosphorus (P) and thus accepted differences for N and K. Consequently plots with slurry fertilization received N, P and K at levels of 1.13, 1.41 and 1.17 LU ha^{-1} respectively. Fertilization with the manure compost treatment was carried out at levels corresponding to 1.18, 1.53 and 1.09 LU ha^{-1} for N, P and K respectively. Average yearly nutrient inputs are given in Table 3.

Biodynamic field preparations were sprayed on the relevant plots three times per season and composting additives were added at the beginning of manure composting and when filling the slurry tanks. For a detailed description of the biodynamic preparations see Carpenter-Boggs *et al.* (2000).

The three factors – tillage, fertilization and preparations – were fully factorized. This resulted in eight treatments, each replicated four times. The 32 plots were arranged in a strip-plot design. The plot size was 12 m x 12 m, allowing the use of regular-sized farming equipment. Soil samples were taken and yields measured in an inner 8 m x 8 m parcel.

2.2 Site conditions

The soil type at the experimental site was a Stagnic Eutric Cambisol with 45% clay content (coefficient of variance (cv.) 15%) and a pH_{H2O} of 7.1 (cv. 4%). It was enriched in ammonia acetate-ETDA extractable phosphorus and potassium due to extensive application of manure from livestock (swine) in pre-study conventional management.

Before the experiment started, the field was under conventional management and had been managed organically for 7 years in accordance with European Union Regulation (EEC) No. 834/2007. Ploughing depth was 22 cm under conventional farming and 15 cm under organic farming prior to the start of the experiment. C_{org} in the ploughed soil depth was therefore distributed relatively homogenously.

The mean annual precipitation at the site was 1000 mm. In rainy periods the soil can be waterlogged for some days. Mean annual temperature was 8.9 °C.

2.3 Crops

A ley-based rotation was established (Table 1). Cereal and sunflower grains, cereal straw, the oat-clover intercrop, grass clover and silage maize were removed from the field. In the RT system only a winter pea catch crop was established before silage maize and incorporated in spring. Grain yields under RT, except for sunflower, were lower than under CT (CT: winter wheat 5.18 Mg dry matter (DM) ha^{-1}, sunflower 3.19 Mg DM ha^{-1}, spelt 2.43

Mg DM ha^{-1}; RT: winter wheat 4.43 Mg DM ha^{-1}, sunflower 3.33 Mg DM ha^{-1}, spelt 2.23 Mg DM ha^{-1}); fodder crops had higher yields under RT (CT: oat-clover intercrop 0.82 Mg DM ha^{-1}, grass clover 2006 7.51 Mg DM ha^{-1}, grass clover 2007 7.79 Mg DM ha^{-1}, silage maize 12.27 Mg DM ha^{-1}; RT: oat-clover intercrop 0.87 Mg DM ha^{-1}, grass clover 2006 9.66 Mg DM ha^{-1}, grass clover 2007 9.60 Mg DM ha^{-1}, silage maize 16.48 Mg DM ha^{-1}.

2.4 Soil sampling

Soil samples were taken at the beginning of the experiment on October 1st 2002 (after harvest of silage maize), on March 15th 2005 (standing crop: spelt) and on September 25th 2008 (after harvest of silage maize) in all 32 experimental plots. Twelve individual cores (diameter 3 cm) per field plot were separated into 0-10 cm and 10-20 cm soil depth layers and thereafter bulked to one composite sample per plot and layer. Soils were then sieved through a 5-mm mesh and kept at 3°C until they were analysed.

2.5 Chemical soil analysis

2.5.1 Measurement of pH and C_{org}

The pH of dried samples (60°C, 24 h) was measured in a soil suspension with deionized water (1:10, w/v). C_{org} was measured after wet oxidation of 1 g dry soil in 20 ml concentrated H_2SO_4 and 25 ml 2 M $K_2Cr_2O_7$ in accordance with Swiss standard protocols (FAL *et al.*, 1996).

2.5.2 Measurement of nutrient contents

Soluble nutrients P and K were extracted with CO_2-saturated water (P_{CO2}, K_{CO2}) according to Swiss standard protocols (FAL *et al.*, 1996). The plant-available exchangeable fraction of P ($P_{Aac\text{-}EDTA}$) was extracted with ammonium acetate-EDTA. Phosphate in the extract was measured after complex formation with added ammonium molybdate in a spectrophotometer at 750 nm. Available K in the ammonium acetate-EDTA extract ($K_{Aac\text{-}EDTA}$) was measured by atom absorption spectrometry at 766.5 nm (FAL *et al.*, 1996).

2.6 Soil microbial analyses

All soil microbial analyses were carried out on moist soil samples adjusted to a water content corresponding to 40-50% of maximum water retention capacity.

2.6.1 Chloroform fumigation extraction

Soil microbial biomass C (C_{mic}) and N (N_{mic}) were estimated by chloroform fumigation extraction (CFE) in accordance with Vance *et al.* (1987). CFE was done in triplicate on 20 g (dry matter) subsamples that were extracted with 80 ml of a 0.5 M K_2SO_4 solution. Total organic C (TOC) in soil extracts was determined by infrared spectrometry after combustion at 850°C (DIMA-TOC 100, Dimatec, 45276 Essen, DE). Total N was subsequently measured in the same sample by chemoluminescence (TNb, Dimatec, 45276 Essen, DE). Soil microbial biomass was then calculated according to the formula: $C_{mic} = EC / k_{EC}$ where EC = (TOC in fumigated samples – TOC in control samples) and $k_{EC} = 0.45$ (Joergensen and Mueller, 1996a). $N_{mic} = EN / k_{EN}$ where EN = (total N extracted from fumigated samples – total N extracted from control samples) and $k_{EN} = 0.54$ (Joergensen and Mueller, 1996b).

2.6.2 Soil dehydrogenase activity

Dehydrogenase activity (DHA) was measured according to Tabatabai (1982) in 5 g soil samples incubated at 30°C for 24 h in the presence of an alternative electron acceptor (triphenytetrazoliumchloride, TTC). The red-coloured product (triphenylformazan, TPF) was extracted with acetone and measured in a spectrophotometer at 546 nm.

