



HAL
open science

Wildflower strips of 2.5-year-old promote earthworms and enchytraeids (Annelida, Oligochaeta) in arable fields

Céline Pelosi, Michel Bertrand, Antoine Gardarin

► To cite this version:

Céline Pelosi, Michel Bertrand, Antoine Gardarin. Wildflower strips of 2.5-year-old promote earthworms and enchytraeids (Annelida, Oligochaeta) in arable fields. *European Journal of Soil Biology*, 2024, 122, pp.103644. 10.1016/j.ejsobi.2024.103644 . hal-04623177

HAL Id: hal-04623177

<https://hal.science/hal-04623177v1>

Submitted on 25 Jun 2024

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution 4.0 International License



Original article

Wildflower strips of 2.5-year-old promote earthworms and enchytraeids (Annelida, Oligochaeta) in arable fields

C. Pelosi^{a,*}, M. Bertrand^b, A. Gardarin^b^a INRAE, Avignon Université, UMR EMMAH, 84000, Avignon, France^b Université-Paris Saclay, INRAE, AgroParisTech, UMR Agronomie, 91120, Palaiseau, France

ARTICLE INFO

Handling Editor: Guénola Pérès

Keywords:

Agriculture
Agroecology
Terrestrial invertebrates
Cropping system
Perennial flower strips
Biodiversity

ABSTRACT

In the last 50 years, humans have increased crop yields due to intensive agricultural practices and by homogenizing cultivated lands (e.g., larger and more uniform fields). However, this land management practice has led to serious environmental issues, and now, the importance of heterogeneity and semi-natural landscape elements in production systems is acknowledged. Perennial habitats, such as flower strips, could play a key role in agroecosystem sustainability, but little is known about their effects on earthworm and enchytraeid (Annelida: Oligochaeta) communities. The aim of this study was to assess earthworms and enchytraeids in 2.5-year-old wildflower strips that were sown in the middle of arable fields in northern France. Samples (soil, earthworms and enchytraeids) were collected at ten locations, in flower strips and in adjacent cropped fields. The same number of earthworm species was found in both habitats, but more enchytraeid species were detected in the flower strips than in the adjacent cropped fields. Moreover, the total abundance of earthworms and enchytraeids significantly increased in the flower strips compared with the adjacent cropped fields, by 69 % and 61 %, respectively. Flower strips had a significant positive effect on anecic and endogeic earthworms but not on the abundance of epigeic earthworms, which was highly variable among the samples, although on average, it was seven times greater in the flower strips than in the cropped fields. Although the flower strips were sown only 2.5 years earlier, significant changes were observed in the soil Oligochaeta communities. These findings advocate for sowing flower strips within cultivated land as a source of soil biodiversity in the current changing environment. Considering the positive role of flower strips on biodiversity and particularly on the studied tiny soil engineers, these perennial landscape elements should be more widely considered to support the agroecological transition.

1. Introduction

Through the use of pesticides, tillage and landscape simplification, agriculture strongly affects biodiversity [1], which is currently massively declining [2,3]. Pesticide-free or no-till cropping systems have beneficial effects on biodiversity, including soil fauna [4,5]. Moreover, ecological features in or around fields, such as hedges and flower strips, can increase the diversity of aboveground and belowground organisms [6–8]. As these perennial habitats are weakly disturbed or not disturbed, they constitute refuges for wildlife within agricultural landscapes, mainly in intensive agricultural contexts [9]. For many years, removing these ecological features has been common agricultural practice, but recent reports underline the key role they play in agroecosystem sustainability and agroecological transition [3].

Perennial habitats, such as flower strips and beetle banks, protect

populations of epigeic arthropods and soil organisms against disturbances in the vicinity of cropped habitats [10,11]. Dense vegetation with complex architecture as well as accumulated plant litter offer more stable microclimate conditions throughout the year than does the cropped field surface [12]. The permanent plant cover and diverse vegetation of perennial habitats provide resources for the soil decomposer community through root exudates and plant litter deposition [13, 14]. Therefore, perennial habitats, such as flower strips, promote soil organisms (e.g., earthworms and enchytraeids) that are also a trophic resource for many invertebrates, birds and mammals and consequently enhance biodiversity at higher trophic levels.

However, the effects of planting flower strips on soil organisms are poorly known. Terrestrial Oligochaeta communities are modified (e.g., in terms of abundance, species diversity, and vertical distribution) by soil tillage and by the use of pesticides and organic matter inputs [15,

* Corresponding author. 228 route de l'Aérodrome, CS 40 509, 84 914 AVIGNON Cedex 9, France.

E-mail address: celine.pelosi@inrae.fr (C. Pelosi).

<https://doi.org/10.1016/j.ejsobi.2024.103644>

Received 26 May 2023; Received in revised form 6 June 2024; Accepted 20 June 2024

Available online 24 June 2024

1164-5563/© 2024 The Authors. Published by Elsevier Masson SAS. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

16]. Therefore, earthworms and enchytraeids are used as indicators of land use and management [17,18]. However, there are very few studies on the effects of flower strip implementation within arable fields on these invertebrates. Kohli et al. [19] reported that the earthworm total abundance, biomass and species richness increased in a rotational maize field turned into wildflower strips. This indicates that strips with wildflowers are suitable habitats for earthworms. Some species, such as *Lumbricus terrestris*, a common epi-aneic earthworm, were more favoured than others, potentially due to their sensitivity to soil tillage and their epigeic behaviour [20]. In long-term set-aside plots sown with a meadow seed mixture, earthworm density, biomass and richness were favoured by the absence of mowing [21]. In contrast, nothing is known about the effects of flower strips on enchytraeids, which might be less sensitive to environmental factors (e.g., pesticides, soil tillage, salinity) than earthworms [22–24]. However, several studies have suggested that enchytraeids are even more important than earthworms in conventionally cultivated arable land because they are less sensitive to ploughing (they can even be favoured by minimum tillage [25]) and are more active metabolically [26,27].

The aim of this study was to assess earthworm and enchytraeid (Annelida: Oligochaeta) communities in 2.5-year-old wildflower strips in arable cropped fields. To this end, these invertebrates were sampled in both habitats (flower strips and cropped fields) at ten locations near Paris, France. Our initial hypotheses were that i) the abundance and diversity of earthworms and enchytraeids are greater in wildflower strips than in arable cropped fields and that ii) the beneficial effect of flower strips is less important for enchytraeid than earthworm communities because the former are less sensitive to soil tillage.

