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Microbial residues as indicators of soil restoration in South African secondary pastures

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ABSTRACT

Prolonged intensive arable cropping of semiarid grassland soils in the South African Highveld resulted in a significant loss of C, N and associated living and dead microbial biomass. To regenerate their soils, farmers converted degraded arable sites back into secondary pastures. The objective of this study was to clarify the contribution of microorganisms to the sequestration of C and N in soil during this regeneration phase. Composite samples were taken from the topsoils of former arable land, namely Plinthustalfs, which had been converted to pastures 1–31 years ago. Amino sugars were determined as markers for microbial residues in the bulk soil and in selected particle-size fractions. The results showed that when C and N contents increased during the secondary pasture usage, the amino sugar concentration in the bulk soil (0–5 cm) recovered at similar magnitude and reached a new steady-state level after approximately 90 years, which corresponded only to 90% of the amino sugar level in the primary grassland. The amino sugar concentration in the clay-sized fraction recovered to a higher end level than in the bulk soil, and also at a faster annual rate. This confirms that especially the finer particles contained a high amount of amino sugars and were responsible, thus, for the restoration of microbially derived C and N. The incomplete recovery of amino sugars in bulk soil can only in parts be attributed to a slightly coarser texture of secondary grassland that had lost silt through wind erosion. The soils particularly had also lost the ability to restore microbial residues below 5 cm soil depth. Overall, the ratios of glucosamine to muramic acid also increased with increasing duration of pasture usage, suggesting that fungi dominated the microbial sequestration of C and N whereas the re-accumulation of bacterial cell wall residues was less pronounced. However, the glucosamine-to-muramic acid ratios finally even exceeded those of the primary grassland, indicating that there remained some irreversible changes of the soil microbial community by former intensive crop management.

1. Introduction

Soil degradation by prolonged arable cropping usually goes along with a loss of soil organic matter (SOM) (Oldman, 1998; Lobe et al., 2001; Brodowski et al., 2004) and of associated living and dead microbial biomass (Amelung et al., 2002; Postma-Blaauw et al., 2010). Such SOM losses were notably high in arable sites of the South African Highveld, because of the Plinthustalfs' sandy and coarse texture (Lobe et al., 2001, 2002, 2005). To regenerate the soils and hence increase their fertility, restoring the SOM content seems essential (Obi and Ebo, 1995). One strategy to restore SOM content and to increase soil fertility is a change in land usage (Conant et al., 2001). Therefore, the degraded sites in the grassland region of semiarid South Africa have been continuously converted back into grassland for soil regeneration. This conversion started about 30 years ago and has continued until today. Yet, a complete recovery of SOM content was almost impossible, and 10–95 years of secondary pasture management will be needed to restore SOM content (in 0–20 cm) to a new equilibrium, which will be most pronounced near the surface (Preger et al., 2010). Several other studies also indicated that in secondary pastures the SOM content does not restore to a level common in natural grasslands (Potter et al., 1999; Knops and Tilman, 2000; Preger et al., 2010; see also Guo and Gifford, 2002 for a review). Little is known, however, on the role of microorganisms for the sequestration of C and N, and about the mechanisms regulating the possible accrual of microbial residues in former degraded arable sites of this semiarid climate.

As long as substrate limitations existed in the former degraded soils, labile plant residues are practically not accumulated but are rapidly converted into microbial biomass (Robles and Burke, 1998).

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When the microorganisms die, their stable cell wall components still exist in soil. As amino sugars are not synthesized in significant amounts by plants, the concentrations and patterns of amino sugars in soil may be used, therefore, to evaluate the contribution of bacteria and fungi to the sequestration of C and N in soil (Amelung et al., 1999; Dai et al., 2002; Glaser et al., 2004; Appuhn and Joergensen, 2006). Acid hydrolysis of SOM has revealed that approximately 5–10% of soil N is present as amino sugars (Mengel, 1996; Schulten and Schnitzer, 1998). Fluctuations in living biomass do not alter the amino sugar content in soil because most of the amino sugars occur in dead microbial cells (Chantigny et al., 1997; Guggenberger et al., 1999; Kandeler et al., 2000). The free amino sugars turn over rapidly and provide a suitable substrate for microbial respiration and new biomass formation (Roberts et al., 2007), i.e., the free amino sugars scarcely add to total amino sugar content (Schulten and Schnitzer, 1998). Also, Brodowski et al. (2004) found that D-alanine in soil (another excellent marker for peptidoglycan, similar to muramic acid) are almost non-existent in free forms in soil, confirming that the same is unlikely for muramic acid.

The individual amino sugars in the soil have various microbial origins (Parsons, 1981). Muramic acid is solely synthesized by bacteria and is a compound of their peptidoglycan cell walls, whereas fungal cell wall contains glucosamine in significant greater quantities than bacterial cell walls (Chantigny et al., 1997; Guggenberger et al., 1999). Less is known about the origin of galactosamine in soil. It has also sometimes been used as an indicator for bacterial gums and tissue (Kögel and Bochter, 1985; Amelung, 2001; Liang et al., 2007a,b), but it may also be produced by fungi (Glaser et al., 2004; Engelking et al., 2007). The origin of mannosamine is not known to date.