2.7 Nutrient balances

Nutrient balances for N, P and K were calculated on a field basis. Wheat grains and straw, oat-clover intercrop, sunflower seeds, spelt grains and straw, grass-clover, winter pea and silage maize samples were analysed for nutrient concentrations. Nitrogen was determined after Kjeldahl digestion. For measuring P and K, samples were incinerated at 600°C and the ash extracted with concentrated hydrochloric acid. N and P concentrations were determined photometrically and K via atom absorption spectrometry. Nutrients (N, P, K) in slurry and manure compost were extracted with hydrochloric acid after the samples had been incinerated at 600°C and were analysed as specified above. Biological N-fixation by legumes, atmospheric deposition, leaching and gaseous emissions of nutrients were not considered in the balances. The winter pea catch crop contained 62 kg N ha^{-1} and was exclusively incorporated into the soil of the RT system. Although we assume that the whole quantity of catch crop N was not fixed biologically, we included it in the nitrogen budget.

2.8 Statistics

The statistical model we used involved Tillage as the main factor in the strip-plot design and Fertilization * Preparations as a combined factor. Soil microbial properties and nutrient contents were calculated with a general linear model to test for significance using SPSS 15.0 software (SPSS Inc., 2006). Linear contrasts were then calculated for fertilization and preparations using SAS 9.1 software (SAS Institute Inc., 2002-2003). A mixed model in SAS was used to perform a time line analysis for pH and C_{org} with the fixed factors Tillage, Fertilization*Preparations and Year (Year as a repeated measure). The Block was used as a random factor.

3. Results and discussion

After the first six-year crop rotation period of our experiment, statistical analysis revealed no effects of fertilization and only slight effects of preparations on the investigated properties while the response to tillage was strong, especially in the 0-10 cm soil depth layer. We found a distinct stratification of C_{org}, microbial biomass C and N, microbial activity, plant-available P and K under RT, while they were distributed relatively homogenously throughout 0-20 cm under CT.

3.1 pH and C_{org}

We used a mixed model ANOVA with repeated measures in 2002, 2005 and 2008 to identify significant changes of pH and C_{org} over the first crop rotation period. The factors Year in both soil layers and Tillage in the 0-10 cm soil layer showed significant effects. There was no effect of the combined factor Fertilizer*Preparations. The interaction Year*Tillage significantly affected pH and C_{org} in the 0-10 cm soil layer (Table 4).

Soil pH decreased significantly from 2002 to 2005 under both tillage treatments and in both soil depths. When compared with the initial values of 2002, pH values in 2008 were significantly lower only under RT (Table 5 & Fig. 1). The decrease of pH was highest under RT in 0-10 cm (- 0.17, $p < 0.01$).

According to Rasmussen (1999), soil acidity under RT increases in the long run by 0.2-0.3 units in topsoil, which may be due to an accumulation of organic acids in the superficial layer (Pronin, 2003). These findings are confirmed by the results of our experiment. On the other hand, CT may prevent ions from leaching by turning the soil and thus retard acidification of the topsoil (Friedel *et al.*, 1996). Seasonal differences causing the lower levels measured in spring 2005 cannot be excluded.

Under RT C_{org} in the 0-10 cm soil layer in 2008 was 19% higher ($p < 0.001$) than the initial values in 2002. This represents an increase from 2.19% C_{org} to 2.61% C_{org} within six years. C_{org} remained constant under CT (Table 5 & Fig 1). No significant differences were found in the 10-20 cm soil layer and there were no effects of fertilization or preparations.

C_{org} is considered an important indicator of soil fertility. The increase in C_{org} in our experiment in the 0-10 cm soil layer under RT measured in 2005 (Berner *et al.*, 2008) continued between 2005 and 2008. In a meta-study, Ogle *et al*. (2005) found C_{org} increased by 16% in 0-30 cm depth after 20 years of no tillage in a temperate wet climate. Alvarez (2005) found no differences in C_{org} accumulation between NT and RT. In this meta-study, the amount of C_{org} integrated over 30 cm soil depth under NT and RT was 14% higher than under CT, if only long-term experiments were taken into account. The increase in C_{org} took place only in 0-15 cm, no differences were reported below 15 cm, which corresponds to our findings. Our results are in line with Pekrun and Claupein (1998), Rasmussen (1999), Kladivko (2001) and Koch and Stockfisch (2006). Under organic farming conditions, Emmerling (2007) reported an increase in C_{org} of 7-10% in the surface layer after 10 years of RT, with no differences below the tilled layer. Other studies found an increase in C_{org} in the superficial layer but a decrease in the untilled soil layers below (Angers *et al.*, 1993; Angers *et al.*, 1997; Kay and VandenBygaart, 2002). Insufficient plant material left on the field may be a reason for the failure of RT to enhance C_{org}. By contrast, input of organic matter via crop rotation and organic fertilizers in our experiment were high compared to other RT trials (Table 2). This seems to be important, as Baker *et al*. (2007) argue that C_{org} gains in most cases are based only on near-surface samples (0-30 cm). They disappear when deeper sampling (below 30 cm) is included. Under organic farming conditions, Schulz *et al*. (2008) found no increase in C_{org} after 12 years of RT but C_{org} was altered by inducing a ley phase into the crop rotation and with amendment of manure. Many authors (Kouwenhoven and Boer, 1997; Pekrun and Claupein, 1998; Drinkwater *et al.*, 2000) argue that periodical use of mouldboard ploughing may be inevitable in organic farming to control weed problems. However, high losses of C_{org} , have been reported after single mouldboard ploughing in a RT system, proportional to the previous gain under NT or RT (Koch and Stockfisch, 2006; Conant *et al.*, 2007). Organic fertilizers, especially manure, enhance more stable fractions of C_{org} (Christensen, 1988; Wander and Traina, 1996) and thus C_{org} accumulated under RT in stocked organic farming systems may be more resistant to decomposition after mouldboard ploughing than in stockless systems. Peigné *et al*. (2007) think it likely that the combined effect of organic farming and RT could also improve the soil organic matter content and consequently soil nutrient reserves in stockless organic systems and call for further research on this issue.

3.2 Soil microbial biomass and activity

Soil microbial biomass (C_{mic}, N_{mic}) and microbial activity (DHA) were strongly stratified under RT, whereas they were relatively homogenously distributed throughout the profile under CT. Soil microbial biomass was increased under RT in the 0-10 cm soil layer, C_{mic} being 37% ($p < 0.01$) and N_{mic} 35% ($p < 0.05$) higher than under CT (Table 6). Under RT, C_{mic} was also increased by 10% ($p < 0.05$) in the 10-20 cm soil layer, whereas N_{mic} showed no significant difference between the two tillage treatments.

