Opusc. Zool. Budapest, 2017-2018, 48(Supplementum 2): 73–81

Data on the Lumbricidae and Enchytraeidae from six forest sites in Germany and Portugal

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Abstract. The complex interactions between primary production, soil and soil organisms (in particular Lumbricidae and Enchytraeidae) during organic matter decomposition have developed over evolutionary time under specific habitat conditions. Thus, in order to study these processes, four forest sites in Germany (two in southern Hesse, two in eastern Saxony) and two sites in Central Portugal were selected to study both the oligochaete fauna and litter decomposition (almost all of them with a pH <4). Here, the results of the first part of this his work, i.e. the abundance and the species composition of the oligochaete fauna, are described. All samplings were performed using standard ISO-methods, with different temporal intensity. In detail, nine common Central European earthworm species were found at the six study sites plus one unidentified species at a Portuguese site. Abundance was usually low (< 10 Ind/m^2); higher numbers (72.5 and 17.5 Ind/m^2) were detected only at the two Saxonian sites. In total, 53 enchytraeid species were found; 43 of them already described. At four sites densities fell within the expected range for German deciduous forests (20 000 – 80000 Ind/m^2). Finally, notes on the taxonomy and biogeography of several potworm species are given.

Keywords: Annelida, species number, species composition, global climate change

INTRODUCTION

According to recent research on climate-change scenarios, Central European weather will become more extreme, with intermittent very hot and dry summers during the next 50−100 years (Schär *et al*. 2004, Schmidli *et al*. 2007). Tree species currently used in forestry are not adapted to such conditions, thus drought-resistant Mediterranean tree species might be an alternative (Pflug & Brüggemann 2012, Bussotti *et al.* 2015). Litterfall from trees represents a key resource for the below-ground ecosystem, but it is not known whether the complex process of litter decomposition, which depends on the soil organism community (mainly bacteria, fungi and a wide range of soil invertebrates), is or will be impacted by these climatically driven changes in forest composition (Blankinship *et al*. 2011 but see Wall *et al.* 2008). Soil organism communities are adapted in various ways to local conditions, *e.g*. soil properties or climatic factors. As part of the German BiK-F project (www. www.bik-f.de; today belonging to the Senckenberg Museum, Frankfurt), the question was raised whether climate-change driven transformations of forest-tree composition will affect these organisms and their functions, such as their role in litter decomposition. Specifically, one of the research questions is whether soil organisms in Germany, having been adapted to litter from Central European tree species, such as beech (*Fagus sylvatica*) and oaks, *e.g. Quercus pubescens*, can handle litter from southern European trees (mainly oaks). Lumbricid and enchytraeid species were selected as representatives of the whole community due to their important role

urn: lsid:zoobank.org:pub:915C27D1-178B-4186-A2D4-B68E8C2BCBE9 *published: 11, May 2018*
HU ISSN 2063-1588 (online), HU ISSN 0237-5419 (print) *http://dx.doi.org/10.18348/opzool.2017.S2.73*

in litter decomposition under central European conditions (Blouin *et al*. 2013).

The oligochaete studies were performed at different intensities in six forest sites (two in Saxony [eastern Germany], two in Hesse [western Germany] and two in Central Portugal), representing areas with different climate systems. Thus, the main question of the whole project was: will the local soil organism community be able to decompose "foreign" litter, or will local soil nutrient cycles be disrupted after introduction of the "foreign" trees? In this paper we report on the description of the oligochaete fauna at the six study sites in Germany and Portugal. Abundance and community composition of the fauna are important for the interpretations of the litter decomposition studies which are still in progress.

MATERIAL AND METHODS

In order to address the research question, three geographical regions in Europe were selected, one in the continental and one in the Atlantic region of Germany, and one in the Mediterranean region (Central Portugal). The study sites in eastern Germany are located in the continental climate zone with a mean annual yearly temperature (MAT) average of 8.5°C. These average increases by approx. 2°C when shifting to the sites in western Germany, representing the oceanic climate zone. An additional further 3°C increase in yearly temperature MAT is found at the study sites in Portugal which are influenced by the Mediterranean climate zone (Peel *et al.* 2007). These differences are within the range of predicted temperature increases for Germany and Portugal.

