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Temporal variation in surface and subsoil abundance and function of the soil microbial community in an arable soil

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ABSTRACT

Many studies of the microbial ecology of agricultural ecosystems focus on surface soils, whereas the impacts of management practice and season on soil microbial community composition and function below the plough zone are largely neglected. Deep soils have a high potential to store carbon; therefore any management driven stimulation or repression of microorganisms in subsoil could impact biogeochemical cycling in agricultural sites. The aim of this study was to understand whether soil management affects microbial communities in the topsoil (0–10 cm), rooted zone beneath the plough layer (40–50 cm), and the unrooted zone (60–70 cm). In a field experiment with different crops [wheat (*Triticum aestivum* L.) and maize (*Zea mays* L.)] and agricultural management strategies (litter amendment) we analysed microbial biomass as phospholipid fatty acids (PLFAs) and enzyme activities involved in the C-cycle (β-glucosidase, N-acetyl-β-D-glucosaminidase, β-xylosidase, phenol- and peroxidase) across a depth transect over a period of two years. Wheat cultivation resulted in higher bacterial and fungal biomass as well as higher enzyme activities at most sampling dates in comparison to maize cultivated plots, and this effect was visible to 50 cm depth. Litter application increased bacterial and fungal biomass as well as hydrolytic enzyme activities but effects were apparent only in the topsoil. In winter high microbial biomass and enzyme activities were measured in all soil layers, possibly due to increased mobilization and translocation of organic matter into deeper soil. Hydrolytic enzyme activities decreased with depth, whereas oxidative enzyme activities showed no decrease or even an increase with depth. This could have been due to differing sorption mechanisms of hydrolytic and oxidative enzymes. Specific enzyme activities (enzyme activity per microbial biomass) were higher in the deeper layers and possible reasons are discussed.

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1. Introduction

Soils have a great potential to store carbon throughout entire soil profiles (Lorenz and Lal, 2005). The global carbon stock within the first metre of soils has been estimated at 1500–2000 Pg (Janzen, 2005). Although over 50 percent of the global organic carbon pools in soil are found below 30 cm depth (Jobbagy and Jackson, 2000), the contribution of microorganisms to carbon dynamics in subsoils has received far less attention than in topsoils (Rumpel and Kögel-Knabner, 2011). Focussing on top- as well as on subsoils in agricultural ecosystems is important, because soil management (e.g. soil tillage, crop type, N amendment, and residue management such as mulching) may influence not only the input and turnover of organic C (Clapp et al., 2000; Lorenz and Lal, 2005), but also alter subsoil processes related to plant nutrient acquisition (Harrison et al., 2011).

Soil microorganisms play an important role in the formation and turnover of SOM by decomposition of plant residues and remineralization of nutrients (Bardgett et al., 2005). Their size, community composition and function can therefore be used to investigate decomposition and deduce SOM turnover in soil. Soil microbial communities are not uniformly dispersed throughout the soil profile, but reflect patches of available resources, predominantly plant litter and roots. Crop type or management strategy therefore affect community structure and distribution. For example, vegetation dependant factors such as plant species influence the size and composition of microbial communities (Moore-Kucera and Dick, 2008) through amount, availability and quality of exudates, distribution of roots in the soil profile, and through the quality of plant residues. With agricultural amendments such as litter application the additional substrates are likely to affect microbial community and their function throughout the

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soil profile. Studies in natural ecosystems indicate that increased nutrient availability modifies microbial assemblages not only in topsoils but also in subsoils of the vadose zone (Schütz et al., 2009).

To understand SOM dynamics in top- and subsoils it is also important to take into account abiotic changes in depth. For example, higher absolute amounts of minerals in deeper soil layers can result in higher stabilization potential of organic matter with minerals at depth (Rasse et al., 2005). Also seasonally dependant abiotic factors such as temperature and soil moisture can have a strong influence on biomass and activity of microbes, and seasonal effects can even be higher than treatment effects (Debosz et al., 1999; Bell et al., 2010). Moreover, climatic forcing in the topsoil, e.g. drying/wetting or freezing/thawing cycles, triggers the release of mobile organic dissolved and particulate substances (MOPS) (Majdalani et al., 2008). Transient flow conditions affecting organic matter further occur as a result of cell death and lysis during drying, disruption of soil structure due to mechanical stress, and harvesting practices which increase fractured plant residues. Overall these multiple factors result in an enhanced transport of MOPS (Totsche et al., 2007), DOM (Kalbitz et al., 2000) and colloids (Cheng and Saiers, 2009) in the soil profile, depending on season.

It is widely thought that substrate quality is lower in subsoil than in topsoil, suggesting that soil organic matter is less degradable at depth. Both substrate pools and microbial biomass generally decline (Blume et al., 2002; Bausenwein et al., 2008; Gelsomino and Azzellino, 2011), and activity also decreases with increasing soil depth (Fang and Moncrieff, 2005). However, in studies where assimilation or mineralization activities were normalized to the size of the microbial biomass, these specific activities showed either similar values within the soil profile or even an increase with depth (Blume et al., 2002; Gelsomino and Azzellino, 2011).