Despite 8% higher average values in the 10-20 cm layer, tillage effects on the C_{mic}-to-N_{mic} ratio were not significant. However, a 7% higher C_{mic}-to-N_{mic} ratio ($p < 0.05$) was found with the use of biodynamic preparations.

The C_{mic}-to-C_{org} ratio, which is considered to be an indicator of biological soil fertility (Sparling, 1992; Stockfisch *et al.*, 1999), was 14% higher ($p < 0.05$) under RT in the 0-10 cm soil layer. Microbial activity (DHA) was increased by 57% ($p < 0.05$) under RT compared to CT in the 0-10 cm soil depth. In the 10-20 cm layer DHA was significantly enhanced by 17% as compared to CT ($p < 0.05$).

Microbial biomass and activity are considered to be early indicators of changes in soil properties induced by tillage regimes (Kandeler *et al.*, 1999). A strong differentiation of the microbial biomass between tilled and untilled layers under RT was found in our experiment; this corresponds with results obtained by other authors (Alvarez *et al.*, 1995; Kandeler *et al.*, 1999; von Lützow *et al.*, 2002; Emmerling, 2007). No clear stratification of the microbial biomass was found by Angers *et al*. (1993) with a silage maize rotation and low input of organic matter. While we also found a significant increase in microbial biomass C in the 10-20 cm soil depth layer, others report no difference (Friedel *et al.*, 1996: ATP contents; Kandeler *et al.*, 1999) or less microbial biomass (Stockfisch *et al.*, 1999; Emmerling, 2007) in the untilled layer. Microbial biomass is strongly affected by freshly added organic matter (von Lützow *et al.*, 2002). Friedel *et al*. (1996) accordingly found a high dependence of microbial biomass distribution in the soil on the amount of fresh, decomposable organic matter in a tillage experiment. The high input of organic matter in the Frick trial (Table 2) combined with the reduction of tillage may be the reason for the high levels of microbial biomass C and N, even in the untilled 10-20 cm soil layer. Application of manure in our experiment – in contrast to the stockless experiment described by Emmerling (2007), which was also conducted under organic farming conditions with a ley-based crop rotation – may be a crucial factor in the dynamics of the microbial populations. Heinze *et al*. (in press) found enhanced microbial biomass as a result of application of manure.

Diversified crop rotations, reduction of tillage and adoption of organic farming are reported to result in a more fungal-dominated microbial community (Six *et al.*, 2006). A higher C_{mic}-to-N_{mic} ratio in the undisturbed 10-20 cm soil depth layer under RT in our experiment supports these findings as it indicates a higher proportion of fungi and older cells in the total microbial biomass, whereas younger cells and a bacteria-dominated microflora would be reflected in a decrease in the C_{mic}-to-N_{mic} ratio (Joergensen, 1995). Guggenberger *et al*. (1999) and Emmerling (2007) found an increase in fungi in the upper soil layer under NT and RT. This , however, was not indicated by changes in the C_{mic}-to-N_{mic} ratio in 0-10 cm soil depth in our experiment. The regular use of the rototiller may have prevented the development of fungal biomass in the tilled layer in our experiment. Additionally, in the experiment described by Emmerling (2007) the green fallow and cereal straw was mulched and remained at the field, leaving high amounts of lignin and cellulose as a favourable substrate for the fungal population.

The C_{mic}-to-N_{mic} ratio increased in our experiment with the use of biodynamic preparations in 0-10 cm. In contrast, Fließbach *et al*. (2007) found a lower C_{mic}-to-N_{mic} ratio for a treatment with compost and biodynamic preparations compared to a manured conventional system. However, they were unable to say whether this effect is caused by composting or by the biodynamic preparations. No effects of biodynamic preparations on soil biology properties were found by Carpenter-Boggs *et al*. (2000), when compared to compost without preparations.

The C_{mic}-to-C_{org} ratio in 0-10 cm under RT was 14% higher than that under CT. This difference was already apparent after three years of our trial (Berner *et al.*, 2008) and is now smaller than the differentiation by C_{org}. This confirms the results obtained by Stockfisch *et al.* (1999), who consider the C_{mic}-to-C_{org} ratio to be an early indicator of an enhancement of C_{org}. Angers *et al*. (1993) found a C_{mic}-to-C_{org} ratio three times higher under RT compared to CT in 0-16 cm after 11 years of silage maize rotation and low input of organic matter. An increase in the C_{mic}-to-C_{org} ratio of 16% in the superficial layer was also reported by Emmerling (2007).

We found significantly higher microbial activity (DHA) under RT in both soil layers. Emmerling (2007) found soil respiration and alkaline phosphomonoesterase significantly higher in 0-15 cm under RT but no difference in the soil layer below. Similar results were obtained by von Lützow *et al*. (2002) and Kandeler *et a*l. (1999). Von Lützow *et al*. (2002) report higher microbial biomass and activity in clay soils because the conditions for microorganisms are more stable, although C_{org} is less accessible to the microbial community.

3.3 Phosphorus and potassium

As with C_{org}, microbial biomass and microbial activity, a clear stratification, especially of soluble P_{CO2} and K_{CO2},was found after 6 years under RT. P_{CO2} in the 0-10 cm soil layer in 2008 was 72% higher ($p < 0.05$) than CT, while the exchangeable $P_{Aac\text{-}EDTA}$ was only 27% higher ($p < 0.05$) (Table 6). K_{CO2} was 40% ($p < 0.1$) higher than CT in the 0-10 cm layer in 2008 and $K_{Aac\text{-}EDTA}$ +23% ($p < 0.05$) higher. There were no significant tillage effects in the 10-20 cm soil layer and no effects of fertilization or preparations in both layers.

The small differences between the nutrient budgets for P and K in CT and RT cannot be the reason for the high differences in nutrient contents between the two tillage systems. The surplus of K was even higher under CT (Table 3). Yields of forage crops (grass clover and silage maize) were higher under RT; larger quantities of crop residues and root biomass were also left on the field. Rasmussen (1999) reported a significant increase in plant-available P in 0-5 cm soil depth under RT in various studies, while available P in 10-20 cm remained stable or even decreased. A stratification of plant-available P similar to C_{org} was also found by Emmerling (2005), whereas the total amount of plant-available P remained constant in 0-25 cm depth. Vu *et al*. (2009) found a concentration of plant-available P in 0-10 cm under NT. A high accumulation of C_{org} was closely related to organic P dynamics, as organic P accumulates only when C availability is high (Bünemann *et al.*, 2006). Plant-available K in the top layer increased under RT whereas there were no differences between RT and ploughed soil in 10-20 cm (Rasmussen, 1999).