In each region, two study sites with four 20×20 m plots each were selected in order to represent different forest histories and conditions within a climatic region, *i.e.* forests were only replicated within sites but not necessarily between sites of an area. The two eastern German sites are located in the state of Saxony: Viereichen and Ullersdorf, both near Görlitz. They represent two undisturbed deciduous forests, mainly consisting of different species of oaks (mainly *Quercus robur* with an age of about 100 years), hornbeam (*Carpinus betulus*) and few common beech (*Fagus sylvatica*) trees. At Viereichen, rarely small leaf linden (*Tilia cordata*) does occur.

The two south-western German sites are located in the state of Hesse: Rüsselsheim (disturbed mixed forest near human settlements) and Lampertsheim (mixed deciduous forest in the Rhine valley, very sandy soil and dry conditions). These two were most intensively studied. The Rüsselsheim site is originally an open *Pinus sylvestris* forest containing solitary old (>80 years) *Quercus robur* trees. After disturbed by fire in 2008, the area was partly re-planted with *Q. ilex*, *Q. frainetto* and *Q. pubescens*. At Lampertheim, the dominant tree species is also *Q. robur* (*ca*. 110 years old). Saplings of the same oak species as in Rüsselsheim (plus *Q. petraea*) where planted just outside of the undisturbed sampling area (Dorow *et al*. 2012). St. Olaia is a calcareous hill in the coastal plain that has been continuously occupied by forest since the Neolithic times. The vegetation is mainly composed by the tree species *Quercus fagineae* and *Quercion ilicis* and various herb and shrub species (*e.g*. *Vinca difformis*). Cerdeira has been occupied by forest since about 50 years ago when the area was no longer explored for agriculture. The vegetation is mainly composed by the tree species *Quercus robur*, *Q. pyrenaica*, *Castanea sativa*, *Pinus pinaster* and *Eucalyptus globulus* and the bracken fern (*Pteridium aquilinum*).

The main characteristics of the six study sites and their most important soil properties are given in Table 1. Soils differ strongly in terms of texture, mainly regarding the percentage of sand and silt. The clay content differs only slightly (*i.e.* between 5 and 10%). Except for St. Olaia, which had a pH of 5.0, all other sites had acid soils with pH-values between 3.2 and 3.7. Regarding the organic matter content, a wide range was covered, from 6.2 % in Lampertheim up to 21.3 % at Cerdeira. Despite these differences, the C/N-ratio was not strikingly different, ranging from 13.1 in Ullersdorf and 20.3 in Rüsselsheim. Soil moisture was measured gravimetrically (*i.e.* loss in % weight after drying at 105°C) in parallel to the individual oligochaete samplings. It can change quickly, but the two Hessian sites are certainly drier than the other four sites.

Table 1. Main characteristics and most important soil properties of the six study sites. Mean temperature = mean of daily measures during the study period. Mean annual precipitation (mm/year) data are long-term averages collected from the nearest climate station.

Property	Viereichen	Ullersdorf	Rüsselsheim	Lampertheim	Cerdeira	St. Olaia
Region	**Saxony (Germany)**		**Hesse (Germany)**		**Central Portugal**	
Coordinates	51°22′54” N 14°41′54” E	51°14′15” N 14°50′27” E	49°57′13” N 8°24′54” E	49°35′08” N 8°28′46” E	40°09′43” N 8°19′41” W	40°17′06” N 8°71′52” W
Text. Sand (%)	62.3	19.5	67.8	75.4	35.1	48.9
Text. Silt (%)	32.2	70.2	21.9	19.0	57.2	43.0
Text. Clay (%)	5.5	10.3	10.3	5.6	7.7	8.1
Texture class	Medium silty sand	Weakly loamy silt	Medium loamy sand	Weakly silty sand	Sandy silt	Silty-loamy sand
pH ($CaCl_2$)	3.7	3.2	3.5	3.6	3.5	5.0
Organic Material (%)	9.5	13.4	9.4	6.2	21.3	16.7
C/N	14.9	13.1	20.3	17.1	15.7	13.0
Ø Soil Moisture (%)	30.1	79.5	37.8	30.9	41.8	30.6
Ø Temperature (C°)	8.4	9.9	11.1	7.3	12.8	15.1
Ø Ann. Precipitation	672	635	658	695	958	900

Sampling of soil organisms

Earthworms (Lumbricidae) were sampled according to the ISO standard 23611-1 (2006), *i.e.* by a combination of hand-sorting and formalin extraction. Five randomly selected samples were taken per plot. The two Hessian sites were sampled 2009, twice in spring and autumn each. The remaining four sites (two in Saxony and two in Portugal) were sampled once in April and June 2014, respectively. Each individual worm was morphologically determined using the keys of Graff (1953), Bouché (1972) and Sims & Gerard (1999). The worms were stored in 90% ethanol.