2. Materials and methods

2.1. Sampling area and design

The study was carried out in the northern part of France, a region dominated by arable farmland (mostly cereals and oilseed rape) in open field landscapes. In fall 2018, at ten farms managed using organic, conservation or conventional farming and with different cropping systems (Table 1), a perennial flower strip was sown with a mixture of 42 native and mostly perennial species across a field to divide it into two parts. Each flower strip measured approximately 4–6 m wide and 200–800 m long. Strips were mown once per year in winter. The strips did not receive any chemical treatment or any intentional fertilisation, although some drift could have occurred despite the absence of

observation. In spring 2021, winter cereals, oilseed rape, or lucerne were grown in the selected cropped fields (Table 1). Earthworms and enchytraeids were sampled in flower strips and in adjacent cropped fields 50 m from the strip (Fig. 1). All the sample points were located approximately 30 m from the grassy roadside.

2.2. Soil, earthworm and enchytraeid sampling

In spring 2020, for soil analysis (Table S1), three soil samples (0–20 cm depth) were taken from each field and from each flower strip (Fig. 1) using a 5-cm Ø soil auger. Then, they were combined to obtain one composite sample per site. Soil characteristics were measured at the Laboratoire d'Analyse des Sols, Institut national de recherche pour l'agriculture, l'alimentation et l'environnement (Arras, France). This laboratory's quality is certified by COFRAC (French accreditation committee) for soil characteristic analysis. Briefly, soils were dried at room temperature, disaggregated, homogenised and then sieved (2-mm-sieve). The following soil characteristics were measured: pH (by water suspension), organic matter and nitrogen contents (by dry combustion, in g kg^{-1}), grain size distribution (clay <2 μm , silt 2–20 μm , and sand >20 μm), total calcium carbonate (CaCO_3 ; in g kg^{-1}), and total

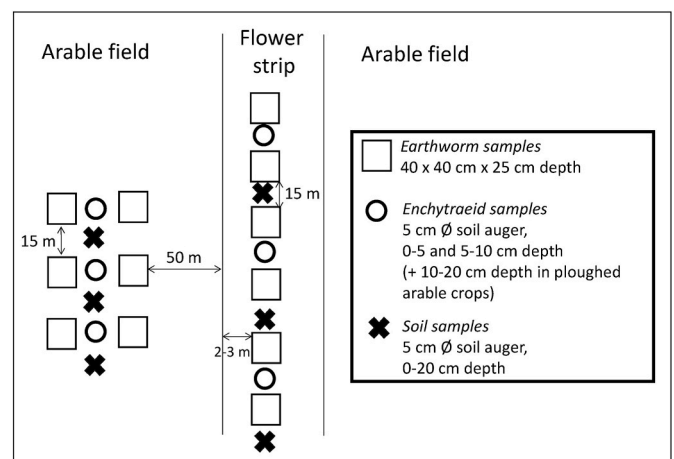


Fig. 1. Sampling at each of the ten studied fields. In each field, earthworms, enchytraeids and soil were sampled in the flower strip and in the adjacent cropped field.

Table 1

Description of the cropping systems implemented at the ten fields where earthworms and enchytraeids were sampled. TFI, Treatment Frequency Index is used for the assessment of pesticide pressure at different scales, from field to national level; it is defined as the mean number of treatments per hectare with commercial products, weighted by the ratio of the dose used to the recommended dose. As a reference, the average TFI for wheat in France was 4.9 in 2017 [28]. In France, organic agriculture follows the European standard n° 834/2007 (the use of synthetic pesticides and fertilizers is forbidden). Conservation agriculture is based on: i) the absence of soil disturbance other than sowing with a light tine or disc, ii) maximum soil coverage by intercropping and residues, and iii) diversification of cultivated species over time (rotation) and in space (intercrops). In conventional farming, tillage and synthetic inputs are used. The cropping fields were classified as conventional, conservation or organic in 2021, but some have recently transitioned to another system, thus explaining the TFI of 1.7 in a field classified as organic.

Field number	Agriculture system	Agriculture system history	Crop in 2021	Mean annual TFI (2018–2021)	Total number of moldboard ploughing operations (2018–2021)	Mean number of other tillage operations/year (2018–2021)
1	Conservation	10 years	Winter wheat	6.9	0.0	0.3
2	Conservation	10 years	Winter barley	3.4	1.0 ^a	0.3
3	Conservation	5 years	Oilseed rape	6.8	0.0	0.0
4	Conventional	>30 years	Oilseed rape	5.0	1.0	2.3
5	Conventional	>30 years	Winter wheat	5.9	2.0	1.3
6	Conventional	>30 years	Winter barley	6.2	2.0	2.5
7	Organic	2 years, after conventional	Lucerne	0.0	2.0	4.7
8	Organic	2 years, after conservation	Triticale undersown to white clover	1.7	0.0	4.0
9	Organic	20 years	Winter wheat	0.0	4.0	5.5
10	Organic	20 years	Winter wheat	0.0	4.0	5.7

^a Exceptional intervention.

phosphorus (P_2O_5 ; in $g\ kg^{-1}$) by inductively coupled plasma mass spectrometry. The soil texture at the ten sampling locations was mainly silty loam composed of 61 % silt, 24 % clay and 15 % sand (Table S1). The pH at all sampling sites ranged between 7.2 and 8.2. Similar to all the measured soil parameters (Table S1), the mean (\pm standard deviation) organic matter content was similar in the flower strips ($25.3 \pm 7.2\ g\ kg^{-1}$) and in the cropped fields (25.6 ± 7.3).

Soil moisture was measured by weighing soil samples (0–20 cm depth, taken from the composite sample used for soil analysis, see above) before and after drying at $105\ ^\circ C$ for 48 h. The mean soil moisture ($n = 10$) was 29.8 % (± 4.6) in the flower strips and 29.6 % (± 2.7) in the cropped fields.

Earthworms and enchytraeids were sampled in March 2021. At a depth of 10 cm, the minimum and maximum soil temperatures during the sampling period (5 days) were 6.5 and $8.0\ ^\circ C$, respectively (at the meteorological station of Thiverval Grignon, 50 km west of the sampling area). For the earthworms, six samples were taken from each cropped field and flower strip (Fig. 1) at the ten sampling locations. A $40\ cm \times 40\ cm \times 25\ cm$ -deep block of soil was extracted. Earthworms were hand-sorted and preserved in 70 % alcohol before counting and weighing each individual without emptying the gut contents. The development stage of each earthworm was noted. Earthworms were considered sub-adults if they had full tubercula pubertatis but no clitellum and adults if they had a clitellum. They were considered juvenile if they had neither a tubercula pubertatis nor a clitellum. Sub-adults and adults were identified at the species level using the identification keys described by Sims and Gerard [29]. Juveniles were identified at the species level on the basis of the morphological characteristics of each species and the specific form they take in alcohol compared with adults. When identification at the species level was impossible, individuals were classified as 'unidentified'.

