EARTHWORMS AFFECT MINERALIZATION OF DIFFERENT ORGANIC AMENDMENTS IN A MICRO COSM STUDY

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Abstract

The efficiency of organic amendment applications as a sustainable practice is directly related with activity of soil organisms and their interactions. Earthworms (Lumbricidae) are one of the most important group of soil organisms, which can critically affect organic matter mineralization by incorporating, decomposing and redistributing soil surface organic matter.

To study the decomposition and mineralization of two organic amendments (i.e. barley straw and cattle manure) a number of 30 microcosms was set up using polypropylene tubes filled with soil (FAO: luvisol). Half of the microcosms were inoculated with two species of earthworms (Lumbricus terrestris and Aporrectodea caliginosa), while half remained without animals. Different lengths of ¹³C labelled barley straw and cattle manure were put on the surface of each column. After 45 days of incubation, the organic matter consumption was recorded and earthworm casts and soil (top 10 cm) were analysed.

The results showed that the organic matter consumption rate is higher in manure treatments compared with straw treatments. The amount of consumed manure was about 63% from added quantity in earthworm treatments, respectively 26% in control treatments. The straw consumption rate was far less only 19% and 20% of added straw being consumed in earthworm treatments, respectively 13% and 15% in control treatments. However, the earthworms preferred a straw length of 2.5 cm.

Earthworm activity stimulated microbial biomass, improved Nt and N-NO₃⁻ content of the soil and reduced C/N ratio.

INTRODUCTION

The demands for a sustainable agriculture are bound to introduce changes in production systems of modern agriculture. One of the major challenges of sustainable agriculture is to maintain and improve soil fertility when high production with low costs is expected. A possible solution to this challenge could be the use of organic amendments for a more regenerative agriculture with a better understanding of soil biology and below ground processes (Sherwood, 2000). Even if organic fertilizers are widely used today, more research is needed to understand the nutrient release and their availability for plants uptake. These processes are directly related with soil organism activity and their interactions.

Earthworms are considered as one of the most important faunal group in agroecosystems because of their effects on soil structure formation and on nutrient cycling (Marinissen and de Ruiter, 1993). Lee (1985) reported a density of 5-100 earthworm individuals m⁻² with a corresponding biomass of 0.5-20 g m⁻² fresh weight in cropland. Bouché (1977) defined three main ecological types of earthworms which process soil organic matter differently: epigeic species are small-sized, live in soil organic horizons and preferentially consume litter or dung; anecic species are large sized and mix plant fragments
and mineral particles ingesting during their burrowing through the soil and feeding on the surface; *endogeic* species are medium sized, live in organo-mineral horizons and feed on soil more or less enriched with organic matter. The boundaries between these ecological forms are not sharp and numerous intermediate forms could be described (Jégou et al., 1998). In agroecosystems, anecic and endogeic species reach high densities, affecting significantly the decomposition and mineralization of organic matter. Shipitalo et al. (1987) reported that feeding activity of earthworms and consequently the net effect of their activity is highly dependent on food quality. Accordingly, when different kinds of organic material are used as amendments in agriculture, a favourable effect should appear in earthworm activity.

Mackay and Kladivko (1985), showed that in presence of active earthworm populations the rate of residue breakdown can be increased significantly. When straw was applied to the field, the mass loss could be 21% higher in the presence of earthworms compared with their absence (Jensen, 1985). Curry and Byrne (1992) reported in Ireland a straw mass disappearance of 53% during eight months in the presence of an earthworm population with 57 mature individuals m$^{-2}$. Cortez et al. (1989) reported for *Nicodrilus giardi*, a consumption rate of 4.8-7.1 mg g$^{-1}$ live worm day$^{-1}$ in a laboratory study. Bohlen et al. (1999) showed that *L. terrestris* greatly increased incorporation of manure into the soil after two weeks, while the rye straw incorporation started after 10 weeks. Edwards and Lofty (1982) reported that farmyard manure and sewage cake application increased the earthworm population of four species, with a maximum of 184% more for *L. terrestris*.