Potworms (Enchytraeidae) were sampled according to the ISO standard 23611-3 (2006) with soil-corers (Ø 5.5 cm). Ten samples were randomly taken per plot. Each sample was divided to litter layer and 0–4 cm mineral soil layer. The Hessian sites were sampled four times between autumn 2008 and spring 2010, while the others were sampled only once, in April or June 2014. Potworms were collected by wet extraction in the laboratory at room temperature, about 20°C. Extraction duration for litter-layer samples was 1–2 days and for mineral soil samples was 3–4 days. Morphological identification was performed using the key and checklist of Schmelz & Collado (2010; 2012).

RESULTS

Abundance, species number and composition of earthworms

All three pairs of sites and, also, the two sites within a region differed considerably in abundance and/or species number (Table 2). In Hesse, very low density (0.4 – 3.4 Ind/m^2) and species richness were found. In Saxony we found considerable differences between the two sites. At Viereichen both abundance and species number were high (80.5 Ind/m^2; 6 species), but at Ullersdorf the respective numbers were clearly lower (17.5 Ind/m^2; 3 species). In Portugal, the differences were even more pronounced; at Cerdeira almost no earthworms were found (0.4 Ind/m^2; 1 species), while at St. Olaia both parameters were much higher (7.9 Ind/m^2; 4 species).

Abundance, species number and composition of enchytraeids

Due to the different sampling efforts at the six sites, it is difficult to compare the results directly, but the number of enchytraeids was in tendency, with two exceptions, in a similar range, i.e. about 40 000 to 50 000 Ind/m^2 (Tables 3 and 4). In Rüsselsheim their average number was almost always lower (about 25 000 Ind/m^2), while at Viereichen on average twice as many enchytraeids were found as at all other sites.

Table 2. Abundance (Ind/m^2), standard deviation (SD), species number and composition of earthworms at the six study sites. Note that earthworms were sampled four times at the two Hessian sites, but only once at the other sites (therefore, mean values are given for Rüsselsheim and Lampertheim). n.d. = not determined due to low numbers

Genus / Species	Vieleichen	Ullersdorf	Rüsselsheim		Lampertheim		Cerdeira	St. Olaia
	Spring 2014	Spring 2014	Autumn 2009	Spring 2010	Autumn 2009	Spring 2010	Spring 2014	Spring 2014
Aporrectodea s.l. sp.	23.0	0.5	-	-	-	-	-	2.5
Aporrectodea caliginosa (Savigny, 1826)	14.5	-	-	-	-	-	-	-
Aporrectodea rosea (Savigny, 1826)	0.5	0.5	-	-	-	-	-	3.0
Lumbricus sp.	12.0	4.5	2.0	-	1.3	-	-	-
Lumbricus rubellus Hoffmeister, 1843	14.0	3.5	0.8	1.2	1.3	0.4	0.4	-
Lumbricus terrestris Linnaeus, 1758	1.5	-	-	-	-	-	-	-
Dendrobaena/-drilus sp.	2.5	0.5	-	-	-	-	-	-
Dendrobaena attemsi (Michaelsen, 1902)	4.0	5.0	-	-	-	-	-	-
Dendrobaena octaedra (Savigny, 1826)	-	-	-	-	0.5	-	-	-
Dendrodrilus rubidus (Savigny, 1826)	0.5	-	-	-	-	-	-	-
Octolasion sp.	-	-	-	-	-	-	-	0.5
Octolasion cyaneum (Savigny, 1826)	-	-	-	-	-	-	-	1.5
Octolasion tyrtaeum (Savigny, 1826)	-	-	-	-	0.3	-	-	-
Unknown species	-	-	-	-	-	-	-	0.5
Undetermined	8.0	3.0	0.5	-	-	-	-	-
Total ± SD.	80.5 ± 32.2	17.5 ± 3.4	3.3 ± 1.7	1.2 n.d.	3.4 ± 2.1	0.4 n.d.	0.4 n.d.	8.0 ± 2.0
Species number per site	6	3	1	1	3	1	1	4