One important function of soil microorganisms is the degradation of insoluble polymers like cellulose, lignin, and chitin into smaller subunits by extracellular enzymes. It has been shown that substrate presence induces respective enzyme synthesis (Suto and Tomita, 2001) and therefore enzyme activities can be used to yield information about availability of particular substrates in soils (Geisseler and Horwath, 2009). Whereas hydrolytic enzymes (e.g. β-glucosidase, N-acetyl-β-D-glucosaminidase, xylosidase) are responsible for the decay of organic substrates with faster turnover times like carbohydrates or chitin, oxidative enzymes (e.g. phenol- and peroxidase) have an important function in the degradation of SOM components with slower turnover times (e.g. lignin) (Horwath, 2007). Specific enzyme activity (enzyme activity per unit microbial biomass) gives further information about the production and/or stabilization of enzymes (Kandeler and Eder, 1993; Taylor et al., 2002; Allison et al., 2007).

The present study investigated the effects of frequently cultivated crop types (maize and wheat) and management (litter and no litter) on microbial community composition (PLFAs), and its function (enzyme activities) at three different depths (topsoil, rooted zone beneath plough layer, unrooted zone) in an arable field over a period of two years.

We hypothesized that crop type, management strategy (litter amendment) and season have more pronounced effects on microbial properties in the top- than in the subsoil, as surface communities are more exposed to mechanical, chemical or vegetation changes. Further, we expected that not only lower microbial abundance, but also changes in physiology of soil microorganisms (their enzyme production and expression), as well as abiotic interactions between microorganisms, substrates, and soil enzymes may drive C dynamics in subsoils.

2. Materials and methods

2.1. Field site and soil samples

The main field experiment investigating carbon flow in belowground food webs was set up by the University of Göttingen (Lower Saxony, Germany) on arable land (51°33′N, 91°53′E; 158 m a.s.l.) in April 2009 (Kramer et al., 2012). The area has a temperate climate with mean annual precipitation of 720 mm and mean air temperature of 7.9 °C. Dominant soil types at the site are Luvisols and Cambisols with partially stagnic properties (IUSS, 2007). The clay and sand fractions decrease from 7.0 and 5.8 to 6.8 and 4.8% (w/w) from the Ap1 to the Bv2 horizon, respectively, whereas the silt fraction increases from 87.2 to 88.4% (w/w). The soil bulk density of the site increases from 1.38 g cm^{-3} in the Ap1 to 1.68 g cm^{-3} in the Bv2 horizon. The pH_{CaCl_2} in the Ap1 to the Bv2 (>65 cm) horizon increases from 6.0 to 7.0. The C_{org} and total N content decrease from the Ap1 to the Bv2 (>65 cm) horizon from 11.6 to 1.8 mg g^{-1} dry weight and 1.2 to 0.3 mg g^{-1} dry weight, respectively (for details see Kramer et al., 2012; Pausch and Kuzyakov, 2012).

Four treatments were established which were differentiated by crop type (wheat vs. maize) and management strategy (litter or no litter application). To allow feasible agricultural management a strip design was chosen, with wheat (*Triticum aestivum* L) cultivated in the first (north) and maize (*Zea mays* L.) in the second (south) strip, each strip with 10 plots of 24 × 24 m. Before sowing, soil was tilled with a chisel plough to a depth of 12 cm. In the first vegetation period (2009) winter wheat ("Julius", sown at 224 kg ha^{-1}) and maize ("Ronaldinio", sown at 34 kg ha^{-1}) were grown. In the second period (2010) the varieties used were summer wheat ("Melon", sown at 224 kg ha^{-1}) and hybrid maize ("Fernandez", sown at 26 kg ha^{-1}). Fertilization practice was as follows: on the maize plots ammonium nitrate urea solution (2009: 122.4 kg N ha^{-1}; 2010: 79.2 kg N ha^{-1}) and di-ammonium phosphate (2009/2010: 32.4 kg N ha^{-1} and 82.8 kg P ha^{-1}) were applied twice, shortly before and after seeding. The wheat plots received granular NS fertilizer (21.0 kg N ha^{-1}, 24.0 kg S ha^{-1}) in March 2009 and ammonium nitrate urea solution between 39.5 and 61.3 kg N ha^{-1} in April, May and June in both 2009 and 2010. After harvest in early November 2009 chopped maize litter excluding cobs (0.8 kg m^{-2} dry weight equivalent to 0.35 kg C m^{-2}) was applied on 5 randomly chosen plots from the 10 plots in each strip to establish the Corn Maize (CM) and Wheat + maize Litter (WL) treatments. The other 5 plots of each strip were the plots without litter addition and designated Fodder Maize (FM) and Wheat (W) treatments. In November 2010 the harvested maize litter from this year was applied.

Soil samples were taken with a soil corer to 70 cm depth and separated as follows: topsoil (0–10 cm), rooted zone beneath the plough layer (40–50 cm), and unrooted zone (60–70 cm). Ten soil cores randomly distributed on each plot were taken between plants. The soil from each depth was mixed and homogenized. Soil samples were cooled and transported to the laboratory. Soils were sieved (<2 mm), water content was gravimetrically determined (105 °C for 24 h), and samples were frozen at −24 °C. All data presented here are expressed on a soil dry weight basis.

Soil samples were collected three times a year; in summer, autumn, and winter. Summer sample collections were July 2009 and 2010 (high root exudation) and autumn collections in September 2009 and 2010 (shortly before maize harvest). Winter sample collections (highest translocation of MOPs) were in December 2009 and in January 2011. The second winter collection was delayed due to heavy snow in December 2010. For more details see Kramer et al. (2012).

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