



## Original article

# Microbial use and decomposition of maize leaf straw incubated in packed soil columns at different depths

Nils Rottmann<sup>a,\*</sup>, Jens Dyckmans<sup>b</sup>, Rainer Georg Joergensen<sup>a</sup>

<sup>a</sup> Department of Soil Biology and Plant Nutrition, University of Kassel, Nordbahnhofstr. 1a, 37213 Witzenhausen, Germany

<sup>b</sup> Centre of Stable Isotope Research and Analysis, Georg August University Göttingen, Büsgenweg 2, 37077 Göttingen, Germany

## ARTICLE INFO

## Article history:

Received 17 June 2009

Received in revised form

14 September 2009

Accepted 29 September 2009

Available online 8 October 2009

Handling editor: Bryan Griffiths

## Keywords:

Microbial biomass C

Microbial residues

Ergosterol

Particulate organic matter

$\delta^{13}\text{C}$

## ABSTRACT

An experiment was carried out to investigate the decomposition and microbial use of maize leaf straw incubated in packed soil columns at different depths. The straw was incorporated into the top layer at 0–5 cm depth and into the bottom layer at 15–20 cm depth of a sandy or a loamy soil. Microbial biomass C was significantly increased after adding straw to the bottom layer of both soils. After adding straw to the top layer, this increase was significantly lower in the sandy soil and significantly higher in the loamy soil. Maize straw application significantly increased the ergosterol-to-microbial biomass C ratio in both soils from 0.26% to a mean content of 0.72% after adding straw to the top layer and to a mean content of 1.11% after adding straw to the bottom layer. The calculation of the maize-derived CO<sub>2</sub> production revealed that the mineralization rates of maize C were always higher in the sandy soil, with a mean of 20%, than in the loamy soil, with a mean of 14%. The application of maize straw always significantly increased the soil organic matter-derived CO<sub>2</sub> production. This increase was stronger in the loamy soil than in the sandy soil and stronger after application of the maize straw to the top layer than to the bottom layer. On average, 100% of the maize straw C was recovered in the different fractions analysed. In the layers with maize leaf straw application, 28% of the maize C was recovered as particulate organic matter (POM) > 2 mm and 32% as POM 0.4–2.0 mm, without a significant difference between the two soils and the depth of application. In the layers with maize leaf straw application, 19% of the maize C was recovered as microbial residue C and 3.1% as microbial biomass C. In the three layers without straw, the microbial biomass incorporated a further 2.4% of the maize C in the sandy soil, but only 0.9% in the loamy soil. Considerable amounts of substrate C were transferred within the microbial biomass over a decimetre distance. The finer pore space of the loamy soil seems to obstruct the transfer of maize-derived C. This was especially true if the maize leaf straw was added to the bottom layer.

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## 1. Introduction

Decomposition of plant residues and the resulting release of nutrient elements are key functions of soil microorganisms [49]. Straw is an important organic residue added to soil in farming systems without livestock or straw-less livestock keeping. In these systems, the capability of straw decomposition is important to minimize negative effects on the following crop, caused by N immobilization [9,20] and the release of growth-inhibiting low molecular substances [10,27]. Straw is mainly decomposed by saprotrophic fungi [5,9] and forms hot spots of high biological

activity after incorporation into the soil [24,37,47], with an increased demand for nutrients and oxygen.

The oxygen supply for straw decomposition differs under field conditions depending on the depth of incorporation [26]. In reduced tillage systems using, for example, rotary cultivators, the straw is mixed into the first 5–10 cm, whereas the straw is buried mainly at 20–30 cm depth by mouldboard ploughing [48]. Nutrient and oxygen supply to decomposing soil microorganisms is also strongly affected by the soil texture, i.e. sandy soils provide larger pore space with more oxygen, but less nutrients to the microbial community [46,51]. However, not only the detritusphere, i.e. the few millimetres of soil attached to the organic residues [24,37,47] is important for the decomposition of straw, also the surrounding bulk soil may contribute to the nutrient supply [7]. Fungi are able to transport nutrients over relatively large distances within their hyphal network, connecting soil microhabitats rich in

\* Corresponding author. Tel.: +49 5542 98 1517.