Table 3. Abundance (Ind/m^2) and standard deviation (SD) of enchytraeids at Rüsselsheim and Lampertheim

		Hesse (Germany)	
Year	**Season**	**Rüsselsheim**	**Lampertheim**
2008	Autumn	21 520 ± 3 475	75 640 ± 8 286
2009	Spring	41 940 ± 5 841	57 800 ±2 800
2009	Autumn	13 920 ± 2 994	13060 ± 715
2010	Spring	22 700 ± 7 569	48 440 ± 12 517
Average	-	**25 020 ± 11 232**	**48 735 ± 26 324**

Enchytraeid abundance was fairly similar at the two Portuguese sites. At the two Hessian sites, samples were taken four times between the autumn of 2006 and the spring of 2011. Variation in abundance were less pronounced in Rüsselsheim (variation of a factor of three) than in Lampertheim (variation of a factor of six), but no specific pattern was visible.

In total, 53 enchytraeid species were identified (Table 4). Forty-three of them have already been described or are known to science; the remaining ten require descriptions. Not surprisingly, all but one of the undescribed taxa were found at the two Portuguese sites. Enchytraeids of Portugal are poorly known (Schmelz & Collado 2013). The number of enchytraeid species per site varied between 11 (Cerdeira) and 27 (St. Olaia); species numbers at the German sites were in-between. Thirty and 31 species, respectively, were only found at the sites in Germany (four sites) or in Portugal (two sites). Eleven species out of 52 occurred both in Portugal and Germany. Only one species, *Enchytronia parva*, was found at all six sites. At most sites no (Rüsselsheim) or few, *i.e.* 1-4 (Lampertheim, Viereichen, Ullersdorf, and Cerdeira) species occurred which were not found at other sites. The exception is St. Olaia, where 20 species were found which were not found at other sites.

DISCUSSION

Abundance, species number and composition of earthworms

All but one of the earthworm species found are well-known throughout Europe, *i.e.* no species with a restricted regional distribution was detected. The exception was one small worm found at St. Olaia, which was not well enough preserved to be determined. It resembled *Proctodrilus antipai*, a central-European species occurring between central France and the Black Sea (Bouché 1972, Csuzdi & Zicsi 2003).

The low abundances and species numbers at most sites can be explained by the acidic soils with pH-values mainly below four, as indicated by typical acidophilous species of the genera *Dendrobaena/Dendrodrilus* and, partly, *Lumbricus*. The low abundance at the Hessian and Portuguese sites can be explained by site-specific characteristics, *i.e.* very sandy and dry conditions soil in Lampertheim, human disturbance in Rüsselsheim and shallow soil layers, partly on slopes, in Cerdeira. With such low abundances, it is clear that the ecological role of earthworms at these sites is small (*cf.* Blouin *et al.* 2013). The exception is Viereichen with a similarly low pH, but with twice the average earthworm densities reported for German deciduous forests (Jänsch *et al.* 2013). This situation cannot be presently explained.

Abundance, species number and list of enchytraeids

Enchytraeid densities at the six sites varied between 25 000 Ind/m^2 (Rüsselsheim) and almost 100 000 Ind/m^2 (Viereichen). Three out of the four German sites as well as both Portuguese sites are within the "normal" range (51 241 ± 30 677 Ind/m^2) for German deciduous forests, as determined in a recent literature review (Römbke *et al.* 2013). According to the same source, such German deciduous forests "should" harbor 12.4 ± 5.5

Table 4. Abundance (Ind/m^2), standard deviation (SD.), number and composition of enchytraeid species at the six study sites (mean values for the Hesse sites). Species names with four letter codes in brackets indicate forms with new character combinations, probably new species.

