Geoderma 226–227 (2014) 79–84

Contents lists available at ScienceDirect

Geoderma

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

Microbial residue indices down the soil profile after long-term addition of farmyard manure and mineral fertilizer to a sandy soil

André Sradnick [a,*], Meike Oltmanns [b], Joachim Raupp [c], Rainer Georg Joergensen [a]

[a] Department of Soil Biology and Plant Nutrition, University of Kassel, Nordbahnhofstr. 1a, 37213 Witzenhausen, Germany
[b] Institute for Biodynamic Research, Brandschneise 5, 64295 Darmstadt, Germany
[c] Agric-science.org, Geinsheimer Str. 3, 64521 Gross-Gerau, Germany

ARTICLE INFO

Article history:
Received 24 July 2013
Received in revised form 4 March 2014
Accepted 5 March 2014
Available online 31 March 2014

Keywords:
Farmyard manure
Soil organic C
Amino sugars
Fungal C to bacterial C ratio

ABSTRACT

Long-term organic fertilization may control the accumulation of organic matter in subsoil. The objective of this study was to evaluate the effects of long-term farmyard manure application in comparison with mineral fertilization on the accumulation of amino sugars as indices for microbial residues down to 1 m depth at a sandy site that exhibits highly heterogeneous pH conditions. In relation to maximum values in topsoil at 90–100 cm depth, the SOC content decreased to roughly 24% and the total N content to 16% of the maximum values, leading to an increased soil C/N ratio from 11 to values around 16 in all treatments. The relative contribution of microbial residue C to SOC decreased with depth from 68% at 0–25 cm to 24% at 50–100 cm. In the subsoil, the stocks of microbial residue C were increased by manure in comparison with mineral fertilization, but not the stocks of SOC. This suggests that manure-induced priming effects increase the microbial turnover at 50–100 cm depth. Manure fertilization promoted the formation of bacterial residues in the topsoil at 0–25 cm depth, but not in the subsoil. Below the topsoil, the fungal C to bacterial C ratio decreased from 2.6 at 0–25 cm depth to 2.1 at 50–100 cm depth. Below the topsoil, the ratio of fungal to bacterial residues continuously decreased with depth from 2.7 to 1.7 at 90–100 cm depth, without fertilizer effects. Possible reasons for this decrease, such as effects of pH on the subsoil microbial community, a higher sensitivity of fungi to the absence of fresh organic matter or to an unfavourable composition of the subsoil atmosphere, need further investigations.

1. Introduction

Fertilization with cattle manure is an important means for improving soil fertility under arable conditions, by increasing the stocks of soil organic carbon (SOC) and microbial biomass in comparison with mineral fertilization (Edmeades, 2003; Esperschütz et al., 2007; Heinze et al., 2010). Most studies on the effects of organic fertilizers have focused exclusively on the topsoil down to 10 or 30 cm depth, where the contents of SOC and densities of microorganisms and roots are highest (Esperschütz et al., 2007; Joergensen et al., 2010). The proportion of SOC stored at 30 to 100 cm depth ranges between roughly 40 and 60% of the SOC stock at 0 to 30 cm depth (Batjes, 1996). The amount of microbial biomass C stored in the subsoil may reach similar values to that in the topsoil (Lavahun et al., 1996). However, the microbial biomass C to SOC ratio drastically declines with depth, as subsoils are depleted of fresh energy-rich plant material in comparison with topsoils (Jörgensen et al., 2002). This leads to a relative increase in the contribution of microbial residues to subsoil OC, as the soil microorganisms have to survive mainly on the decompositions of highly processed SOC (Boström et al., 2007; Liang and Balser, 2008; Rumpel and Kögel-Knabner, 2011).