For a better understanding of complex interaction between earthworms and different types of organic matter used as amendments in agriculture, a laboratory study using microcosms was conducted at the Institute of Agroecology of the FAL in Braunschweig. The aims of our study were to investigate the relationship between food quality, food consumption and soil proprieties of two different organic sources, commonly used as amendments in agriculture, related with two earthworm species. The study was performed with different lengths of $^{13}$C labelled barley straw and cattle manure as organic sources and two earthworm species, *Lumbricus terrestris* as primary decomposer, and *Aporrectodea caliginosa* as secondary decomposer.

**MATERIAL AND METHODS**

In order to meet the objectives, a number of 30 microcosms was conducted using polypropylene columns (30 cm high, 12 cm in diameter) filled with soil. The soil was a luvisol derived from loess (clay 12%, silt 85%, sand 3%) with a pH (CaCl$_2$) of 6.3, a total organic carbon content (Corg) of 1.4%, a total nitrogen content (Nt) of 0.14% and a carbon/nitrogen ratio (C/N) of 10. The soil was collected from field and sieved through a 5 mm mesh sieve to remove large fragments of organic matter. After sieving, the soil was frozen at -20°C for seven days, than thawed at room temperature for 3 days, than frozen again for seven days. This freezing-thawing cycle killed the soil fauna whereas some nematoda and protozoa may have survived (Huhta et al., 1989). After freezing – thawing procedure the columns were filled with soil to a bulk density of 1.25 g cm$^{-3}$. The water content of the soil was fit at 20% and the bottom part of the columns was closed with a 20µm mesh.

Than, the organic sources were added to the columns in a quantity of 9.3 g for each microcosm. We used different lengths of $^{13}$C labelled barley straw (C/N ratio 30) and cattle manure (C/N ratio 12) which were spread uniformly at the soil surface of the columns. The first treatment (T1) received a mixture of 2.5 cm and 10 cm lengths of straw in equal quantities (i.e. 4.15 g), the second treatment received a mixture of 2.5 cm, 5 cm and 10 cm
lengths of straw in equal quantities (i.e. 3.1 g), while the third treatment (T3) received 9.3 g of cattle manure. The control treatments (C1, C2, C3) received the same amount and combination of organic material like T1, T2, T3. Immediately after addition of organic material, a combination of two earthworm species, one individual of *Lumbricus terrestris* and two individuals of *Aporrectodea caliginosa*, were inoculated in earthworm treatments (T1, T2, T3) while non-earthworm treatments (C1, C2, C3) remained without earthworms. The worms were collected from a field site, and maintained in the same type of soil like that used in experiment, for a two week adaptation period. Before inoculation the earthworm weight was recorded.

The microcosms were set up in an experimental design with three different organic treatments (two lengths of barley straw, three lengths of barley straw and cattle manure) and two earthworms treatments (with and without earthworms). Each treatment had 5 replicates resulting in a total number of 30 microcosms. The microcosms were incubated at dark and 15°C during 45 days.

After the incubation period the remaining organic material was collected and weighted and the community consumption rate in mg food g⁻¹ live weight day⁻¹ was recorded. After removing of organic material, 15 g of casts were collected from the soil surface. The casts were dried at 110°C overnight for total carbon (Ct) and nitrogen (Nt) analysis. Then, the first 10 cm of the soil columns were sampled and the columns destroyed carefully to collect the earthworms. The earthworms were weighted, killed by freezing and kept at -20°C for further analyses.

From each column, the collected soil was separately processed according with the analyses which were made it: 10 g of soil was kept at 4°C for microbial biomass analysis, 25 g was frozen for N-NO₃⁻ analysis, 15 g was air dried for pH measurement and 25 g was dried at 110°C overnight for Ct and Nt measurement.

The microbial biomass was estimated using substrate induced respiration method (SIR) described by Anderson and Domsch (1978). N-NO₃⁻ measurement was done with a Skalar-photometer after extraction with 250 ml 0.0125M CaCl₂. The pH of the soil was measured electrochemically in CaCl₂ solution with Sartorius Professional Meter PP-25. Ct and Nt was measured by dry combustion with LECO TruSpec CN.