For the enchytraeids, three samples were taken from each cropped field and from each flower strip (Fig. 1) at the ten sampling locations. Sampling was carried out following the ISO 23611-3 method [30] with a 5-cm \varnothing soil auger at 0–5 cm and 5–10 cm depths in the flower strips and the non-ploughed fields (conservation agriculture, Table 1) because most enchytraeids generally stay in the uppermost soil layers [31]. In the ploughed fields (Table 1), samples were taken at 0–5, 5–10, and 10–20 cm depths because enchytraeids can be redistributed at different depths by soil tillage [32,33]. Each sample was transferred separately into a plastic bag and stored at $4\ ^\circ C$. Enchytraeids were extracted using wet funnel extractors under light from incandescent light bulbs [34,35]. All individuals were kept in Petri dishes with tap water and counted. Adult and sub-adult individuals were identified at the species level under a light microscope (up to 400x magnification) using the keys described by Schmelz and Collado [36].

2.3. Statistical analysis

At each site and for both flower strips and cropped fields, earthworm abundance and biomass were determined for the whole sample and for the three main ecological categories: epigeic, anecic and endogeic. For enchytraeids, the total abundance and abundance of r-strategists (i.e., life strategy, according to Graefe and Schmelz [37]) were determined. Abundance and biomass were expressed as individuals m^{-2} and $g\ m^{-2}$, respectively. The effects of habitat (cropped field vs. flower strip) on earthworm and enchytraeid abundance, biomass and richness were assessed. Generalized linear mixed models and the glmmTMB package [38] were used, assuming a Gaussian error distribution or a negative binomial error distribution to account for data overdispersion, depending on the response variable. The models included replicate numbers nested within the habitat as a random effect to account for repeated measurements. The significance of the fixed effects (habitat and cropping system) was evaluated with type II analyses of deviance with the Wald chi-squared test and the ANOVA function of the car package [39]. Interactions between explanatory variables were not

tested due to the insufficient size of the dataset. The distribution of model residuals (normality, homoscedasticity, outliers) was checked and confirmed with the DHARMA package [40]. To visualise how the cropping systems may have influenced the difference between flower strips and cropped fields, ratios of the total abundance and species richness in flower strip to cropped field were calculated for both earthworms and enchytraeids in all the fields, according to the cropping systems. Statistical analyses were performed with R software version 3.1.3 [41].

3. Results

3.1. Earthworms

The total earthworm abundance and total biomass were significantly greater ($1.81 \cdot 10^{-7}$ and $5.73 \cdot 10^{-8}$, respectively) in the flower strips than in the adjacent cropped fields (238.5 ± 143.8 individuals m^{-2} versus 141.0 ± 74.8 individuals m^{-2} ; and $93.6\ g\ m^{-2} \pm 49.2$ versus $54.5\ g\ m^{-2} \pm 33.2$, respectively) (Fig. 2a–b). The positive effect of flower strips on abundance and biomass was also significant when earthworms were divided into ecological categories, except for the mean abundance of epigeic earthworms ($p = 0.06$), although it was seven times greater in the flower strips than in the cropped fields (Table 2).

Overall, two epigeic species, three anecic or epi-anecic species (considered anecic), and six endogeic or epi-endo-anecic species (considered endogeic), were found at the ten sampling locations (Table 2). The most abundant species was *Aporrectodea caliginosa*, which represented 55 % of the total abundance in the flower strips and 50 % in the cropped fields across the ten sampling locations (Table 2). Two

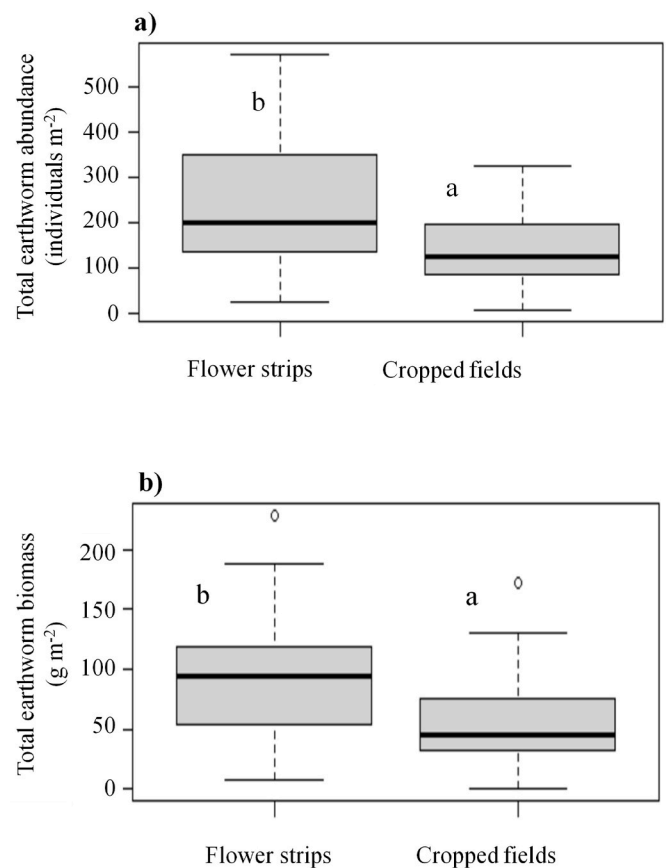


Fig. 2. Total earthworm a) abundance (individuals m^{-2}) and b) biomass ($g\ m^{-2}$) in flower strips and cropped fields ($n = 60$ observations in 10 fields for both). Significant differences between habitats ($p = 0.05$) are indicated with different letters.

Table 2

Effect of the habitat and of the cropping system on the abundance (individuals m^{-2}) and biomass ($g\ m^{-2}$) (standard-deviations in brackets) of the different species and on ecological categories of earthworms. The effect of habitat (FS: flower strips, CF: cropped fields) and cropping systems (conventional, conservation, organic farming) were tested with a generalized mixed effect model ($n = 120$ observations in ten fields); significant results are in bold.