Genus / Species	**Saxony (Germany)**		**Hesse (Germany)**		**Portugal**	
	Vier-eichen	**Ullers-dorf**	**Rüssels-heim**	**Lam-pertheim**	**Cerdeira**	**St. Olaia**
Achaeta abulba Graefe, 1989	-	80	1050	10480	-	-
Achaeta affinis Nielsen & Christensen, 1959	22940	1000	10920	16600	12960	-
Achaeta bibulba Graefe, 1989	-	-	220	260	-	-
Achaeta bifollicula Chalupský, 1993	-	-	860	80	-	-
Achaeta camerani (Cognetti, 1899)	820	11580	380	1580	-	-
Achaeta unibulba Graefe, Christensen & Dózsa-Farkas, 2005	-	-	-	-	-	60
Achaeta sp. (HEAL)	-	-	-	-	-	600
Achaeta sp. (PAFF)	-	-	-	-	-	2680
Achaeta sp. (PAFU)	-	4560	-	-	-	-
Bryodrilus ehlersi Ude, 1892	-	-	10	170	-	-
Buchholzia appendiculata (Buchholz, 1862)	4640	-	-	-	-	14840
Buchholzia sp. (OLAI)	-	-	-	-	-	1080
Cernosvitoviella cf. *minor* Dózsa-Farkas, 1990	-	-	-	-	-	20
Cognettia cognettii (Issel, 1905)	-	60	-	40	12200	-
Chamaedrilus/ Cognettia chlorophilus * Friend, 1913	7000	17020	5410	6180	-	320
Enchytraeus bigeminus Nielsen & Christensen, 1963	-	-	-	20	-	-
Enchytraeus buchholzi Vejdovsky, 1879	1420	-	-	-	540	3000
Enchytraeus bulbosus Nielsen & Christensen, 1963	-	-	-	-	-	240
Enchytraeus sp. (GRAN)	2020	-	10	90	820	1240
Enchytraeus norvegicus Abrahamsen, 1969	11020	120	70	165	1760	-
Enchytronia parva Nielsen & Christensen, 1959	8440	180	340	2090	1500	840
Enchytronia sp. (MINO)	-	-	-	-	-	120
Enchytronia sp. (TENU)	120	-	-	-	160	-
Enchytronia pygmaea Graefe & Schmelz, 2017						100
Fridericia auritoides Schmelz, 2003	-	-	-	-	-	720
Fridericia benti Schmelz, 2002	60	-	-	-	-	-
Fridericia bisetosa (Levinsen, 1884)	2980	-	-	-	-	1980
Fridericia bretscheri Southern, 1907	-	-	-	-	-	2460
Fridericia brunensis Schlaghamerský, 2008	320	-	30	-	-	-
Fridericia bulboides Nielsen & Christensen, 1959	1220	-	-	180	-	-
Fridericia ciliotheca Schmelz & Collado, 2013	-	-	-	-	-	460
Fridericia connata Bretscher, 1902	240	-	20	-	-	200
Fridericia cylindrica Springett, 1971	-	-	-	-	-	40
Fridericia dura (Eisen, 1879)	620	-	-	-	-	-
Fridericia isseli Rota, 1994	-	-	-	-	-	340
Fridericia larix Schmelz & Collado, 2005	-	-	-	-	-	160
Fridericia monochaeta Rota, 1995	-	-	-	-	800	300
Fridericia paroniana Issel, 1904	-	-	-	-	-	5520
Fridericia ratzeli (Eisen, 1872)	740	-	20	60	-	-
Fridericia striata (Levinsen, 1884)	280	-	-	90	3,120	1780
Fridericia tuberosa Rota, 1995	-	-	-	-	-	240
Fridericia sp. (PANO)	-	-	-	-	1,080	-
Fridericia sp. (PERT	-	-	-	-	-	1980
Marionina argentea (Michaelsen, 1889)	-	-	-	-	-	40
Marionina clavate Nielsen & Christensen, 1961	-	5100	5340	1160	-	-
Marionina filiformis Nielsen & Christensen, 1959	-	-	-	60	-	-

Genus / Species	Saxony (Germany)		Hesse (Germany)		Portugal	
	Vier-eichen	Ullers-dorf	Rüssels-heim	Lam-pertheim	Cerdeira	St. Olaia
Marionina simillima Nielsen & Christensen, 1959	-	-	-	-	5460	-
Marionina sp. (SIM?)	20	-	-	-	-	-
Mesenchytraeus glandulosus(Levinsen, 1884)	40	-	-	-	-	-
Mesenchytraeus pelicensis Issel, 1905	120	80	30	-	-	-
Oconnorella cambrensis(O'Connor, 1963)	-	6680	310	7800	-	-
Oconnorella tubifera (Nielsen & Christensen, 1959)	14360	220	-	1400	-	-
Stercutus niveus Michaelsen, 1888	-	80	-	-	-	-
Abundance (Ind/m^2)	**95820 ± 37300**	**46760 ± 29470**	**25020 ± 11232**	**48735 ± 26324**	**40400 ± 26545**	**41360 ± 26483**
Species number per site	**21**	**13**	**16**	**19**	**11**	**27**