**RESULTS AND DISCUSSIONS**

During the incubation period, the earthworm biomass decreased to a weight loss of 17% in T1, 11.5 % in T2, respectively 10.8 % in T3. A decreasing in weight was reported also by Shipitalo et al (1988), Cortez et al. (1989), Bohlen et al. (1995). The greatest decrease occurred in T1 while the smallest was observed in manure treatment (T3). Bohlen (1995) suggested that microcosm experiments can stress earthworms so that they refuse food for a short period especially when food quality is low. Straw is known to be a poor source of nitrogen comparing with manure so that straw is less palatable for earthworms. This was also suggested by Cortez et al. (1989) who reported that straw became more palatable for earthworms during incubation when it was partially decomposed.

The quantity of organic material which disappeared from the soil surface during the incubation, expressed in percent from initial amount, varied between treatments being quite similar in straw treatments, i.e. 19% respectively 21%, and reached 63% in manure treatment. As we already mentioned, the manure is consumed by organisms immediately after application, while straw is consumed after some time.
The community consumption rate expressed as mg food g\(^{-1}\) live weight day\(^{-1}\) was two times greater in manure treatment compared with straw treatments (Table 1). The values recorded for straw consumption were smaller than those reported by Cortez et al. (1989) (4.8 – 7.1 mg food g\(^{-1}\) live weight day\(^{-1}\) for *Nicodrilus giardi* fed with wheat straw) but are quite similar with those reported by Shipitalo et al. (1988) for *L. terrestris* fed with corn leaves and bromegrass leaves (6 respectively 2 mg food g\(^{-1}\) live weight day\(^{-1}\)). A significant difference (\(p < 0.01\)) was found for a consumption rate of 2.5 cm straw length comparing with 5 cm, respectively 10 cm straw lengths in both straw treatments, indicating that earthworms preferred the 2.5 cm straw length.

Tab. 1. Earthworm biomass and consumption rate of earthworms community feeding on barley straw of different lengths and cattle manure (mean±SD)

<table>
<thead>
<tr>
<th>Treatment</th>
<th><em>A. caliginosa</em> biomass (g)</th>
<th><em>L. terrestris</em> biomass (g)</th>
<th>Total earthworm biomass (g)</th>
<th>Community consumption rate (mg food g(^{-1}) live wt. d(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2.5 cm straw</td>
<td>5 cm straw</td>
<td>10 cm straw</td>
<td>Manure</td>
</tr>
<tr>
<td>T1</td>
<td>1.49</td>
<td>5.07</td>
<td>6.57</td>
<td>3.8±0.7</td>
</tr>
<tr>
<td>T2</td>
<td>1.07</td>
<td>5.14</td>
<td>6.21</td>
<td>3.2±0.5</td>
</tr>
<tr>
<td>T3</td>
<td>1.69</td>
<td>5.96</td>
<td>7.66</td>
<td>1.9±0.5</td>
</tr>
</tbody>
</table>

Comparing with the start values, after 45 days of incubation all soil parameters had a significant difference (\(p < 0.01\)) indicating that organic amendments added to the soil have affected soil proprieties. This effect was more evident for pH and N-NO\(_3\) values (Fig.1, Fig.2).

There is a significant difference (\(p < 0.01\)) between earthworm treatments (T1, T2, T3) and non earthworm treatments (C1, C2, C3) regarding soil Nt values, extractable soil N-NO\(_3\) and soil microbial biomass. Earthworms did not affect significantly pH values and soil organic carbon content (Table 2).

According with our data, earthworms increased N mineralization during 45 days for 1.3 %, 4.4 %, 5.9 % in T1, T2 and T3. An increase in N mineralization as result of earthworm activity was also reported by other authors (Bohlen et al 1994, Blair et al. 1997). The effect of earthworms on nitrogen mineralization could be explained by direct consumption of organic material, digesting/excreting process, casting activity, favourable effects on microbial activity and influences on soil properties (Marinissen and Ruiter, 1993). In our experiment, the higher mineralization rate in earthworm treatments (T1, T2, T3) could be explained by higher consumption rate and a higher microbial biomass determined. As a result of nitrogen
mineralization, extractable N-NO$_3^-$ values are higher in earthworm treatments compared with control (Table 2). The nutrient source and earthworm activity had a significant effect on N-NO$_3^-$ extractable amounts with greater values in manure treatments compared with the straw treatments. This could be explained by the differences in quality of organic sources used as amendments, manure having a high amount of nitrogen which could be mineralized during decomposition.