Species	Ecological category(ies)	Abundance (ind·m ⁻²)		Effect of habitat p-value	Effect of cropping system	Biomass (g·m ⁻²)		Effect of habitat p-value	Effect of cropping system
		FS	CF			FS	CF		
<i>Lumbricus rubellus</i>	epigeic	0.3 (0.7)	0.0 (0.0)			0.2 (0.5)	0.0 (0.0)		
<i>Lumbricus castaneus</i>	epigeic	14.8 (43.9)	1.8 (4.3)			2.5 (7.4)	0.2 (0.6)		
Epigeic indet.	epigeic	0.0 (0.0)	0.2 (0.7)			0.0 (0.0)	0.0 (0.1)		
Total epigeic earthworms		15.1 (43.8)	2.0 (4.3)	0.0577	0.9705	2.7 (7.4)	0.3 (0.6)	0.0004	0.5815
<i>Lumbricus terrestris</i>	epi-aneic	7.7 (12.8)	4.7 (5.6)			3.8 (4.3)	4.9 (5.8)		
<i>Aporrectodea longa</i>	aneic	11.3 (16.2)	5.3 (10.2)			18.6 (24.5)	6.8 (7.9)		
<i>Aporrectodea giardi</i>	aneic	1.6 (2.7)	0.7 (1.1)			5.5 (8.6)	2.6 (4.1)		
<i>A. longa/A. giardi</i> (indet.)	aneic	17.4 (19.8)	15.5 (22.5)			8.8 (8.4)	8.6 (12.8)		
Total aneic earthworms		37.9 (39.5)	26.3 (26.1)	0.0153	0.8447	36.7 (37.4)	22.8 (18.5)	0.0191	0.8412
<i>Allolobophora chlorotica</i>	epi-endo-aneic	13.1 (19.7)	11.7 (18.4)			2.8 (4.3)	2.6 (4.2)		
<i>Aporrectodea caliginosa</i>	endogeic	131.8 (101.3)	71.7 (51.3)			37.5 (23.5)	18.7 (13.6)		
<i>Aporrectodea rosea</i>	endogeic	6.5 (9.1)	2.6 (2.2)			1.4 (1.8)	0.8 (0.7)		
<i>Aporrectodea icterica</i>	endogeic	33.3 (28.6)	25.7 (26.2)			11.9 (9.6)	9.1 (9.1)		
<i>Allolobophora muldali</i>	endogeic	0.1 (0.3)	0.0 (0.0)			0.0 (0.0)	0.0 (0.0)		
<i>Octolasion cyaneum</i>	epi-endo-aneic	0.2 (0.7)	0.1 (0.3)			0.5 (1.7)	0.2 (0.5)		
Endogeic indet.	endogeic	0.5 (1.3)	1.0 (1.4)			0.0 (0.1)	0.1 (0.1)		
Total endogeic earthworms		185.5 (113.6)	112.8 (51.0)	6.51·10⁻⁷	0.2277	54.2 (26.9)	31.5 (17.7)	3.83·10⁻⁷	0.2117

species (*Allolobophora muldali* and *Lumbricus rubellus*) were present only in the flower strips (Table 2). For *Lumbricus castaneus*, the standard deviation was very high because its abundance was very high in one flower strip (i.e., 139.6 individuals m^{-2} , field 7; Table 1 and Table A1) and very low (≤ 5 individuals m^{-2}) in the other nine flower strips. The mean number of earthworm species was 3.7 ± 1.0 in the flower strips

and 3.4 ± 1.1 in the cropped fields (p value = 0.23).

The ratios of the total abundance in flower strip to cropped field was higher in one organic field than in the other systems for earthworms (Fig. 3a), highlighting potential differences in the effect of flower strips depending on the agricultural system, although cropping systems effects were never significant (Table 2; Table A2).

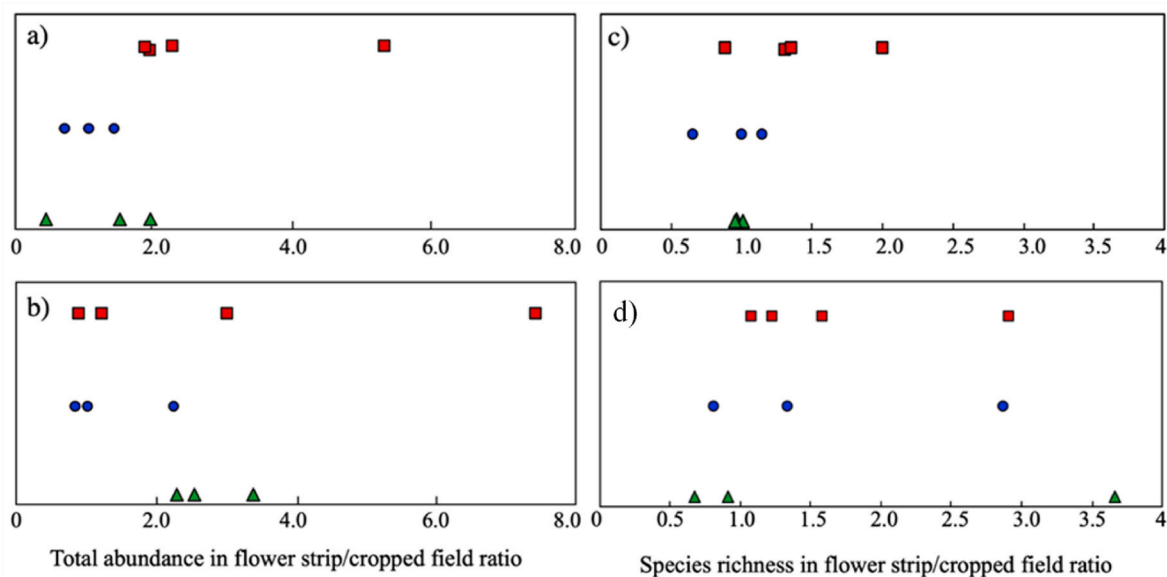


Fig. 3. Stripchart plots illustrating the ratio of the total abundance (a, b) or of species richness (c, d) in flower strip to cropped field for earthworms (a, c) and enchytraeids (b, d). Green triangles, blue circles and red squares for conventional ($n = 3$), conservation ($n = 3$) and organic agriculture ($n = 4$), respectively.

3.2. Enchytraeids

In total, 1475 and 913 individuals were found in wildflower strips and cropped fields, respectively. The total enchytraeid abundance and species richness were greater in the flower strips than in the cropped fields ($25,041 \pm 18,684$ individuals m^{-2} versus $15,466 \pm 16,186$ individuals m^{-2} , $p = 0.00228$; and 6.9 ± 2.5 species versus 5.1 ± 3.2 species, $p = 0.00536$, respectively) (Fig. 4a–b). Cropping system had no significant effect.

Seven different genera and 34 species of enchytraeids were recorded, as well as nine potentially new, undescribed species (1 from *Buchholzia*, 2 from *Achaeta* and 6 from *Fridericia*) (Table 3). The most abundant species were *Enchytraeus bulbosus* and *Enchytraeus buchholzi*, which represented 25 % and 20 % of the total enchytraeid abundance, respectively, followed by *Fridericia christeri* (7 % of the total abundance). These three species were found at all ten sampled sites. Three species were found at seven sites, but each represented less than 6 % of the total abundance (Table 3). The percentage of r-strategists was slightly lower ($p = 0.04$) in flower strips ($40 \% \pm 24$) than in cropped fields ($50 \% \pm 28$) (Table 3), without any cropping system effect ($p = 5.38 \cdot 10^{-9}$).