3.4 Nutrient budgets

To ensure both short-term productivity and long-term sustainability, achieving a balance between inputs and outputs of nutrients within the farm system is crucial (Watson et al., 2002). As nitrogen fixation by legumes was not considered in our calculation, nitrogen budgets are clearly negative for all treatments. The deficit was -33 kg N ha^{-1} yr^{-1} in the slurry treatment compared to -42 kg N ha^{-1} yr^{-1} with application of manure compost. P input and removal were balanced, while there was a surplus in K in all treatments over the first crop rotation period (Table 3). K surplus in the slurry treatment (48 kg K ha^{-1} yr^{-1}) was twice that in the manure compost treatment (24 kg K ha^{-1} yr^{-1}).

The N-deficit in the nutrient budget was higher and the calculated surplus of K lower under RT than under CT. Removal of N and K under RT was higher, mainly due to the higher yields of grass-clover and silage maize. Phosphorus input and removal were almost balanced over the first crop rotation period for all treatments.

If biological N-fixation is considered, N-surpluses of up to 60 kg N ha^{-1} yr^{-1} (Berry *et al.*, 2003) are reported for organic farms in the UK. In a survey considering 88 European organically managed farms, Watson *et al*. (2002) found an average N surplus of 82 kg ha^{-1} yr^{-1} for dairy farms.

Stockless organic farms show P-deficits, while farms with livestock can compensate by importing P in additional feed and bedding material (Watson *et al.*, 2002; Berry *et al.*, 2003). Negative balances of P for organic dairy farms in Norway were reported by Steinshamn *et al*. (2004). P deficits were also reported by Emmerling (2005) in a stockless trial with RT under organic farming conditions. We found no deficit of P in our experiment, which may be explained by relatively high inputs of P as compared to N and K, due to relatively low element contents for N and K in the applied manures.

K budgets calculated for organic crop rotations show both surpluses and deficits (Watson *et al.*, 2002). Rotations with large return of manure had K surpluses or balanced K budgets, which is also the case in our experiment.

To summarize, our results confirm the current data: stratification of C_{org}, microbial biomass, microbial activity and soil nutrients were often observed after adoption of RT. In the 10-20 cm soil layer, we found no differences in C_{org} and soil nutrients between the two tillage systems. Interestingly, soil microbial biomass C and microbial activity (DHA) were also higher in the untilled layer of RT. We found no similar results in the literature and assume that comparatively high inputs of organic material via crop rotation and manure are important factors in our experiment. The goal of current research is to target the role of soil types and of clay minerals in particular and especially the hydraulic dynamics and aeration of tillage systems.

4. Conclusions

After the first crop rotation period of six years, only tillage provoked significant responses in soil fertility indicators. Hardly any effects of the fertilization treatments and the use of biodynamic preparations were observed. We found a strong stratification of C_{org}, microbial biomass, microbial activity (DHA) and soil nutrients such as P and K in the RT tillage system. Enhancement of these properties in the superficial soil layer under RT lead to the conclusion that RT is a suitable method for increasing soil fertility in organic farming systems. Average yields during the six years under RT in our experiments were 11% higher than those obtained under CT. In conclusion, our RT system has demonstrated its capacity to provide a balanced performance with respect to several ecological services of agroecosystems, such as primary production, maintenance of natural resources and soil fertility, nutrient supply and support of high biodiversity (Björklund *et al.*, 1999).

The results presented here reflect the situation after six years of RT under organic farming conditions. In their review, Kay and VandenBygaart (2002) found results of changes in C_{org} obtained by different investigators to be most consistent when measurements were made more than 15 years after initiating the tillage trial. Further development of soil fertility indicators needs to be assessed, also with respect to carbon sequestration in an organic farming system with diversified ley-based crop rotation and organic fertilization, as carbon sequestration of RT systems is still a matter of controversy (Baker *et al.*, 2007). Further research on the combined effects of organic farming and RT on this issue is necessary.

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Table 1
Dates of soil tillage in the different tillage systems

System	Crop	Tillage	Date
Conventional	Winter Wheat	Plough	11 October 2002
		Rototiller	30 October 2002
	Intercrop – Oat-Clover	Rototiller	19 August 2003
	Sunflower	Plough	26 February 2004
		Rototiller	22 April 2004
	Spelt	Plough	8 November 2004
		Rototiller	16 November 2004
	Clover-Grass	Rototiller	13 August 2005
	Silage Maize	Plough	25 February 2008
		Rototiller	9 May 2008
Reduced	Winter Wheat	Rototiller	30 October 2002
		Chisel	6 August 2003
	Intercrop – Oat Clover	Rototiller	19 August 2003
	Sunflower	Rototiller	22 April 2004
	Spelt	Rototiller	16 November 2004
	Clover-Grass	Rototiller	13 August 2005
	Catch crop – Winter Pea	Stubble Cleaner	14 September 2007
		Chisel	15 September 2007
		Rototiller	11 October 2007
	Silage Maize	Stubble Cleaner	9 May 2008
		Rototiller	9 May 2008

Table 2
Organic matter (OM) input (manure, slurry and green manure) in the first crop rotation period (2003-2008) (t OM ha^{-1})

Year	Crop	Tillage		Fertilization		Preparations	
		Conventional	Reduced	Slurry	Manure Compost	Without	With
2003	Wheat	2.41	2.41	2.07	2.75	2.45	2.37
2004	Sunflower	0.94	0.94	0.72	1.17	0.90	0.98
2005	Spelt	1.52	1.80	1.08	2.24	1.69	1.64
2006	Clover-Grass	2.29	2.31	2.28	2.32	2.35	2.25
2007	Clover-Grass	1.33	1.33	1.38	1.28	1.34	1.32
2008	Silage Maize	0.64	2.39	0.48	0.80	0.62	0.67
Total		9.13	11.18	8.01	10.6	9.35	9.23
Average yearly Input		1.52	1.86	1.33	1.76	1.56	1.54

Silage Maize 2008: RT including pea green manure; OM: organic matter

Table 3
Nutrient budgets on a field basis for nitrogen (N), phosphorus (P) and potassium (K) for the first crop rotation period (2003-2008)