Table 2. F values and levels of significance (p < 0.05) from 2-ANOVA for each factor (organic matter type and earthworms) and each dependent variable (soil parameters)

<table>
<thead>
<tr>
<th></th>
<th>Microbial biomass</th>
<th>N-NO$_3^-$</th>
<th>Corg</th>
<th>Nt</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>OM type</td>
<td>19.14*</td>
<td>39.7*</td>
<td>4.29</td>
<td>5.38</td>
<td>5.81*</td>
</tr>
<tr>
<td>Earthworms</td>
<td>90.75*</td>
<td>68.08*</td>
<td>1.46</td>
<td>12.66*</td>
<td>1.09</td>
</tr>
<tr>
<td>OM type × earthworms</td>
<td>1.17</td>
<td>11.19*</td>
<td>1.48</td>
<td>0.9</td>
<td>1.04</td>
</tr>
<tr>
<td>R$^2$</td>
<td>0.84</td>
<td>0.87</td>
<td>0.35</td>
<td>0.51</td>
<td>0.38</td>
</tr>
</tbody>
</table>

Soil microbial biomass was significantly higher (p < 0.01) in earthworm treatments than in non earthworm treatments (Fig. 3, Table 2). Earthworms increased soil microbial biomass carbon with 18%, 13% respectively 17% in T1, T2, T3 compared with corresponding control treatments. Other authors (Scheu, 1987; Anderson, 1988; Bohlen et al. 1997) reported also a higher microbial biomass in the presence of earthworms, while Bohlen and Edwards (1995) showed that soil microbial nitrogen could decrease in the presence of earthworms. Ruz-Jerez et al. (1992) reported also a decrease in microbial biomass carbon after 11 weeks of laboratory incubation when earthworms were present. In our case, the higher microbial biomass could be explained by a relative high consumption rate and a corresponding higher casting activity (data not shown). Microbial biomass is higher in earthworm casts than in surrounding soil as Scheu (1987) reported, so that we can expect a higher microbial biomass when cast amounts are high. It might be expected that microbial biomass would have been higher in manure treatments than in straw treatments. Actually, the straw treatments (T1, T2) have a higher microbial biomass carbon than the manure treatment (T3) (Fig. 3). Different quality of organic sources could be responsible for that. The manure contained readily mineralizable substrates that increased microbial biomass shortly after application. Bohlen et al. (1999) reported a high peak of microbial biomass nitrogen two weeks after manure adding and a decrease during the next eight weeks. It seems that a higher microbial biomass carbon in our experiment was reached around the middle of the incubation period and decreased thereafter. On the other hand, straw is usually poor in nutrients (Wessen and Berg, 1986) and the decomposition process may take a long time.

The recalcitrant compounds, (i.e lignin) require a greater investment of energy to be decomposed, so that we could assume that microbial growth started after some time in straw treatments and reached a higher level at the end of the experiment. As we already discussed, we belive that earthworms started feeding on straw in the late part of the experiment, when
straw became more available. This period corresponds with the higher microbial biomass carbon value in straw treatments.

CONCLUSIONS

Results of our experiment indicated that earthworm feeding activity was dependent on available organic sources, earthworms consuming more cattle manure than straw. The smallest straw length (2.5 cm) was consumed more intensively compared to the longest straw pieces. This fact should be taken into account for mulching measures in the field.

Earthworms activity increased mineralization rate of organic sources and had influence on soil properties such as microbial biomass, nitrate, total nitrogen and pH.

BIBLIOGRAPHY

14. Ruiz-Jerez B. E., P. R. Ball, R. W. Tillman, 1992, Laboratory assessment of nutrient release from a pasture soil receiving grass or clover residues, in the presence or absence of Lumbricus rubellus or Eisenia fetida, Soil biology & biochemistry, Vol. 24, No. 12, p. 1529-1534