The depletion of fresh energy-rich plant material also changes the microbial community structure with depth, especially the ratio of fungi to bacteria, i.e. the two microbial groups that dominate the biomass of soil microorganisms by more than 95% (Joergensen and Emmerling, 2006). Ekelund et al. (2001) and Fierer et al. (2003) detected disproportionate decreases in fungal biomass with depth in comparison with bacterial biomass, using direct microscopy in a forest soil and PLFA in arable and a grassland soil, respectively. Not only depth but also organic fertilization might have significant effects on subsoil microbial processes and SOC storage, caused by promoting bacteria (Heinze et al., 2010; Scheller and Joergensen, 2008), a higher formation of dissolved organic C (Kaiser and Kalbitz, 2012; Liang et al., 2012), and increased root growth (Chirinda et al., 2012). However, the input of easily available C by these two processes may lower the SOC contents, as priming effects make stable subsoil OC available to soil microbial decomposition (Fontaine et al., 2007).

It has repeatedly been shown that amino sugars are useful indicators for the accumulation of microbial residues also in the subsoil (Appuhn

* Corresponding author at: Leibniz Institute of Vegetable and Ornamental Crops Grossbeeren/Erfurt e.V., Theodor-Echtermeyer-Weg 1, 14979 Grossbeeren, Germany.
E-mail address: sradnick@igzev.de (A. Sradnick).

http://dx.doi.org/10.1016/j.geoderma.2014.03.005
0016-7061/

et al., 2006; Liang and Balser, 2008). Soil amino sugars are stabilized in soil (Glaser et al., 2004) and survive the biomass they originate from (Guggenberger et al., 1999). Fungal cell walls are the major source of glucosamine (Joergensen and Wichern, 2008), whereas bacterial cell walls, especially in the murein skeleton of Gram-positive species, are the exclusive source of muramic acid (Appuhn and Joergensen, 2006). Amino sugar analysis gives important information on the relative contribution of fungi and bacteria to the fraction of soil microbial residues (Joergensen and Wichern, 2008). For this reason, the ratio of fungal to bacterial residues is a sensitive tool for investigating the effects of organic fertilizers (Joergensen et al., 2010) and soil pH (Sradnick et al., 2014).

The objective of this study was to evaluate the effects of long-term farmyard manure application in comparison with mineral fertilization on the accumulation of amino sugars as indices for microbial residues down to 1 m depth at a sandy site that exhibits highly heterogeneous pH conditions (Heinze et al., 2010). The underlying hypotheses are: (1) The relative contribution of microbial residues to SOC increases with depth, due to the survival of microorganisms on the decompositions of highly processed SOC. (2) In the manure treatment, the higher C input by roots and soluble organic matter increases subsoil OC sequestration, despite the possibility of priming effects. (3) The ratio of fungal to bacterial residues generally declines with depth and is lower in the manure treatment, due to preferential stimulation of bacterial growth.

2. Materials and methods

2.1. Experimental site

The depth profiles were taken from field A of a long-term field trial of the Institute of Biodynamic Research, Darmstadt, Hessia, Germany (49°50′N, 8°34′E) at 100 m above sea level (Heinze et al., 2010; Raupp and Oltmanns, 2006). The long-term experiment was established in 1980 on a Haplic Cambisol (FAO-WRB, 2006) with 86% sand, 9% silt, and 5% clay, which has been developed from alluvial sediments of the river Neckar. The mean annual temperature is 9.5 °C and the mean annual precipitation is 590 mm. The experiment was based on a strip design in split blocks with four replicate plots (5 m × 5 m) of each treatment, with fertilizer type applied at three different rates (Heinze et al., 2010; Raupp and Oltmanns, 2006). Composted cattle farmyard manure (CM) without straw return and mineral fertilizer (MIN, i.e. calcium ammonium nitrate, superphosphate, potassium chloride, since 1996 potassium magnesia) with the return of straw, given at the highest rate, were compared for the current study. The application rate was 140 kg N ha^{-1} for cereals and 150 kg N ha^{-1} for root crops. No N fertilizer was added in the years with legumes in the crop rotation. The CM treatment received approximately 10% higher mean annual C input rates in comparison with the MIN treatment (Heinze et al., 2010; Heitkamp et al., 2009). The mean annual inputs of SOC, Nt and P were 1300, 121, 28 and 930, 111, 100 kg ha^{-1} $year^{-1}$ for the CM and the MIN treatment respectively (Heinze et al., 2010; Heitkamp et al., 2009; Raupp and Oltmanns, 2006).