The calculated abundance ratios for enchytraeids were similar regardless of the farming system (conservation, conventional, or organic farming) (Fig. 3b). The dispersion was also greater between fields managed by organic agriculture than between those managed by conservation or conventional agriculture. The mean ratios of the species richness of enchytraeids and earthworms in flower strips to that in cropped fields were also very dispersed between fields (Fig. 3c–d) and were not significantly different. The mean ratio ($n = 10$) of the total abundance in flower strips to that in cropped fields was not different ($p = 0.46$) for enchytraeids (2.47 ± 1.99) and earthworms (1.88 ± 1.34).

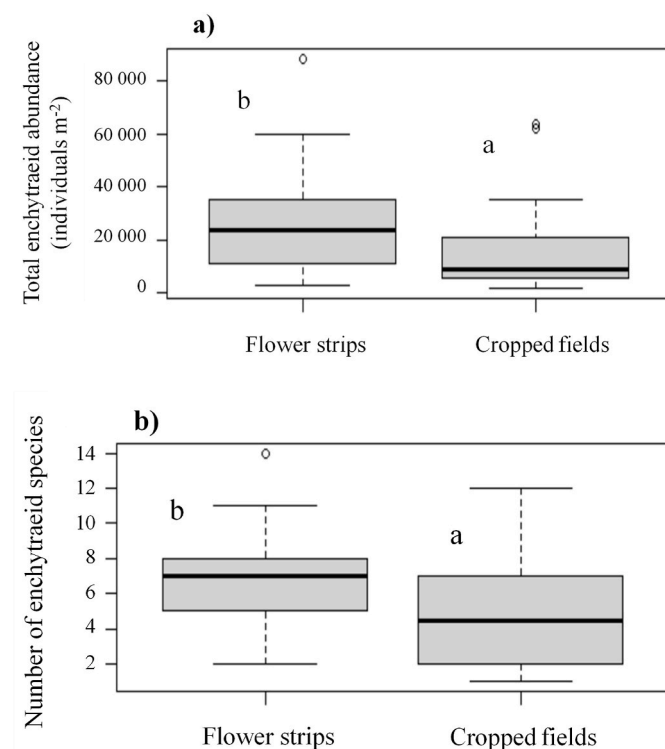


Fig. 4. a) Mean total abundance (individuals m^{-2}) and b) diversity (number of species) of enchytraeids in flower strips and cropped fields ($n = 30$ observations in 10 fields for both). Significant differences between habitats ($p = 0.05$) are indicated with different letters.

4. Discussion

At 2.5 years after sowing, flower strips increased the total abundance of earthworms and enchytraeids, favouring anecic and endogeic earthworms and also enchytraeid diversity compared with the adjacent arable cropped fields. This might be due to the decrease in physical and chemical disturbances and the suitable feeding conditions for Oligochaeta (Annelida) in the perennial flower strips [12,21]. These positive effects of flower strips were also found for above-ground biodiversity, with perennial and older flower strips being more effective than younger flower strips [42]. The increase in earthworm and enchytraeid abundance in flower strips is explained first by internal recovery, which depends on the individuals present in the original cropped field and originates from population increase through reproduction after the soil management changes [43]. External recovery may also have occurred through the immigration of individuals from neighbouring areas (e.g., adjacent grassy roadsides) by active or passive dispersal [44–46]. External recovery may explain why some species were found only in flower strips (e.g., *L. rubellus*) where individuals could settle due to suitable local conditions. As different earthworm species do not have the same dispersal and colonization capabilities [46], diversity can increase slowly in flower strips. Moreover, the seed mixtures (i.e., plant community species composition) used in the different studied flower strips can influence plant community development and consequently earthworm communities [21]. Therefore, it is important to assess the long-term effects of flower strips. Future studies could determine whether flower strips allow field colonization. This could be done by assessing whether a gradient of abundance appears after a few years between the flower strip and the cropped fields, as observed by Hof and Bright [47]. More generally, the dynamics of earthworm and enchytraeid populations should be regularly assessed in these less disturbed habitats to measure the speed (i.e., slope of the curve) of the increase in abundance and diversity and the change in carrying capacity (i.e., plateau of the curve that represents the maximum population size that can be sustained by the environment) of these preserved habitats.

In our cropped fields, the abundances of earthworms and enchytraeids were close to those generally found in agricultural systems: approximately 140 and 15,000 individuals m^{-2} , respectively [48,49]. Moreover, the most abundant earthworm (*A. caliginosa*) and enchytraeid (*E. buchholzi* and *E. bulbosus*) species are species typically found in cropped fields in temperate regions [23,50]. For earthworms, the combination of hand-sorting and a chemical expellant (i.e., ethological methods) is recommended, particularly to properly assess anecic earthworms [51]. However, Callaham and Hendrix [52] reported that the formalin extraction method was more effective than hand-sorting for the collection of *L. terrestris* but not for any other species. Moreover, liquid extraction involves a chemical (e.g., formalin or mustard oil) that is not necessarily allowed in organic farming. Here, we chose to use hand-sorting only, so the abundance of adult anecic and epi-anecic earthworms may have been underestimated and greater differences between flower strips and arable cropped fields potentially masked. This choice could also have underestimated their abundances under conservation agriculture with lower tillage intensity. Nine potentially new species of enchytraeids (not described so far) were found in the ten sampling sites near Paris. Enchytraeids have been less studied than earthworms, although they have functional roles similar to those of earthworms in soils (i.e., soil organic matter degradation and soil porosity dynamics) but at a smaller scale [53]. Therefore, more studies are needed to better describe the diversity of these soil animals and to assess the effects of anthropogenic activities on enchytraeids.

The studied cropped fields were managed differently, with either conventional, organic or conservation agriculture techniques. In all three agricultural systems, the abundance and biomass of all earthworm species and two-thirds of the enchytraeid species were greater in the flower strips than in the adjacent cropped fields. This positive effect of

Table 3

Number of individuals of the different enchytraeid species found in the 10 sampling sites at two (in flower strips and unploughed crops) or three (in ploughed crops) different soil depths in flower strips and cropped fields, and life strategy of the identified species (r or k, according to Graefe and Schmelz, 1999). *Fridericia* sp. indet. are juveniles that could not be identified at the species level. *Fridericia* sp. 1 to 6 are potentially new species (undescribed to date) that could not be named (but the individuals could be linked to a morphospecies).

