



## Mechanisms of short-term soil carbon storage in experimental grasslands

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### ABSTRACT

We investigated the fate of root and litter derived carbon in soil organic matter and dissolved organic matter in soil profiles, in order to explain mechanisms of short-term soil carbon storage. A time series of soil and soil solution samples was investigated at the field site of The Jena Experiment between 2002 and 2004. In addition to the main experiment with C3 plants, a C4 species (*Amaranthus retroflexus* L.) naturally labeled with <sup>13</sup>C was grown on an extra plot. Changes in organic carbon concentration in soil and soil solution were combined with stable isotope measurements to follow the fate of plant carbon into the soil and soil solution. A split plot design with plant litter removal versus double litter input simulated differences in biomass input. After 2 years, the no litter and double litter treatment, respectively, showed an increase of 381 g C m<sup>-2</sup> and 263 g C m<sup>-2</sup> to 20 cm depth, while 71 g C m<sup>-2</sup> and 393 g C m<sup>-2</sup> were lost between 20 and 30 cm depth. The isotopic label in the top 5 cm indicated that 115 g C m<sup>-2</sup> and 156 g C m<sup>-2</sup> of soil organic carbon were derived from C4 plant material on the no litter and the double litter treatment, respectively. Without litter, this equals the total amount of 97 g C m<sup>-2</sup> that was newly stored in the same soil depth, whereas with double litter this clearly exceeded the stored amount of 75 g C m<sup>-2</sup>. Our results indicate that litter input resulted in lower carbon storage and larger carbon losses and consequently accelerated turnover of soil organic carbon. Isotopic evidence showed that inherited soil organic carbon was replaced by fresh plant carbon near the soil surface. Our results suggest that primarily carbon released from soil organic matter, not newly introduced plant organic matter, was transported in the soil solution. However, the total flow of dissolved organic carbon was not sufficient to explain the observed carbon storage in deeper soil layers, and the existence of additional carbon uptake mechanisms is discussed.

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### 1. Introduction

Increasing carbon storage in soils is one option helping to mitigate increasing atmospheric CO<sub>2</sub> concentrations and global climate change (Prentice, 2001; Lal, 2004b). In terrestrial ecosystems plants are able to reduce atmospheric CO<sub>2</sub> and bind it in biomass. The input of this plant material to the soil as roots or litter stores carbon only during the short term, however (Parton et al., 1988; Jenkinson, 1990). Roots are considered to be a more stable form of carbon supply to the soil than litter (Denef and Six, 2006). The latter causes priming of microbial decomposition and thus also faster degradation of present soil organic carbon (SOC).

Numerous investigations have shown that organic carbon stocks in soils are determined by the land use. Changing arable fields to

managed grasslands, as on our field site, increases carbon concentrations in soil within a few years (Romkens et al., 1999; Balesdent et al., 2000; Lal, 2004a). At the same time the carbon distribution in the soil profile changes. In contrast to the homogeneous input of plant material to soil in grasslands is controlled by the aboveground litter layer and the root distribution. As 70–75% of the root biomass in grasslands is located in the top 15 cm of the soil (Gill et al., 1999; Gleixner et al., 2005) organic carbon concentrations increase in the main rooting zone but decrease beneath this zone. Recent experimental evidence demonstrates that the type and diversity of plant species in grasslands plays an important role for carbon transfer into the soil and is able to modify carbon storage under a given land use scheme (Tilman et al., 2006; Steinbeiss et al., in press). As higher plant biodiversity leads to larger plant biomass (Lambers et al., 2004; Roscher et al., 2005; Balvanera et al., 2006) and therefore a larger biomass input into the soil, it is generally assumed that differences in input amounts (not quality of the input material) are responsible for the observed variation in soil carbon storage (Catovsky et al., 2002; Skinner et al., 2006). Soil microorganisms, however, might be

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especially activated by the input of fresh and easily decomposable plant material if a higher plant diversity increases the variability of compounds available as a nutrient source (Wardle et al., 1999; Hooper et al., 2000; Stephan et al., 2000). Different groups of soil microorganisms, such as bacteria or fungi, use different carbon sources and therefore complement each other in resource utilization (Fontaine et al., 2003; Kramer and Gleixner, 2006). This so-called priming effect (defined as “strong short-term changes in turnover of soil organic matter caused by comparatively moderate treatments of the soil” (Kuzyakov et al., 2000)) not only transforms plant biomass to soil organic carbon but results in the decomposition and mobilization of already present SOC (Fontaine et al., 2004; Fontaine and Barot, 2005). This mobilized carbon can be transported in soil solution and is exported from the microbial active zone near the roots. Water moving downwards then carries dissolved organic carbon (DOC) to deeper soil horizons, where it is preserved from complete mineralization. Gradually the transported compounds are readsorbed to soil particles and contribute to carbon storage deeper in the soil profile (Kalbitz et al., 2005; Lajtha et al., 2005). So far it is unclear to what extent dissolved organic carbon in soil solution originates from plant decomposition products or mobilized soil organic carbon, and to what extent organic carbon is relocated by transport processes.

In our current study, stable carbon isotopes and their natural variability in plants with different photosynthetic pathways were used as tool to follow plant-derived carbon into the soil and soil solution carbon pools (Balesdent and Mariotti, 1996; Gleixner et al., 2002). As most plants in our geographical region possess a C3 photosynthetic pathway, the isotopic signature in soil organic carbon reflects typical  $\delta^{13}\text{C}$  values of C3 material in the range of about  $-25\text{‰}$  to  $-27\text{‰}$ . Growing plants with C4 photosynthesis, that show  $\delta^{13}\text{C}$  values between  $-12\text{‰}$  and  $-15\text{‰}$ , enabled us to quantify the proportion of fresh carbon incorporated into the soil and soil solution.