2.2. Sampling and soil chemical analysis

During September 2009, the soil samples were taken two months after cropping. The samples were collected at 5 cm steps down to 100 cm depth, using a steel corer (Eijkelkamp SC/SE diameter 4 cm). The soils were sieved (<2 mm), adjusted to 50% water holding capacity and stored in polyethylene bags at 4 °C for several weeks until soil biological analysis. A sub-sample was dried and finely ground for chemical analyses. The pH was determined in water using a soil to water ratio of 1 to 2.5. Total C and N were measured by gas chromatography using a Vario EL (Elementar, Hanau, Germany) analyser. For determining mobile C, 5-g samples were extracted with 20 ml of 0.5 M K_2SO_4 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 using a Dimatoc 100 automatic analyser (Dimatec, Essen, Germany).

2.3. Amino sugars

The amino sugars muramic acid (MurN), mannosamine (ManN), glucosamine (GlcN), and galactosamine (GalN) were determined according to Appuhn et al. (2004) as described by Indorf et al. (2011) using OPA (o-phthalaldehyde) derivatisation. Moist samples of 0.5 g soil were hydrolysed with 10 ml 6 M HCl, for 6 h at 105 °C. Chromatographic separations were performed on a Hyperclone C_{18} column (125 mm length × 4 mm diameter) at 35 °C, using a Dionex (Germering, Germany) P 580 gradient pump, a Dionex Ultimate WPS — 3000TSL analytical autosampler with in-line split-loop injection and thermostat and a Dionex RF 2000 fluorescence detector set at 445 nm emission and 330 nm excitation wavelengths. Fungal C was calculated by subtracting bacterial GlcN from total GlcN as an index for fungal residues, assuming that MurN acid and GlcN occur at a 1 to 2 molar ratio in bacterial cells (Engelking et al., 2007): mmol fungal C g^{-1} dry weight = (mmol GlcN − 2 × mmol MurN) × 9. Bacterial C was calculated as an index for bacterial residues by multiplying the concentration of MurN by 45 (Appuhn and Joergensen, 2006). Microbial residue C was estimated as the sum of fungal C and bacterial C.

2.4. Statistical analysis

The results presented in the tables are arithmetic means and expressed on an oven-dry basis (about 24 h at 105 °C). The significance of differences between the fertilizer treatments and the effects of soil pH, depth, and soil organic matter on these differences was determined by analyses of covariance (ANCOVA), by normalizing the fertilizer treatment effects for differences in soil pH and depth. For this analysis, the results of soil samples collected at different depths were recalculated into stocks using bulk density data and summarized to three depth zones: (1) the topsoil (0–25 cm), representing the plough layer with incorporation of fertilizer and straw, (2) the intermediate zone (25–50 cm), specifying a highly heterogeneous transition zone, and (3) the subsoil (50–100 cm). Differences between means were tested using least significant differences (LSD) at $P < 0.05$. All statistical analyses were performed using SPSS, version 16.

3. Results

Soil pH values increased from around 7 in the topsoil to about 7.5 in the subsoil of the MIN and to 8.0 in that of the CM treatment (Fig. 1).

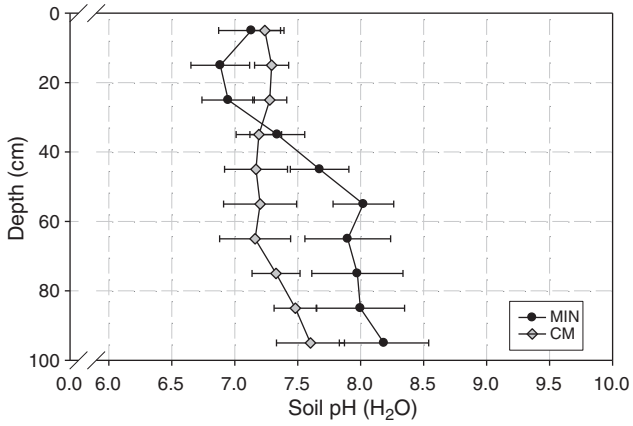

Fig. 1. Soil pH–H_2O at 10 depths. MIN = mineral fertilizer, CM = composted farmyard manure; bars show one standard error of mean (n = 8).