	N (kg ha^{-1} yr^{-1}) #		P (kg ha^{-1} yr^{-1})		K (kg ha^{-1} yr^{-1})	
Tillage	Conventional	Reduced	Conventional	Reduced	Conventional	Reduced
Input	102	116	26	26	158	165
Yield	130	153	25	26	116	136
Surplus	-27	-37	1	0	43	30
Fertilization	Slurry	Manure Compost	Slurry	Manure Compost	Slurry	Manure Compost
Input	107	101	24	28	172	152
Yield	140	143	25	25	123	128
Surplus	-33	-42	-1	2	48	24
Preparations	Without	With	Without	With	Without	With
Input	106	102	26	26	163	161
Yield	142	141	25	25	126	125
Surplus	-36	-38	1	1	37	36

Values presented are annual means. # biological nitrogen fixation not considered

Table 4
F-values and significance levels of the mixed-model with repeated measures in 2002, 2005 and 2008 for pH_{H20} and soil organic carbon (C_{org})

	pH_{H20}				C_{org}			
	0-10 cm		10-20 cm		0-10 cm		10-20 cm	
Tillage	19.94	*	0.69		84.01	**	5.35	
Fertilization*Preparations	1.61		0.89		1.77		0.7	
Tillage*Fertilization*Preparations	1.18		0.58		0.52		1.76	
Year	32.54	***	17.53	**	37.91	***	4.67	(*)
Year*Tillage	8.33	*	0.22		22.82	**	0.47	
Year*Fertilization*Preparations	2.13	(*)	0.36		1.04		0.24	
Year*Tillage*Fertilization*Preparations	2.35	(*)	1.85		2.24	(*)	2.21	(*)

(*) p < 0.1; * p < 0.05; ** p < 0.01; *** p < 0.001

Table 5
Means for pH and soil organic carbon (C_{org}) in 2002, 2005 and 2008 in soil depth layers 0-10 cm and 10-20 cm (relative values to 2002=100 for C_{org} 2005 and 2008 in parentheses). Results of the mixed model t-test for Year*Tillage, indicating significant differences of the means of each treatment in 2005 and 2008 compared with the corresponding means in 2002.

	pH_{H20} 2002		pH_{H20} 2005		pH_{H20} 2008		C_{org} 2002 (%)		C_{org} 2005 (%)		C_{org} 2008 (%)	
	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm
Tillage												
Conventional	7.62	7.60	7.42	7.43	7.55	7.52	2.11	2.05	2.22 (105)	2.19 (107)	2.16 (102)	2.13 (104)
Reduced	7.58	7.59	7.35	7.39	7.41	7.47	2.19	2.16	2.46 (112)	2.28 (106)	2.61 (119)	2.17 (100)
Fertilization												
Slurry	7.62	7.62	7.43	7.44	7.51	7.52	2.12	2.09	2.30 (108)	2.21 (106)	2.33 (110)	2.11 (101)
Manure Compost	7.59	7.57	7.35	7.38	7.46	7.47	2.19	2.13	2.38 (109)	2.26 (106)	2.44 (112)	2.19 (103)
Preparations												
Without	7.65	7.61	7.39	7.42	7.50	7.52	2.13	2.11	2.34 (109)	2.24 (106)	2.41 (113)	2.16 (103)
With	7.56	7.57	7.38	7.40	7.47	7.47	2.17	2.10	2.34 (108)	2.23 (106)	2.36 (109)	2.14 (102)
ANOVA Year*Tillage												
Conventional	-	-	***	***	(*)	n.s.	-	-	*	(*)	n.s.	n.s.
Reduced	-	-	***	**	**	*	-	-	***	(*)	***	n.s.

(*) p < 0.1; * p < 0.05; ** p < 0.01; *** p < 0.001

Table 6
Means of soil microbial biomass C_{mic} and N_{mic}, C_{mic}-to-N_{mic} ratio, C_{mic} to C_{org} (%) and dehydrogenase activity (DHA) in the soil depth layers 0-10 cm and 10-20 cm in 2008, ANOVA for the main effects Tillage and Fertilization*Preparations, linear contrasts for Fertilization and Preparations

	C_{mic} (mg C_{mic} kg^{-1})		N_{mic} (mg N_{mic} kg^{-1})		C_{mic} to N_{mic}		C_{mic} to C_{org} (%)		DHA (µg TPF g^{-1} d^{-1})	
	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm
Tillage										
Conventional	764	794	113	125	6.80	6.38	3.58	3.78	315	328
Reduced	1049	869	153	126	6.94	6.91	4.07	4.05	495	382
Fertilization										
Slurry	900	828	133	125	6.85	6.67	3.89	3.96	404	360
Manure Compost	913	835	134	126	6.89	6.62	3.76	3.87	406	350
Preparations										
Without	914	837	138	127	6.64	6.59	3.82	3.93	418	359
With	899	826	129	124	7.10	6.70	3.83	3.91	392	352
Tillage										
Reduced (%) (100% = Conventional)	137	110	135	101	102	108	114	107	157	117
Fertilization										
Manure Compost (%) (100% = Slurry)	101	101	101	101	101	99	97	98	101	97
Preparations										
With (%) (100% = Without)	98	99	94	98	107	102	100	99	94	98
ANOVA		#								
Tillage	**	*	*	n.s.	n.s.	(*)	*	(*)	*	*
Fertilization*Preparations	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Fertilization	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Preparations	n.s.	n.s.	(*)	n.s.	*	n.s.	n.s.	n.s.	n.s.	n.s.

C_{mic}, soil microbial carbon; N_{mic}, soil microbial nitrogen; C_{org}, soil organic carbon; DHA, dehydrogenase activity; TPF – triphenylformazan
ANOVA for C_{mic} in 10-20 cm depth was calculated with C_{mic}* C_{mic}
(*) $p < 0.1$; * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$

Table 7
Means of nutrient contents in the soil depth layers 0-10 cm and 10-20 cm in 2008, ANOVA for the main factors Tillage and Fertilization*Preparations, linear contrasts for Fertilization and Preparations

	P_{CO2} (mg kg^{-1})		$P_{Aac\text{-}EDTA}$ (mg kg^{-1})		K_{CO2} (mg kg^{-1})		$K_{Aac\text{-}EDTA}$ (mg kg^{-1})	
	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm
Tillage								
Conventional	1.83	1.30	107	108	31,7	26,6	454	438
Reduced	3.15	1.39	136	111	44,5	25,2	559	427
Fertilization								
Slurry	2.38	1.39	119	111	38,2	26,4	500	425
Manure Compost	2.60	1.30	124	107	38,0	25,4	512	440
Preparations								
Without	2.58	1.31	125	107	37,1	26,0	505	429
With	2.40	1.38	119	112	39,1	25,8	507	436
Tillage								
Reduced (%) (100=Conventional)	172	107	127	103	140	95	123	97
Fertilization								
Manure Compost (%) (100=Slurry)	109	93	104	96	100	96	102	104
Preparations								
With (%) (100=Without)	93	105	95	104	105	99	100	102
ANOVA								
Tillage	*	n.s.	*	n.s.	(*)	n.s.	*	n.s.
Fertilizer*Preparations	n.s.	n.s.	n.s.	n.s.	(*)	n.s.	n.s.	n.s.
Fertilization	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Preparations	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.

