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journal homepage: www.elsevier.com/locate/apsoil

Influence of mouldboard plough and rotary harrow tillage on microbial biomass and nutrient stocks in two long-term experiments on loess derived Luvisols

Stefanie Heinze^{a,∗}, Rolf Rauber^b, Rainer Georg Joergensen^a

^a Department of Soil Biology and Plant Nutrition, University of Kassel, Nordbahnhofstr. 1a, 37213 Witzenhausen, Germany ^b Department of Crop Sciences, University of Göttingen, Von-Siebold-Str. 8, 37075 Göttingen, Germany

article info

Article history: Received 6 May 2010 Received in revised form 1 September 2010 Accepted 29 September 2010

Keywords: Rotary harrow Mouldboard ploughing Microbial biomass Ergosterol Basal respiration

ABSTRACT

The nutrient-specific effects of tillage on microbial activity (basal respiration), microbial biomass (C, N, P, S) indices and the fungal cell-membrane component ergosterol were examined in two long-term experiments on loess derived Luvisols. A mouldboard plough (30 cm tillage depth) treatment was compared with a rotary harrow (8 cm tillage depth) treatment over a period of approximately 40 years. The rotary harrow treatment led to a significant 8% increase in the mean stocks of soil organic C, 6% of total N and 4% of total P at 0–30 cm depth compared with the plough treatment, but had no main effect on the stocks of total S. The tillage effects were identical at both sites, but the differences between the sites of the two experiments were usually stronger than those between the two tillage treatments. The rotary harrow treatment led to a significant increase in the mean stocks of microbial biomass $C (+18%)$, N $(+25%)$, and P (+32%) and to a significant decrease in the stocks of ergosterol (−26%) at 0–30 cm depth, but had no main effect on the stocks of microbial biomass S or on the mean basal respiration rate. The mean microbial biomass C/N (6.4) and C/P (25) ratios were not affected by the tillage treatments. In contrast, the microbial biomass C/S ratio was significantly increased from 34 to 43 and the ergosterol-to-microbial biomass C ratio significantly decreased from 0.20% to 0.13% in the rotary harrow in comparison with the plough treatment. The microbial biomass C-to-soil organic C ratio varied around 2.1% in the plough treatment and declined from 2.6% at 0–10 cm depth to 2.0 at 20–30 cm depth in the rotary harrow treatment. The metabolic quotient $qCO₂$ revealed exactly the inverse relationships with depth and treatment to the microbial biomass C-to-soil organic C ratio. Rotary harrow management caused a reduction in the microbial turnover in combination with an improved microbial substrate use efficiency and a lower contribution of saprotrophic fungi to the soil microbial community. This contrasts the view reported elsewhere and points to the need for more information on tillage-induced shifts within the fungal community in arable soils.

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1. Introduction

Tillage is one of the major tools for farmers to control the growth of crops and weeds and to regulate the microbial process of plant residue decomposition and the resulting nutrient release ([Dick, 1992\).](#page--1-0) A reduction in tillage intensity has often been discussed in terms of energy saving, increasing rainfall infiltration, thus reducing erosion, and enhancing C sequestration to reduce CO2 emissions [\(Frede et al., 1994; Paustian et al., 2000\).](#page--1-0)Mouldboard ploughing is still the most widespread method used in humid Central Europe ([Derpsch, 1998; Holland, 2004\),](#page--1-0) especially in organic

E-mail address: heinze@uni-kassel.de (S. Heinze).

farming [\(Kouwenhoven et al., 2002\).](#page--1-0) This type of ploughing can be replaced in the order of decreasing intensity by rotary cultivators [\(Meyer et al., 1996; Ahl et al., 1998\),](#page--1-0) rotary harrows [\(Stockfisch](#page--1-0) [et al., 1999; Jacobs et al., 2009\)](#page--1-0) or grubbers [\(Berner et al., 2008\).](#page--1-0) Most studies focussed on soil organic C and microbial biomass C ([Stockfisch et al., 1999; Wright et al., 2008\)](#page--1-0) as indicators for tillage effects, sometimes in combination with total N and microbial biomass N [\(Jacobs et al., 2009\).](#page--1-0) Information on tillage-specific effects on total P and microbial biomass P are rare [\(Saffigna et al.,](#page--1-0) [1989; Meyer et al., 1996\),](#page--1-0) whereas that on total S and microbial biomass S is entirely lacking.