			Strategy	Number of sites	Flower strips			Cropped fields			TOTAL
					0–5 cm	5–10 cm	TOTAL	0–5 cm	5–10 cm	10–20 cm	
<i>Achaeta</i>	<i>bohemica</i>	Vejdovský, 1879	k	1	5	1	6	0	0	0	0
<i>Achaeta</i>	<i>pannonica</i>	Graefe, 1989	k	1	1	1	2	0	0	0	0
<i>Achaeta</i>	sp. 1		–	3	0	1	1	0	0	3	3
<i>Achaeta</i>	sp. 2		–	1	0	0	0	0	1	1	2
<i>Buchholzia</i>	<i>appendiculata</i>	Buchholz, 1862	r	5	24	5	29	1	0	0	1
<i>Buchholzia</i>	<i>fallax</i>	Michaelsen, 1887	r	3	6	3	9	0	1	1	2
<i>Buchholzia</i>	sp. 1		–	1	0	0	0	0	1	3	4
<i>Enchytraeus</i>	<i>buchholzi</i>	Vejdovský, 1879	r	10	191	58	249	131	63	30	224
<i>Enchytraeus</i>	<i>bulbosus</i>	Nielsen & Christensen, 1959	r	10	177	184	361	115	97	22	234
<i>Enchytraeus</i>	<i>coronatus</i>	Nielsen & Christensen, 1959	r	1	1	0	1	0	1	0	1
<i>Enchytraeus</i>	<i>lacteus</i>	Nielsen & Christensen, 1959	r	1	0	4	4	0	1	0	1
<i>Enchytronia</i>	<i>christenseni</i>	Dózsa-Farkas, 1970	k	7	26	50	76	6	25	20	51
<i>Fridericia</i>	<i>bulboides</i>	Nielsen & Christensen, 1959	k	2	20	0	20	0	0	0	0
<i>Fridericia</i>	<i>christeri</i>	Rota & Healy, 1999	k	10	94	42	136	14	24	3	41
<i>Fridericia</i>	<i>connatifformis</i>	Dózsa-Farkas, 2015	k	2	10	2	12	5	5	0	10
<i>Fridericia</i>	<i>dozsa</i>	Schmelz, 2003	k	3	2	3	5	2	0	0	2
<i>Fridericia</i>	<i>galba</i>	Hoffmeister, 1843	k	7	18	7	25	10	4	6	20
<i>Fridericia</i>	<i>isseli</i>	Rota, 1994	k	5	16	8	24	1	10	11	22
<i>Fridericia</i>	<i>nix</i>	Rota, 1995	k	5	1	16	17	1	0	10	11
<i>Fridericia</i>	<i>paroniana</i>	Issel, 1904	k	2	7	0	7	5	3	0	8
<i>Fridericia</i>	<i>rendsinata</i>	Dózsa-Farkas, 1972	k	1	6	0	6	1	0	0	1
<i>Fridericia</i>	<i>sylvatica</i>	Healy, 1979	k	3	1	2	3	12	16	0	28
<i>Fridericia</i>	<i>tuberosa</i>	Rota, 1995	k	3	5	2	7	4	4	0	8
<i>Fridericia</i>	sp. 1		–	7	0	11	11	1	1	3	5
<i>Fridericia</i>	sp. 2		–	4	1	2	3	0	5	4	9
<i>Fridericia</i>	sp. 3		–	1	0	0	0	0	1	0	1
<i>Fridericia</i>	sp. 4		–	1	2	0	2	0	0	0	0
<i>Fridericia</i>	sp. 5		–	1	8	9	17	0	1	0	1
<i>Fridericia</i>	sp. 6		–	1	1	0	1	0	0	4	4
<i>Fridericia</i>	sp. indet.		–	10	186	119	305	80	78	26	184
<i>Henlea</i>	<i>perpusilla</i>	Friend, 1911	k	4	7	8	15	10	6	0	16
<i>Henlea</i>	<i>ventriculosa</i>	d'Udekem, 1854	k	4	65	1	66	2	0	2	4
<i>Marionina</i>	<i>brendae</i>	Rota, 1995	k	1	0	3	3	0	0	6	6
<i>Marionina</i>	<i>hoffbaueri</i>	Möller, 1971	k	1	0	1	1	0	0	0	0
<i>Marionina</i>	<i>mendax</i>	Rota, 2013	k	2	19	32	51	1	8	0	9
TOTAL (Σ of individuals)					900	575	1475	402	356	155	913

flower strips on earthworm abundance could be related to the agricultural system, but here the low number of fields per system makes it difficult to conclude properly on the effects of these three systems.

Our findings showed that the enchytraeid and earthworm communities were similarly favoured by flower strip sowing. Beylich et al. [49] stated that conditions favouring soil organisms can result in increased activity of these two Oligochaeta groups. Similar to earthworms, enchytraeids are bioindicators of land use and agricultural practices [15, 18], and they can take advantage of the decrease or suppression in soil tillage and pesticide use [54]. As enchytraeids live at the soil surface and consume high quantities of organic matter [33], they could have been more favoured by the supply of organic matter in flower strips (decomposing senescent plants, especially in fall). Last, as enchytraeids have a relatively short life cycle compared with that of earthworms, their populations can increase rapidly when the available resources increase.

5. Conclusion

Perennial habitats, such as flower strips, promote aerial and soil organisms, but few studies have investigated their effects on Oligochaeta. The present study showed that in a short period of time (2.5 years), flower strips could increase the abundance and biomass of earthworms and enchytraeids and also increase enchytraeid diversity. As these soil organisms are involved in key functions related to soil

fertility and are prey for higher trophic levels, the implementation of such linear habitats in the landscape could constitute a source of biodiversity and provide other ecosystem services in agricultural landscapes.

CRedit authorship contribution statement

C. Pelosi: Writing – review & editing, Writing – original draft, Supervision, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **M. Bertrand:** Writing – review & editing, Methodology, Investigation. **A. Gardarin:** Writing – review & editing, Resources, Project administration, Funding acquisition, Formal analysis, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Acknowledgments

This work was funded by INRAE (metaprogram EcoServ) and the Office Français de la Biodiversité. We would like to thank R. Schmelz for the identification of enchytraeid individuals. We thank A. Butier for its help at sampling as well as S. Reno and Y. Capowiez for their help at sampling and during the identification session.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.ejsobi.2024.103644>.