*= *Cognettia sphagnetorum* auct. (partim). See Martinsson *et al.* (2015) for details. The valid genus name is controversial and awaits a ruling by the International Commission on Zoological Nomenclature.

species. Our species number range, 13–21 fits very well to this average richness. To our knowledge, no quantitative studies on enchytraeid abundance and community structure have been carried out at Portuguese forest sites in general so far.

Regarding the biogeographical distribution, it seems that, despite the large geographical distance between the German and Portuguese sites, there is considerable overlap in faunal composition, although the overlap is smaller than in the case of earthworms. The large number of previously known species in Portugal confirmed previous records, reviewed in Schmelz & Collado (2013), who list altogether 32 species for Portugal. Two third of the species have also been recorded in Germany, thus most species belong to both the Portuguese and the German enchytraeid fauna. The high number of species found only in St. Olaia (18), including five species not yet described, indicates that the Portuguese enchytraeid fauna is far from being fully explored. In contrast to Central Europe, many new species were found not only in the species-rich genus *Fridericia* but also in genera such as *Achaeta* or *Enchytronia*. This is especially important when planning to use enchytraeids as indicators of soil quality (i.e. in terms of soil biodiversity). Species already known from other areas can be classified ecologically (*e.g.*, several species found at Cerdeira such as *A. affinis* or *E. norvegicus* are known to be acidophilous).

Five of the six study sites have a pH <4 and the sixth one, St. Olaia, is also acidic, although less so (Table 1), meaning that a relatively small number of acidophilous species (often in high numbers) are expected to occur there. However, it is not clear why at five out of six sites the species number is higher than expected. This may partly be explained by site-specific characteristics which, however, are difficult to identify. For example, in Lampertheim the vegetation pattern is very heterogeneous at a small scale, meaning that open spots and typical forest areas intermingle, providing different ecological niches at small scales. Rüsselsheim, on the other hand, is a forest located close to a densely populated city area, which – after a fire – had partly been afforested about 20 years ago. Thus, it can be considered to represent a disturbed site. However, while this stress may explain the low enchytraeid abundance, it seems to be contradictory to the relatively high species number.

The Cerdeira site is located in a mountainous region, often with shallow (*i.e.* less than 10 cm) soils and steep hills. This means that soils regularly dry up, which may explain the overall low abundance in comparison to such forests in general. A preliminary sampling in summer with dust-dry soil yielded no enchytraeids. Typical species of such acid sites are: *Ac. affinoides*, *Co. cognettii, En. norvegicus* and *Fr. striata*. New geographical elements (to be checked with more

sites) are *Fr. monochaeta*, *Fr.* (PANO), and *Ma. simillima*.

The forest of St. Olaia is the last remnant of a larger forest, being located on a small hill surrounded by agricultural land. The dominance of species from the genera *Enchytraeus* and *Fridericia* indicates neutral soils, which is interesting since the soil pH was measured as being just 5.0. In addition, the absence of *Henlea* species indicates dry conditions. New zoogeographical elements (to be checked ecologically with more sites) are represented by *Fr. auritoides*, *Fr. bretscheri*, *Fr. ciliotheca*, and *Fr. monochaeta*.

Finally, some special remarks on the taxonomy and biogeography regarding the two Portuguese sites can be made. At St. Olaia, *Fr. ciliotheca*, recently described from Coimbra (about 80 km to the west), was found for the first time following its original description. *Fr. larix* was found for the second time following its original description. Thus far it is only known from Ireland (original description) and England (unpublished). Possibly, it is a truly Atlantic species. The record of *Enchytronia pygmaea* at St.Olaia was included in the original description of the species (Graefe & Schmelz 2017).

Acknowledgements − We thank all colleagues in Flörsheim, Görlitz and Coimbra who helped with the field samplings and the participants of the 12th International Symposium on Enchytraeidae (Tihany, Hungary (27–29 June 2016)) for their contributions when discussing this issue. In addition, we would like to thank the referees for their very constructive remarks.

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