Microbial biomass C and microbial biomass N are closely related in C-limited agricultural systems ([Dilly et al., 2003; Joergensen](#page--1-0) [and Emmerling, 2006\)](#page--1-0) where N rarely limits microbial growth [\(Joergensen and Mueller, 1996\).](#page--1-0) Microbial biomass P and microbial biomass S are less intimately connected with microbial biomass C [\(Heinze et al., 2010\).](#page--1-0) One reason is that different storage com-

[∗] Corresponding author. Present address: Department of Environmental Chemistry, University of Kassel, Nordbahnhofstr. 1a, 37213 Witzenhausen, Germany. Tel.: +49 5542 98 1575.

^{0929-1393/\$ –} see front matter © 2010 Elsevier B.V. All rights reserved. doi:[10.1016/j.apsoil.2010.09.011](dx.doi.org/10.1016/j.apsoil.2010.09.011)

ponents, such as polyphosphates and teichoic acids for P ([Grant,](#page--1-0) [1979; Nielsen et al., 2002\),](#page--1-0) are known to be affected by fertilizer management and the ratio of fungi to bacteria [\(Heinze et al., 2010\).](#page--1-0) Saprotrophic fungi, which are the principal decomposers of organic material in soil, especially of lignin ([Lindahl et al., 2007\),](#page--1-0) have the apparent ability to store large amounts of S in their biomass [\(Saggar](#page--1-0) [et al., 1981; Banerjee and Chapman, 1996\).](#page--1-0) As the tillage intensity is reported to have a strong impact on soil fungi [\(Ahl et al., 1998\),](#page--1-0) the analysis of microbial biomass S would give additional information on tillage induced nutrient turnover.

In Göttingen (Lower Saxony, Germany), we had the unique opportunity to investigate two long-term tillage experiments on loess-derived Luvisols, which have been comparing mouldboard ploughing with a combination of rotary harrow and rototiller over a period of around 40 years. This enabled us to examine the nutrient-specific effects of these two tillage systems on microbial activity (basal respiration), microbial biomass (C, N, P, S) indices and the fungal cell-membrane component ergosterol. The underlying hypothesis was that the promotion of fungi by a reduction in tillage intensity will specifically increase P and S storage within the microbial biomass and increase C sequestration by improving substrate use efficiency.

2. Materials and methods

2.1. Experimental site and investigation design

Two tillage systems were investigated at two research sites (Garte and Hohes Feld) close to Göttingen (Lower Saxony, Germany). The experiment at the site Garte (51°29′ N, 9°56′ E, 163 m ASL) was established in 1970 and that at the site Hohes Feld (51◦37 N, 9°53′ E, 151 m ASL) in 1967. The average mean annual precipitation is 645 mm with a mean annual temperature of 8.7 ◦C. The soil type at the two sites was characterized as Haplic Luvisol [\(FAO-](#page--1-0)WRB, [2006\) d](#page--1-0)erived from loess. The mean texture at 0–30 cm depth was 15.1% clay, 72.7% silt, and 12.2% sand at the site Garte ([Ehlers](#page--1-0) [et al., 2000; Reiter et al., 2002\)](#page--1-0) and 17.2% clay, 66.4% silt, and 16.4% sand at the site Hohes Feld ([De Mol, 1996\).](#page--1-0) The mean soil $pH-H_2O$ was 7.7 at the site Garte and 7.5 at the site Hohes Feld. The soils had been mouldboard ploughed for many years before the experiments started.

The main tillage treatments were carried out in spring. The tillage treatments were similar at both sites with regular mouldboard ploughing down to 25–30 cm depth followed by seedbed preparation with a rotary harrow and shallow cultivation down to 6–8 cm depth with a rotary harrow for seedbed preparation ([Ehlers](#page--1-0) [et al., 2000; Reiter et al., 2002\).](#page--1-0) At the site Garte, the experimental design was a randomised complete block design with four replicate plots (20 m \times 40 m). At the site Hohes Feld, a split plot design with three replicate plots (12.8 m \times 36 m) was established due to a smaller dimension of the field. The crop rotation was based on cereals in the long-term and identical at both sites for the past 20 years [\(Reiter et al., 2002\).](#page--1-0) Before this time, the crop rotation at the site Hohes Feld consisted of turnip, wheat, barley, and oats, while at the site Garte oil-seed rape, wheat, barley, rye, and oats were cropped. Additionally, the green manure crops at both sites were rape, oil radish or mustard until 1978/79, while in 1986 and 2005 mustard was only sown on the site Garte. However, in the four years before soil sampling, the crop rotation was balanced between cereals and legumes, with peas (Pisum sativum L.), winter wheat (Triticum aestivum L.), maize (Zea mays L.) and broad beans (Vicia faba L.) at both sites. Since the beginning of this experiment, fertilization was conducted by N addition in accordance with soil nutrient status. Thus, all fields of both sites were fertilized with 164 kg N ha^{-1} a^{-1} on average over the past 10 years, with the