E-mail address: [nils.rottman@uni-kassel.de](mailto:nils.rottman@uni-kassel.de) (N. Rottmann).

energy and poor in nutrient supply with microhabitats poor in energy and rich in nutrients [30].

An experiment was carried out to investigate decomposition and microbial use of maize leaf straw incubated in packed soil columns incorporated at different depths. The straw was applied into the top layer at 0–5 cm depth or into the bottom layer at 15–20 cm depth of sandy or loamy soil. This resulted in the following four underlying hypotheses: (1) a higher percentage of substrate is incorporated into the microbial biomass of the loamy soils due to the lower turnover, (2) decomposition and mineralization rates of an added substrate are faster in the sandy soil than in the loamy soils and (3) the differences in microbial use and decomposition of substrate due to the application of substrate to the top or bottom layer are less different in the sandy soil. Maize straw usually differs from soil organic matter in its  $\delta^{13}\text{C}$  value [38,43,44]. In combination with particulate organic matter analysis to recover unused substrate [31], this makes it possible to follow C sequestration into the fractions of microbial biomass and microbial residues, and  $\text{CO}_2$ . It also makes it possible to monitor the translocation of maize-derived C into layers distant from the detritusphere.

## 2. Material and methods

### 2.1. Soil and plant material

The soils used for the experiment were sampled in March 2007 from the upper 15 cm of an experimental site in Angerstein near Göttingen (Southern Lower Saxony, Germany) and an experimental site of the Institute of Biodynamic Research near Darmstadt (Southern Hesse, Germany). The loamy soil of Göttingen was classified as a Haplic Luvisol (FAO-WRB, Food and Agriculture Organisation of the United Nations – World Reference Base for Soil Resources) with the following characteristics: 16% sand, 66% silt, 18% clay, a pH in  $\text{H}_2\text{O}$  of 7.8,  $14.3 \text{ mg g}^{-1}$  total C, a soil organic  $\delta^{13}\text{C}$  value of  $-26.07\text{‰}$ ,  $1.2 \text{ mg g}^{-1}$  total N and a C/N ratio of 11.9. The sandy soil of Darmstadt was classified as a Haplic Cambisol (FAO-WRB) with 79% sand, 16% silt, 5% clay, a pH in  $\text{H}_2\text{O}$  of 7.5,  $11.9 \text{ mg g}^{-1}$  total C, a soil organic  $\delta^{13}\text{C}$  value of  $-26.47\text{‰}$ ,  $1.2 \text{ mg g}^{-1}$  total N and a C/N ratio of 9.9. The field moist soils were sieved (<2 mm) before the experiment was started. Oven dried ( $60^\circ\text{C}$ ) maize leaf straw (*Zea mays* L.) was cut into small pieces of 2–4 mm. It contained 45.3% C with a  $\delta^{13}\text{C}$  value of  $-12.45\text{‰}$ , 0.57% N and  $0.3 \mu\text{g ergosterol g}^{-1}$ .

### 2.2. Experimental design

The six treatments of the experiment were carried out in four replicates in PVC cylinders (15 cm diameter, 20 cm height) in a greenhouse for 57 day: (1) sandy soil with straw at 0–5 cm depth (sand straw top), (2) sandy soil with straw at 15–20 cm depth (sand straw bottom), (3) sandy soil without straw (sand control), (4) loamy soil with straw at 0–5 cm depth (loam straw top), (5) loamy soil with straw at 15–20 cm depth (loam straw bottom), and (6) loamy soil without straw (loam control). Depending on the treatment, the soils were mixed at a concentration of  $20 \text{ mg g}^{-1}$  maize leaf straw into the respective depth layer. For the sandy and loamy treatments, 4444 g and 4161 g dry soil were used per column, respectively, equivalent to 1111 g and 1040 g per layer. The water content was adjusted to 40% of water holding capacity (WHC), which was gravimetrically controlled and adjusted every seventh day, so that the water content did not decrease below 35% WHC. After destructive sampling at the end of the experiment, the water content was measured separately in each layer 7 days after the last water addition, without detecting differences between the different layers. The temperature was kept constant at  $19 \pm 2^\circ\text{C}$  and the bulk density was adjusted to  $1.4 \text{ g cm}^{-3}$ . The PVC cylinders stood on