In regenerating soils of the Great Plains of North America, the amino sugar concentrations had accumulated more rapidly than did bulk C and N after about 8 years of arable land conversion back into grassland, however, soil compaction and limited fertilizer use likely still inhibited the sequestration of C and N by muramic acid-containing bacteria (Amelung et al., 2001a). Whether similar mechanisms also hold true for the semiarid grasslands of South Africa still warrant clarification. In general, different pools of SOM respond differently to land-use change (e.g., Christensen, 1996; Feller and Beare, 1997; Chantigny et al., 1997) and C-sequestration (e.g., Guggenberger et al., 1999; Liang et al., 2007a; Gulde et al., 2008). These pools may be approached by physical fractionation procedures, of which a simple particle-size fractionation has received wide application (e.g., Christensen, 1996; Skjemstad et al., 2004). Hence, we combined particle-size fractionation with biomarker analyses to better understand the microbial impact on the C and N dynamics in the secondary pasture soils. The overall aim of this study was to clarify the contribution of bacteria and fungi to the sequestration of C and N in secondary pastures. In particular we were interested to see whether, how fast, and to which degree microbial residues can be restored relative to bulk SOM when regenerating former degraded arable land by secondary pasture management.

2. Materials and methods

2.1. Samples

Du Preez and Du Toit (1995) defined an agroecosystem as a region with homogeneous environmental factors such as climate, topography and soil, which influence the yield. Three comparable agroecosystems near Harrismith, Kroonstad and Tweespruit in the Free State Province of South Africa were selected as independent replications of our land-use chronosequence in this study. The three agroecosystems are located in the South African Highveld, a central high plateau at an altitude between 1200 and 2000 m above sea level. The potential natural vegetation of the Highveld is open grassland (Bredenkamp et al., 1996), though geophytes and herbs also grow here. The soils were Plinthustalfs (Soil Survey Staff, 1998), equivalent to Westleigh or Avalon soil forms in the South African soil classification system (Soil Classification Working Group, 1991). The respective WRB soil types are mainly Plinthic Lixisols (WRB, 2006). For more information on the site properties see Table 1. Before being converted to secondary pastures, the sites were used to grow crops. Especially maize, wheat, sunflower and partly oat (as winter fodder for cattle) were cultivated in this area, and fields were plowed up to a depth of 20 cm. The mineral fertilization was in maize: 10–80 kg N, 5–20 kg P, 0–10 kg K ha^{-1} $year^{-1}$; in wheat: 20–50 kg N, 4–11 kg P, 0–10 kg K ha^{-1} $year^{-1}$ and in sunflower 35–50 kg N, 8–15 kg P, 0–4 K ha^{-1} $year^{-1}$. Former wheat yields amounted to 1–2 t ha^{-1} while maize and sunflower yields ranged between 0.8 and 2.5 t ha^{-1} (Lobe et al., 2005).

In each of the three agroecosystems, the following three types of land usage were sampled: secondary pastures, which were converted back to pastures from degraded arable land at variable time intervals (from 1 to 31 years ago), sites currently used as arable land (more than 20 years cultivated) and primary grasslands. The sampling time was in 2005 at the beginning of the rainy season (end of September until the beginning of November). Surface samples were taken from five subsites on each of the primary and secondary grassland sites as well as on the arable land. Prior to sampling, the subsites had to be watered to allow the hand auger to penetrate into the hardened grassland soil. All samples were then air-dried and sieved to 2 mm for analyses (fine earth <2 mm).

2.2. Analyses

Data for total C and N contents taken from Preger et al. (2010) who assessed them after dry combustion (DIN ISO 10964) with the standard elemental analyzer (Fisons NA 2000). There was no detectable inorganic C, so we can regard the total C as entirely organic. The pH-value was determined in deionized water with a soil to solution ratio of 1–2.5.

2.3. Particle-size separates

The particle-size separates of the samples at 0–5 cm depth were those of the study of Preger et al. (2010) which obtained the particle-size fractions coarse sand >250 μm, fine sand 20–250 μm, silt 2–20 μm and clay <2 μm. Briefly, the coarse sand was obtained by using ultrasonic dispersion (60 J mL^{-1}) and wet sieving to 250 μm. For the extraction of clay, the soil was once more treated with ultrasound (440 J mL^{-1}), and then centrifuged in several steps until the supernatant was clear. The clay fraction was flocculated with 10% $MgCl_2$ overnight and subsequently freeze-dried. The residual fine sand and silt fraction were separated using wet sieving to 20 μm (see Amelung and Zech, 1999, for method details). All fractions were dried at 40 °C. We examined the amino sugar content and ratios in the 0–5 and 5–10 cm depth in the bulk soil and additionally in the 0–5 cm depth in the four particle-size fractions. In all the sampled secondary pastures we assumed that the clay fraction indicated the accrual of amino sugars best as beside silt, which is re-allocated by wind erosion; the clay fraction also indicated the loss of amino sugars with prolonged cropping (Amelung et al., 2002). Concerning the other fractions we reduced the time-consuming analyses by only selecting secondary pasture soils at the ages of 10, 17 and 30 years. For these age classes of the secondary pasture sites, we also determined the amino sugar contents for the 10–20 cm depth interval (data not shown). We did not include further samples of this depth interval into our analyses,