The current study focused on the role of aboveground litter input on the development of the soil carbon pool, while root biomass input was held constant between treatments by sowing the same plant species. We hypothesized that (1) the more litter is provided the more carbon is transferred to the soil, and that (2) the soil solution is transporting plant-derived carbon from the soil surface as well as mobilized soil organic carbon into deeper soil horizons for storage.

## 2. Materials and methods

### 2.1. Field site and plot design

All samples were taken at the field site of the Jena Experiment, a managed grassland biodiversity experiment established in spring 2002 on the outskirts of Jena, Germany ( $50^{\circ}55' \text{ N}$ ,  $11^{\circ}35' \text{ E}$ , altitude 130 m). The soil of the field site is classified as Eutric Fluvisol (FAO, 1998) developed from up to 2 m-thick loamy fluvial sediments that are almost free of stones. The texture in the plough horizon (0–30 cm) ranges from sandy loam to silty clay with increasing distance to the river. Thus, sand content varies from 40% near the river to 7% furthest from the river, while silt content ranges from 44% to 69%, respectively. The clay content is in the range of 16–24% and shows almost no spatial trend (Steinbeiss et al., in press). The field site was used as an arable field for the last 40 years and ploughed to a depth of about 30 cm. The organic carbon concentration in 0–30 cm depth at the start of the experiment in spring 2002 was in the range from 10 to 29 g C kg<sup>-1</sup>, corresponding to a carbon stock of 7.3 kg C m<sup>-2</sup> averaged for the whole field site. The carbonate content showed a strong spatial gradient and ranged between 4 and 42 g C kg<sup>-1</sup>.

The main biodiversity experiment consists of 86 plots (each 20 m × 20 m) that contain mixtures of grassland species from a 60-species pool. Plant species were grouped into four functional groups. Detailed information about the field site, the species pool and the main experimental design can be found elsewhere (Roscher et al., 2004). At the same time as the main experiment an additional plot (10 m × 20 m) was established with a C4 plant species (*Amaranthus retroflexus* L.) to achieve a natural isotopic label of carbon that enters the soil and the soil solution. Subsequently, this plot is referred to as C4 plot, while all other plots are C3 plots. The total initial carbon content in 0–30 cm depth on this plot was 7.4 kg C m<sup>-2</sup>. The spatial variability of the soil carbon content within this plot (determined from five independent soil cores) depended on depth and ranged between 7.1% and 25.5%, in which the higher variability was observed in depths below 20 cm. On the C4 plot a split plot design was used to achieve differences in litter input. Thus, in fall 2002 and 2003 the whole plot was mown and the mown aboveground biomass (1000–1500 g m<sup>-2</sup> y<sup>-1</sup> corresponding to 430–650 g C m<sup>-2</sup> y<sup>-1</sup>) was removed from one half of the plot and added to the other half of the plot, resulting in a no litter versus double litter treatment. On C3 plots the harvested aboveground biomass was always removed from the plots.

The population of C4 plants was very dense, which kept weeds small and rare. Consequently, only little weeding was necessary.

### 2.2. Soil sampling and analysis

Stratified soil sampling was performed on all plots before sowing in April 2002 and was repeated in April 2004 to a depth of 30 cm. In 2002, five independent samples per plot were taken using a split tube sampler with an inner diameter of 5 cm (Eijkelkamp Agrisearch Equipment, Giesbeek, Netherlands). Soil samples were dried at 40 °C and subsequently segmented to a depth resolution of 5 cm resulting in six samples per core. All samples were analyzed independently to calculate the spatial variability within the plots. In 2004, only three samples per plot were taken, whereas paired sampling was chosen to avoid additional spatial uncertainty (Lal et al., 2000). C4 treatments were handled like plots such that three samples per treatment were taken. Soil samples were already segmented into their respective depths at the field and mixed to a plot or treatment representative sample per depth (Webster, 2007). Subsequently, samples were dried at 40 °C. All soil samples were passed through a sieve with a mesh size of 2 mm. In 2002, generally no plant remains were found in the soil. Exceptionally appearing visible plant remains were removed using tweezers. Due to much higher proportions of roots in the soil, the samples in 2004 were further sieved to 1 mm according to common root removal methods leaving finest roots in the soil light fraction (Allard et al., 2005; Ostonen et al., 2005; Stevens and Jones, 2006). No additional mineral particles were removed by this procedure. Soil bulk density was determined from the soil sampling in 2004. All soil samples taken with the split tube sampler were weighed and the inner diameter of the soil corer was used for volume calculation.

Total carbon concentration was analyzed on ball-milled subsamples (time 4 min, frequency 30 s<sup>-1</sup>) by an elemental analyzer at 1150 °C (Elementaranalysator vario Max CN, Elementar Analysensysteme GmbH, Hanau, Germany). To determine the organic carbon concentration either the carbonate or the organic compounds had to be removed (Bisutti et al., 2004). We measured inorganic carbon concentration by elemental analysis after removal of organic carbon for 16 h at 450 °C in a muffle furnace. The organic carbon concentration was then calculated from the difference between both measurements (Don et al., 2007). The reliability of this method was tested by measuring certified reference soil material similar to the field site soil (HEKAtech GmbH, Wegberg,

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