a sand bed and were closed at the bottom with a nylon gauze (1 mm mesh size) to afford a natural diffusion of  $\text{CO}_2$  as much as possible and to exclude an artificially high  $\text{CO}_2$  accumulation. For sampling, the soil columns were subdivided into layers of 5 cm thickness. In an identical experiment, the cylinders were closed at the bottom with a PVC lid for measuring  $\text{CO}_2$ . This was necessary to catch all  $\text{CO}_2$  for the exact estimation of C sequestration. Both experiments were carried out at the same time, in the same greenhouse room and therefore under the same temperature and light conditions.

### 2.3. Particular organic matter (POM)

Moist soil (400 g) was initially dispersed in 400 ml 5% sodium chloride, shaken by hand and allowed to stand for 45 min [31,34]. Then the samples were poured gradually onto two sieves of 2 mm and 0.4 mm mesh size and washed with tap water. The aggregates were destroyed by pushing the soil through the sieve during the washing procedure until the water passing through the sieve became clear. The material retained on the sieve was transferred into a beaker. Tap water was added, the bucket was swirled and organic material was separated from the mineral material by flotation-decantation. Swirling and flotation-decantation was repeated several times, until organic particles were no longer visible in the mineral fraction. Then, the mineral fraction was discarded. The fractions POM 0.4–2 mm and POM > 2 mm were washed with distilled water and transferred into crucibles, dried at  $60^\circ\text{C}$ , weighed and milled for further analyses.

### 2.4. Carbon dioxide production

Every third day, closed static PVC chambers (15 cm in diameter, 10 cm height) were placed on the cylinders and closed with a rubber-band for 24 h. Inside the chamber, a beaker containing up to 20 ml 2 M NaOH solution was placed on the soil surface. The amount of 2 M NaOH was adjusted according to the expected flux rate so that usually no more than 60% of the absorption capacity was utilised. Total  $\text{CO}_2$  in the alkali traps was determined by back-titrating the excess NaOH to pH 8.3 with 2 M HCl after precipitation of carbonates with  $\text{BaCl}_2$ . The  $\delta^{13}\text{C}$  was determined in precipitated  $\text{BaCO}_3$ . The titration solution was centrifuged at  $4^\circ\text{C}$  and  $9800 \text{ m s}^{-2}$ . The supernatant was discharged and the pellet stirred in 5 ml of distilled water and centrifuged again. This was repeated three times before freeze-drying the pellet containing the precipitated carbon dioxide [28]. The mean  $\delta^{13}\text{C}$  of  $\Sigma\text{CO}_2\text{-C}$  was calculated for each sampling day and averaged at the end of the experiment.

### 2.5. Microbial biomass indices

Microbial biomass C was estimated by fumigation-extraction [50]. A sub-sample of 20 g soil was separated into two portions. One portion was fumigated at  $25^\circ\text{C}$  with ethanol-free  $\text{CHCl}_3$ , which was removed after 24 h. Fumigated and non-fumigated 10-g samples were extracted with 40 ml of 0.05 M  $\text{K}_2\text{SO}_4$  by 30 min horizontal shaking at  $200 \text{ rev min}^{-1}$  and filtered (hw3, Sartorius Stedim Biotech, Göttingen, Germany). Organic C in the extracts was measured as  $\text{CO}_2$  by infrared absorption after combustion at  $850^\circ\text{C}$  using a Dimatoc 100 automatic analyser (Dimatec, Essen, Germany). Microbial biomass C was calculated as follows  $E_c/k_{EC}$ , where  $E_c$  = (organic C extracted from fumigated soil) – (organic C extracted from non-fumigated soil) and  $k_{EC}$  = 0.45 [54]. Microbial residue C was calculated as maize derived soil organic C without POM and maize-derived microbial biomass C.

The fungal cell-membrane component ergosterol was extracted with 100 ml ethanol from 2 g moist soil by 30 min oscillating

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