References

- [1] S. Adhikari, A. Adhikari, D.K. Weaver, A. Bekkerman, F.D. Menalled, Impacts of agricultural management systems on biodiversity and ecosystem services in highly simplified dryland landscapes, *Sustainability* 11 (2019) 3223.
- [2] F. Geiger, J. Bengtsson, F. Berendse, W.W. Weisser, M. Emmerson, M.B. Morales, P. Ceryngier, J. Liira, T. Tschamtkke, C. Winqvist, S. Eggers, R. Bommarco, T. Part, V. Bretagnolle, M. Plantegenest, L.W. Clement, C. Dennis, C. Palmer, J.J. Onate, I. Guerrero, V. Hawro, T. Aavik, C. Thies, A. Flohre, S. Hanke Fischer, P. W. Goedhart, P. Inchausti, Persistent negative effects of pesticides on biodiversity and biological control potential on European farmland, *Basic Appl. Ecol.* 11 (2010) 97–105.
- [3] S. Díaz, J. Settele, E.S. Brondízio, H.T. Ngo, M. Guèze, J. Agard, A. Arnett, P. Balvanera, K.A. Brauman, S.H.M. Butchart, K.M.A. Chan, L.A. Garibaldi, K. Ichii, J. Liu, S.M. Subramanian, G.F. Midgley, P. Miloslavich, Z. Molnár, D. Obura, A. Pfaff, S. Polasky, A. Purvis, J. Razzaque, B. Reyers, R. Roy Chowdhury, Y.J. Shin, L.J. Visseren-Hamakers, K.J. Willis, C.N. Zayas (Eds.), *IPBES, Summary for Policymakers of the Global Assessment Report on Biodiversity and Ecosystem Services of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services*, IPBES secretariat, Bonn, Germany, 2019, p. 56.
- [4] L. Henneron, M. Aubert, F. Bureau, Y. Dumas, F. Ningre, S. Perret, C. Richter, P. Balandier, M. Chauvat, Forest management adaptation to climate change: a Cornelian dilemma between drought resistance and soil macro-detritivore functional diversity, *J. Appl. Ecol.* 52 (2015) 913–927.
- [5] S.F. Bender, C. Wagg, M.G.A. van der Heijden, An underground revolution: biodiversity and soil ecological engineering for agricultural sustainability, *Trends Ecol. Evol.* 31 (2016) 440–452.
- [6] N. Boatman (Ed.), *Field Margins: Integrating Agriculture and Conservation*, BCPC Monograph, 58, British Crop Protection Council, Farnham, Surrey, 1994.
- [7] E.B. Lowe, R. Groves, C. Gratton, Impacts of field-edge flower plantings on pollinator conservation and ecosystem service delivery - a meta-analysis, *Agric. Ecosyst. Environ.* (2021) 310.
- [8] C. Haaland, R.E. Naisbit, L.F. Bersier, Sown wildflower strips for insect conservation: a review, *Insect Divers. Conserv.* 4 (2011) 60–80.
- [9] R. Marja, D. Kleijn, T. Tschamtkke, A.-M. Klein, T. Frank, P. Batary, Effectiveness of agri-environmental management on pollinators is moderated more by ecological contrast than by landscape structure or land-use intensity, *Ecol. Lett.* 22 (2019) 1493–1500.
- [10] A. MacLeod, S.D. Wratten, N.W. Sotherton, M.B. Thomas, 'Beetle banks' as refuges for beneficial arthropods in farmland: long-term changes in predator communities and habitat, *Agric. For. Entomol.* 6 (2004) 147–154.
- [11] M. Porcel, B. Cotes, J. Castro, M. Campos, The effect of resident vegetation cover on abundance and diversity of green lacewings (Neuroptera: Chrysopidae) on olive trees, *J. Pest. Sci.* 90 (2017) 195–206.
- [12] G.J.K. Griffiths, J.M. Holland, A. Bailey, M.B. Thomas, Efficacy and economics of shelter habitats for conservation biological control, *Biol. Control* 45 (2008) 200–209.
- [13] D.A. Wardle, R.D. Bardgett, J.N. Klironomos, H. Setälä, W.H. van der Putten, D. H. Wall, Ecological linkages between aboveground and belowground biota, *Science* 304 (2004) 1629–1633.
- [14] S. Fujii, M. Berg, J. Cornelissen, Living litter: dynamic trait spectra predict fauna composition, *Trends Ecol. Evol.* 35 (2020).
- [15] C. Pelosi, J. Römcke, Are *Enchytraeidae* (*Oligochaeta*, *Annelida*) good indicators of agricultural management practices? *Soil Biol. Biochem.* 100 (2016) 255–263.
- [16] A.S.A. Johnston, R.M. Sibly, M.E. Hodson, T. Alvarez, P. Thorbek, Effects of agricultural management practices on earthworm populations and crop yield: validation and application of a mechanistic modelling approach, *J. Appl. Ecol.* 52 (2015) 1334–1342.
- [17] T. Liu, X. Chen, X. Gong, I.M. Lubbers, Y. Jiang, W. Feng, X. Li, J.K. Whalen, M. Bonkowski, B.S. Griffiths, F. Hu, M. Liu, Earthworms coordinate soil biota to improve multiple ecosystem functions, *Curr. Biol.* 29 (2019) 3420–3429.
- [18] C. Pelosi, J. Römcke, Enchytraeids as bioindicators of land use and management, *Appl. Soil Ecol.* 123 (2018) 775–779.
- [19] L. Kohli, O. Daniel, F. Schönholzer, D. Hahn, J. Zeyer, *Miscanthus sinensis* and wild flowers as food resources of *Lumbricus terrestris* L., *Appl. Soil Ecol.* 11 (1999) 189–197.
- [20] M.B. Bouché, *Lombriciens de France: Ecologie et Systématique*, INRA Ann. Zool. Ecol. Anim. Publication, 1972, France.
- [21] T. Hyvönen, E. Huusela, M. Kuussaari, M. Niemi, R. Uusitalo, V. Nuutinen, Aboveground and belowground biodiversity responses to seed mixtures and mowing in a long-term set-aside experiment, *Agric. Ecosyst. Environ.* 322 (2021) 107656.
- [22] S. Bart, C. Laurent, A.R.R. Péry, C. Mougin, C. Pelosi, Differences in sensitivity between earthworms and enchytraeids exposed to two commercial fungicides, *Ecotoxicol. Environ. Saf.* 140 (2017) 177–184.
- [23] J. Amossé, S. Bart, A.R.R. Péry, C. Pelosi, Short-term effects of two fungicides on enchytraeid and earthworm communities under field conditions, *Ecotoxicology* 27 (2018) 300–312.