(*) $p < 0.1$; * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$

P_{CO2},CO_2-extractable phosphorus; K_{CO2}, CO_2-extractable potassium

$P_{Aac\text{-}EDTA}$, ammonium acetate-extractable phosphorus; $K_{Aac\text{-}EDTA}$, ammonium acetate-extractable potassium

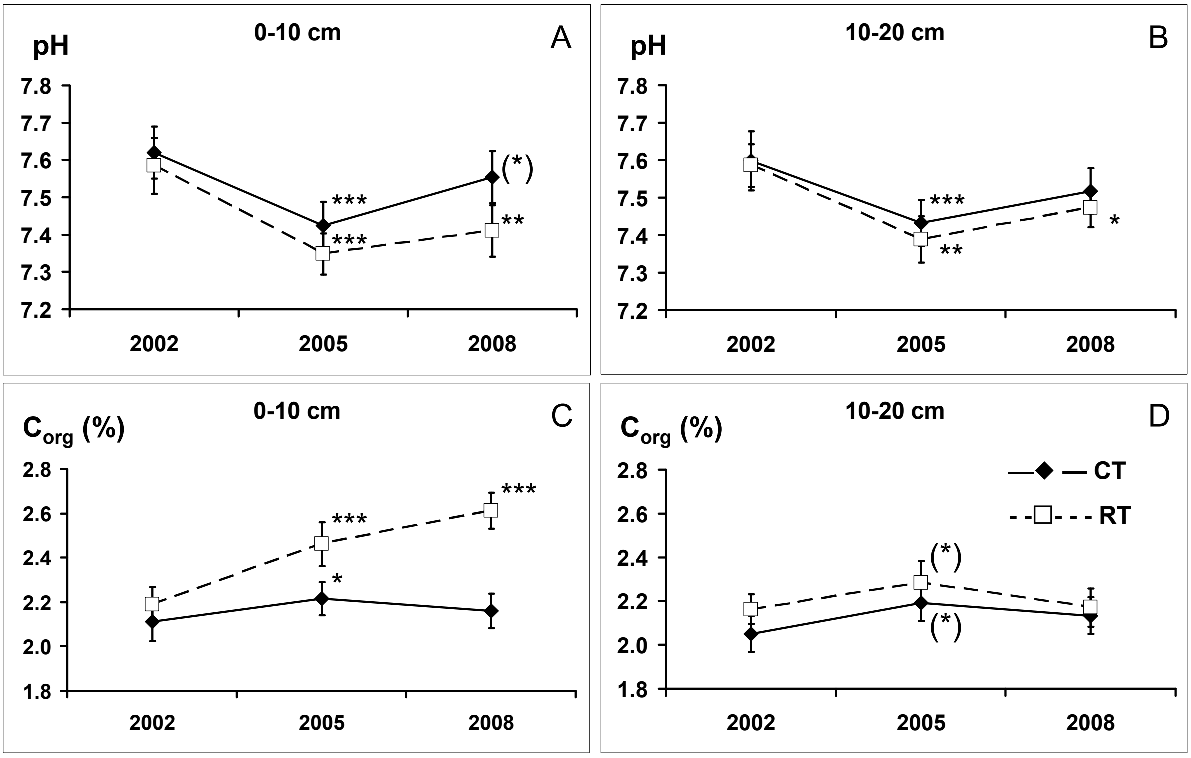
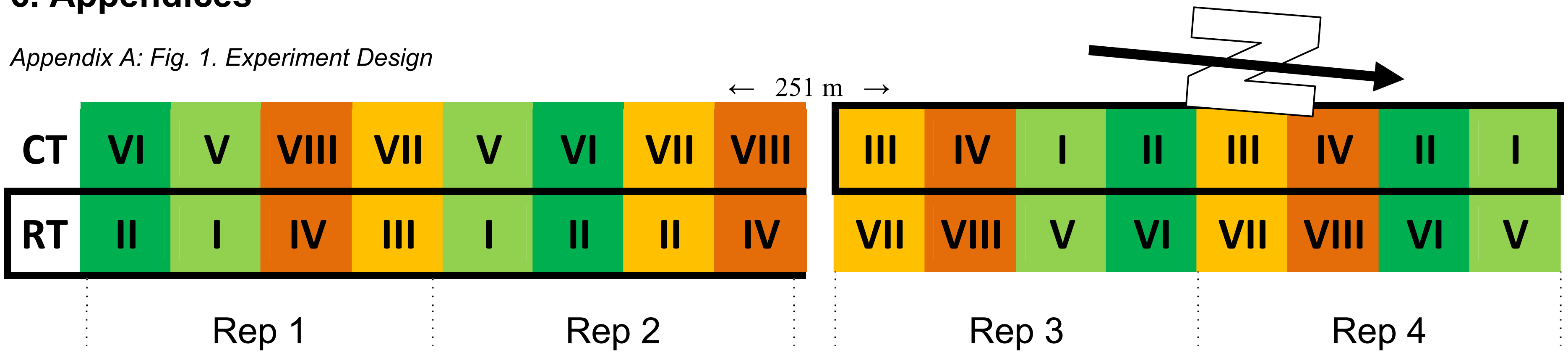


Fig. 1. Means and standard error of the mean of pHH20 and soil organic carbon (Corg) in 0-10 cm and 10-20 cm soil depth for Reduced Tillage (RT) and Conventional Tillage (CT) in the years 2002, 2005 and 2008.