exception of the cultivated legumes. Additionally, 60 kg K ha⁻¹ and 9 kg Mg ha−¹ fertilizers were applied to both sites in 2004 and 2005, with 169 kg K ha⁻¹ and 25 kg Mg ha⁻¹ added in 2006. The site Hohes Feld was fertilized with 46 kg P ha−¹ in 2004 and 2005, whereas the site Garte has not received any P fertilizer since 1997. Since 1987/88, the plant residues of all plots at both sites were chopped and incorporated into the first 3 cm of soil by a rotary cultivator in autumn. Exceptions were the years 1992, 1994 and 2006, when the plant residues of barley, wheat and maize were completely removed from all plots.

2.2. Soil sampling and chemical analysis

All soil samples were taken on 8 March 2007 when the sites lay fallow before sowing of broad beans. The last tillage of the ploughed fields was carried out in April 2006 followed by seedbed preparation for maize, using the rotary harrow twice. The rotary harrow plots were also treated twice at the beginning of May 2006, followed by sowing of maize. In October 2006, the maize was harvested and removed from the plots. In November, all plots were shallowly tilled (3 cm depth) with a rotary cultivator and left over winter. The soils were sampled in a grid design with 4 replicates from each plot at 0–5, 5–10, 10–20, 20–30, and 30–40 cm depth using a steel core with 4 cm diameter at 0–5 and 5–10 cm depth and 8 cm diameter in the deeper layers. This resulted in 16 samples per treatment and depth at the site Garte and 12 replicate samples per treatment and depth at the site Hohes Feld. All samples were sieved (<2.0 mm) to remove roots and crop residues, adjusted to 40% water holding capacity (WHC) and stored in polyethylene bags at 4 ℃ until soil biological analysis was carried out. A subsample was dried (105 \degree C) and finely ground for chemical analysis. The pH was determined in water with $1/2.5$ (w/v). Total contents of C and N were detected by gas chromatography using a Vario MAX (Elementar, Hanau, Germany) elemental analyser. Total S and P were analysed by an HNO3 $^-$ pressure digestion as described by [Chander et al. \(2008\)](#page--1-0) by ICP-AES (Spectro Analytical Instruments GmbH, Kleve, Germany).

2.3. Microbial activity and biomass indices

For measuring basal respiration, 60 g moist soil adjusted to 40% water holding capacity was weighed into 80 ml incubation cylinders made of stainless steel nets ([Hoffmann et al., 2010\).](#page--1-0) The cylinders were placed in 500 ml Pyrex glass jars containing 5 ml 0.5 M NaOH at the bottom and incubated for 7 days at 22 \degree C in the dark. The $CO₂$ evolved was determined by back-titration of the excess NaOH to pH 8.3 using 0.5 M HCl after addition of 5 ml of a saturated BaCl₂ solution. The metabolic quotient qCO_2 [\(Anderson and](#page--1-0) [Domsch, 1990\)](#page--1-0) was calculated as the ratio of basal respiration to the corresponding microbial biomass C at the end of the laboratory incubation.

Microbial biomass C, N, P and S were estimated by fumigation extraction. For determining microbial biomass C and N [\(Brookes](#page--1-0) [et al., 1985; Vance et al., 1987\),](#page--1-0) 10 g moist soil was fumigated at 25 \degree C with ethanol-free CHCl₃, which was removed after 24 h. 10 g of fumigated and non-fumigated soil were extracted with 40 ml of 0.5 M K₂SO₄ by 30 min horizontal shaking at 200 rev min⁻¹ and filtered (hw3, Sartorius Stedim Biotech, Göttingen, Germany). Organic C in the extracts was measured as $CO₂$ by infrared absorption after combustion at 850 $°C$ using a Dimatoc 100 automatic analyser (Dimatec, Essen, Germany). Microbial biomass C was calculated as E_C/k_{EC} , where E_C = (organic C extracted from fumigated soil) − (organic C extracted from non-fumigated soil) and k_{EC} = 0.45 ([Wu et al., 1990\).](#page--1-0) Total N in the extracts was measured using a Dima-N chemoluminescence detector (Dimatec). Microbial

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