A, B: pH. C, D: Soil Organic Carbon (C_{org} (%)). Results of the mixed model t-test for the factor Year*Tillage, stars indicate significant differences of the means of each treatment in 2005 and 2008 compared with the corresponding mean in 2002. (*) $p < 0.1$, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

6. Appendices

Appendix A: Fig. 1. Experiment Design



8 Treatments x 4 Replications = 32 Plots
Plot Size 12 x 12 m

I	Conventional Tillage*Manure Compost*Without Preparations
II	Conventional Tillage*Manure Compost*With Preparations
III	Conventional Tillage*Slurry*Without Preparations
IV	Conventional Tillage*Slurry*With Preparations
V	Reduced Tillage*Manure Compost*Without Preparations
VI	Reduced Tillage*Manure Compost*With Preparations
VII	Reduced Tillage*Slurry*Without Preparations
VIII	Reduced Tillage*Slurry*With Preparations

Appendix B: Detailed Nutrient Budgets

Appendix: Table 1
Nutrient budgets of different Tillage systems on a field basis for nitrogen (N), phosphorus (P) and potassium (K) for the first crop rotation period (2003-2008). Input and Output (Yields) per year.

	N ($kg\ ha^{-1}$) #		P ($kg\ ha^{-1}$)		K ($kg\ ha^{-1}$)	
Tillage	Conventional	Reduced	Conventional	Reduced	Conventional	Reduced
Yields						
2003 wheat - grain	110	97	22	19	22	18
2003 wheat - straw	21	18	4	4	53	48
oat-clover intercrop	34	37	5	6	40	43
2004 sunflower grain	117	123	18	19	26	27
2005 spelt - grain	48	48	13	12	11	9
2005 spelt – straw & spelt	14	15	5	6	32	39
2006 grass clover	182	202	32	33	214	229
2007 grass clover	140	190	25	30	197	269
2008 silage maize	111	191	25	28	100	131
Total yield 2003-2008	778	920	148	156	695	814
Input						
2003 wheat	134	134	50	50	251	251
2004 sunflower	69	69	18	18	90	90
2005 spelt	126	146	27	31	162	200
2006 grass clover	119	122	31	31	214	218
2007 grass clover	114	114	21	21	164	164
2008 silage maize	52	114	7	7	69	69
Total input 2003-2008	614	699	154	158	951	993
-Total yield 2003-2008	778	920	148	156	695	814
Total surplus 2003-2008	-164	-222	6	2	255	179
Input per year	106	102	26	26	163	161
Yield per year	142	141	25	25	126	125
Surplus per year	-36	-38	1	1	37	36

biological nitrogen fixation not considered

Appendix: Table 2
Nutrient budgets of different Fertilization systems on a field basis for nitrogen (N), phosphorus (P) and potassium (K) for the first crop rotation period (2003-2008). Input and Output (Yields) per year.

	N ($kg\ ha^{-1}$) #		P ($kg\ ha^{-1}$)		K ($kg\ ha^{-1}$)	
Fertilization	Slurry	Manure Compost	Slurry	Manure Compost	Slurry	Manure Compost
Yield						
2003 wheat - grain	116	91	22	18	22	18
2003 wheat - straw	21	18	4	4	58	44
oat-clover intercrop	35	35	5	5	40	42
2004 sunflower grain	121	119	18	18	27	26
2005 spelt - grain	47	49	13	12	10	10
2005 spelt - straw+spelt	14	15	5	5	33	37
2006 grass clover	176	208	30	35	200	243
2007 grass clover	156	175	27	28	230	235
2008 silage maize	154	148	27	26	119	113
Total yield 2003-2008	840	858	153	151	740	769
Output						
2003 wheat	127	141	53	48	237	266
2004 sunflower	58	79	9	26	66	115
2005 spelt	98	174	18	41	147	215
2006 grass clover	160	80	39	24	292	141
2007 grass clover	139	90	23	19	204	124
2008 silage maize	60	44	4	9	84	54
Total input 2003-2008	642	609	146	166	1.030	914
-Total yield 2003-2008	840	858	153	151	740	769
Total surplus 2003-2008	-198	-249	-6	15	290	145
Input per year	107	101	24	28	172	152
Yield per year	140	143	25	25	123	128
Surplus per year	-33	-42	-1	2	48	24

biological nitrogen fixation not considered

Appendix: Table 3
Nutrient budgets of different Preparation treatments on a field basis for nitrogen (N), phosphorus (P) and potassium (K) for the first crop rotation period (2003-2008). Input and Output (Yields) per year.

	N (kg ha^{-1}) #		P (kg ha^{-1})		K (kg ha^{-1})	
Preparations	Without	With	Without	With	Without	With
Yield						
2003 wheat - grain	106	101	21	20	21	20
2003 wheat - straw	19	19	4	4	50	52
oat-clover intercrop	36	35	5	5	42	40
2004 sunflower grain	122	118	19	18	27	26
2005 spelt - grain	48	48	13	12	10	10
2005 spelt - straw+spelt	15	14	5	5	35	35
2006 grass clover	194	189	32	32	222	220
2007 grass clover	162	168	28	28	235	231
2008 silage maize	150	152	26	27	116	115
Total yield 2003-2008	852	844	153	151	759	748
Input						
2003 wheat	136	132	51	50	253	249
2004 sunflower	69	69	17	19	87	93
2005 spelt	137	135	30	28	183	179
2006 grass clover	121	120	31	32	216	216
2007 grass clover	120	109	21	21	167	161
2008 silage maize	55	49	6	7	71	67
Total input 2003-2008	637	614	156	157	978	965
-Total yield 2003-2008	852	844	153	151	759	748
Total surplus 2003-2008	-215	-231	3	5	220	216
Input per year	106	102	26	26	163	161
Yield per year	142	141	25	25	126	125
Surplus per year	-36	-38	1	1	37	36

biological nitrogen fixation not considered

*Appendix C: Mixed Model t-test results for the factors Year*Tillage and Year*Fertilization*Preparations*

Appendix: Table 4
Results of mixed model t-test for Year*Tillage and Year*Fertilization*Preparations, indicating significant differences of the means of each treatment in 2005 and 2008 compared with the corresponding means in 2002

	pH_{H20} 2002		pH_{H20} 2005		pH_{H20} 2008		C_{org} 2002 (%)		C_{org} 2005 (%)		C_{org} 2008 (%)	
	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm
ANOVA Year*Tillage												
Conventional	-	-	***	***	(*)	n.s.	-	-	*	(*)	n.s.	n.s.
Reduced	-	-	***	**	**	*	-	-	***	(*)	***	n.s.
ANOVA Year*Fertilization*Preparations												
Slurry, Without Preparations			***	***	n.s.	*			***	*	***	n.s.
Slurry, With Preparations			***	***	***	*			**	(*)	**	n.s.
Manure Compost, Without P.			***	***	**	*			***	*	***	n.s.
Manure Compost, With P.			***	**	**	*			***	*	***	n.s.

(*) p < 0.1; * p < 0.05; ** p < 0.01; *** p < 0.001

Appendix D: Means of pH and C_{org} of each treatment in 2002, 2005 and 2008 (compare Appendix A).

Appendix: Table 5
Means for pH and soil organic carbon (C_{org}) in 2002 in soil depth layers 0-10 cm and 10-20 cm

		pH_{H20} 2002		C_{org} 2002 (%)	
Treatment		0-10 cm	10-20 cm	0-10 cm	10-20 cm
I	CT*MC*P-	7.59	7.59	2.08	2.06
II	CT*MC*P+	7.52	7.46	2.28	2.16
III	CT*SL*P-	7.70	7.65	2.09	2.02
IV	CT*SL*P+	7.61	7.60	2.09	2.05
V	RT*MC*P-	7.64	7.58	2.24	2.23
VI	RT*MC*P+	7.53	7.54	2.23	2.15
VII	RT*SL*P-	7.66	7.63	2.12	2.12
VIII	RT*SL*P+	7.51	7.59	2.16	2.15

CT: Conventional Tillage, RT: Reduced Tillage
SL: Slurry, MC: Manure Compost
P-: Without Preparations, P+: With Preparations

Appendix: Table 6
Means for pH and soil organic carbon (C_{org}) in 2005 in soil depth layers 0-10 cm and 10-20 cm

		pH_{H20} 2005		C_{org} 2005 (%)	
Treatment		0-10 cm	10-20 cm	0-10 cm	10-20 cm
I	CT*MC*P-	7.37	7.41	2.25	2.24
II	CT*MC*P+	7.40	7.40	2.22	2.21
III	CT*SL*P-	7.48	7.51	2.18	2.10
IV	CT*SL*P+	7.45	7.42	2.20	2.22
V	RT*MC*P-	7.31	7.34	2.50	2.31
VI	RT*MC*P+	7.31	7.38	2.55	2.28
VII	RT*SL*P-	7.41	7.43	2.41	2.33
VIII	RT*SL*P+	7.37	7.41	2.39	2.22

CT: Conventional Tillage, RT: Reduced Tillage
SL: Slurry, MC: Manure Compost
P-: Without Preparations, P+: With Preparations

Appendix: Table 7
Means for pH and soil organic carbon (C_{org}) in 2008 in soil depth layers 0-10 cm and 10-20 cm

		pH_{H20} 2008		C_{org} 2008 (%)	
Treatment		0-10 cm	10-20 cm	0-10 cm	10-20 cm
I	CT*MC*P-	7.52	7.48	2.19	2.16
II	CT*MC*P+	7.52	7.48	2.27	2.16
III	CT*SL*P-	7.63	7.60	2.11	2.09
IV	CT*SL*P+	7.56	7.52	2.07	2.13
V	RT*MC*P-	7.44	7.51	2.69	2.28
VI	RT*MC*P+	7.35	7.43	2.60	2.18
VII	RT*SL*P-	7.41	7.48	2.64	2.13
VIII	RT*SL*P+	7.44	7.47	2.51	2.11

CT: Conventional Tillage, RT: Reduced Tillage
SL: Slurry, MC: Manure Compost
P-: Without Preparations, P+: With Preparations

Appendix E: Means of soil nutrients P and K and microbial properties and in soil depth layers 0-10 cm and 10-20 cm for all 8 treatments.

Appendix: Table 8

Means of nutrient contents in the soil depth layers 0-10 cm and 10-20 cm in 2008

		P_{CO2} (mg kg^{-1})		$P_{Aac-EDTA}$ (mg kg^{-1})		K_{CO2} (mg kg^{-1})		$K_{Aac-EDTA}$ (mg kg^{-1})	
		0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm
I	CT*MC*P-	1.93	1.32	111.85	109.52	33.65	27.71	478.88	448.41
II	CT*MC*P+	1.80	1.14	102.09	97.75	31.08	24.31	456.60	446.67
III	CT*SL*P-	1.95	1.20	110.76	104.66	30.75	27.38	432.73	418.58
IV	CT*SL*P+	1.62	1.53	105.69	121.84	31.71	27.35	446.86	438.66
V	RT*MC*P-	3.60	1.46	145.45	113.77	44.63	24.44	563.76	428.99
VI	RT*MC*P+	3.04	1.26	138.83	109.98	43.29	25.46	550.21	436.06
VII	RT*SL*P-	2.81	1.26	131.94	102.64	39.90	24.79	546.43	420.30
VIII	RT*SL*P+	3.09	1.56	130.58	118.60	50.85	26.52	575.32	422.41

P_{CO2},CO_2-extractable phosphorus; K_{CO2}, CO_2-extractable potassium

$P_{Aac-EDTA}$, ammonium-acetate-extractable-phosphorus; $K_{Aac-EDTA}$, ammonium-acetate-extractable potassium

Appendix: Table 9

Means of soil microbial biomass C_{mic} and N_{mic}, C_{mic}-to-N_{mic} ratio, C_{mic} to C_{org} (%) and dehydrogenase activity (DHA) in the soil depth layers 0-10 cm and 10-20 cm in 2008

		C_{mic} (mg C_{mic} kg^{-1})		N_{mic} (mg N_{mic} kg^{-1})		C_{mic} to N_{mic}		C_{mic} to C_{org} (%)		DHA (µg TPF g^{-1} d^{-1})	
Treatment		0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm
I	CT*MC*P-	768	816	120	129	6.42	6.33	3.54	3.90	318	310
II	CT*MC*P+	755	800	108	125	7.08	6.40	3.37	3.77	315	345
III	CT*SL*P-	773	774	118	123	6.60	6.31	3.72	3.75	323	337
IV	CT*SL*P+	758	784	108	122	7.12	6.50	3.71	3.72	304	320
V	RT*MC*P-	1090	881	161	129	6.80	6.86	4.11	3.90	532	384
VI	RT*MC*P+	1038	841	146	123	7.27	6.90	4.03	3.93	459	361
VII	RT*SL*P-	1023	876	152	128	6.75	6.88	3.93	4.17	499	404
VIII	RT*SL*P+	1046	878	153	127	6.94	6.98	4.23	4.21	489	380

C_{mic}, soil microbial carbon; N_{mic}, soil microbial nitrogen; C_{org}, soil organic carbon; DHA, dehydrogenase activity; TPF – triphenylformazan