- [24] O.J. Owojori, A.J. Reinecke, P. Voua-Otomo, S.A. Reinecke, Comparative study of the effects of salinity on life-cycle parameters of four soil-dwelling species (*Folsomia candida*, *Enchytraeus doerjisi*, *Eisenia fetida* and *Aporrectodea caliginosa*), *Pedobiologia* 52 (2009) 351–360.
- [25] C. van Capelle, S. Schrader, J. Brunotte, Tillage-induced changes in the functional diversity of soil biota – a review with a focus on German data, *Eur. J. Soil Biol.* 50 (2012) 165–181.
- [26] P.F. Hendrix, R.W. Parmelee, D.A. Crossley, D.C. Coleman, E.P. Odum, P. M. Groffman, Detritus food webs in conventional and no-tillage agroecosystems, *Bioscience* 36 (1986) 374–380.
- [27] E. Nowak, Enchytraeids (Oligochaeta) in the agricultural landscape, *Pol. J. Ecol.* 52 (2004) 115–122.
- [28] E. Fouillet, L. Delière, N. Chartier, N. Munier-Jolain, S. Cortel, B. Rapidel, A. Merot, Reducing pesticide use in vineyards. Evidence from the analysis of the French DEPHY network, *Eur. J. Agron.* 136 (2022) 126503.
- [29] R.W. Sims, B.M. Gerard, *Earthworms*, FSC Publications, London, 1999.
- [30] ISO 23611-3, *Soil Quality — Sampling of Soil Invertebrates — Part 3: Sampling and Soil Extraction of Enchytraeids*, International Organization for Standardization, Geneva, 2007.
- [31] W.A.M. Didden, Population ecology and functioning of Enchytraeidae in some arable farming systems, in: *PhD Dissertation*, Agricultural University Wageningen, The Netherlands, 1991.
- [32] R. Röhrig, M. Langmaack, S. Schrader, O. Larink, Tillage systems and soil compaction—their impact on abundance and vertical distribution of Enchytraeidae, *Soil Tillage Res.* 46 (1998) 117–127.
- [33] P.C.J. van Vliet, M.H. Beare, D.C. Coleman, Population dynamics and functional roles of Enchytraeidae (Oligochaeta) in hardwood forest and agricultural ecosystems, *Plant Soil* 170 (1995) 199–207.
- [34] F.B. O'connor, Extraction of enchytraeid worms from a coniferous forest soil, *Nature* 175 (1955) 815–816.
- [35] K. Kobeticová, J. Schläghamerský, On the efficiency of three schemes of enchytraeid wet funnel extraction, 5th International Symposium on Enchytraeidae, *Newsl. Enchytraeidae* 8 (2003) 25–31.
- [36] R.M. Schmelz, R. Collado, A guide to European terrestrial and freshwater species of Enchytraeidae (Oligochaeta), *Soil Org.* 82 (2010) 1–176.
- [37] U. Graefe, R.M. Schmelz, Indicator values, strategy types and life forms of terrestrial Enchytraeidae and other microannelids, in: R.K.S. Schmelz (Ed.), *Newsletter on Enchytraeidae* 6, Universitätsverlag Rasch, Osnabrück, 1999, pp. 59–67.
- [38] M. Brooks, K. Kristensen, K. van Benthem, A. Magnusson, C.W. Berg, A. Nielsen, H. Skaug, M. Zächler, B. Bolker, glmmTMB balances speed and flexibility Among packages for zero-inflated generalized linear mixed modeling, *RIO J.* 9 (2017) 378–400.
- [39] J. Fox, S. Weisberg, *An R Companion to Applied Regression*, third ed., Sage, Thousand Oaks CA, 2019. <https://www.john-fox.ca/Companion/index.html>.
- [40] F. Hartig, DHARMA: residual diagnostics for hierarchical (multi-Level/mixed) regression models, R package (2019). version 0.2.4, <https://CRAN.R-project.org/package=DHARMA>.
- [41] V. R Development Core Team, *R: A Language and Environment for Statistical Computing*, R Foundation for Statistical Computing, Austria, 2015. ISBN 3-900051-07-0, URL, <http://www.R-project.org/>.
- [42] M. Albrecht, D. Kleijn, N.M. Williams, M. Tschumi, B.R. Blaauw, R. Bommarco, A. J. Campbell, M. Dainese, F.A. Drummond, M.H. Entling, D. Ganser, G. Arjen de Groot, D. Goulson, H. Grab, H. Hamilton, F. Herzog, R. Isaacs, K. Jacot, P. Jeanneret, M. Jonsson, E. Knop, C. Kremen, D.A. Landis, G.M. Loebe, L. Marini, M. McKeerchar, L. Morandin, S.C. Pfister, S.G. Potts, M. Rundlöf, H. Sardiñas, A. Sciligo, C. Thies, T. Tschamtkke, E. Venturini, E. Veromann, I.M.G. Vollhardt, F. Wäckers, K. Ward, D.B. Westbury, A. Wilby, M. Woltz, S. Wratten, L. Sutter, The effectiveness of flower strips and hedgerows on pest control, pollination services and crop yield: a quantitative synthesis, *Ecol. Lett.* 23 (2020) 1488–1498.
- [43] EFSA Scientific Committee, *Recovery in environmental risk assessments at EFSA*, *EFSA J.* 14 (2016) 4313, <https://doi.org/10.2903/j.efsa.2016.4313>.
- [44] C. Ockleford, P. Adriaanse, P. Berny, T. Brock, S. Duquesne, S. Grilli, A. F. Hernandez-Jerez, S.H. Bennekou, M. Klein, T. Kuhl, R. Laskowski, K. Machera, O. Pelkonen, S. Pieper, M. Stemmer, I. Sundh, I. Teodorovic, A. Tiktak, C. J. Topping, G. Wolterink, P. Craig, F. de Jong, B. Manachini, P. Sousa, K. Swarowsky, D. Auteri, M. Arena, S. Rob, EFSA PPR Panel, Scientific Opinion addressing the state of the science on risk assessment of plant protection products for in-soil organisms, *EFSA J.* 15 (2017) e04690.
- [45] J.C.Y. Marinissen, Colonisation of arable fields by earthworms in a newly reclaimed polder in The Netherlands. Preliminary results, in: G.K. Veeresh, D. Rajagopal, C.A. Viraktamath (Eds.), *Proceedings of the 10th Int. Soil Zoology Colloquium*. Advances in Management and Conservation of Soil Fauna, 1991. India.
- [46] H. Eijsackers, Earthworms as colonizers of natural and cultivated soil environments, *Appl. Soil Ecol.* 50 (2011) 1–13.

- [47] A.R. Hof, P.W. Bright, The impact of grassy field margins on macro-invertebrate abundance in adjacent arable fields, *Agric. Ecosyst. Environ.* 139 (2010) 280–283.
- [48] F. Ricci, L. Bentze, D. Montagne, S. Houot, M. Bertrand, C. Pelosi, Positive effects of alternative cropping systems on terrestrial *Oligochaeta* (*Clitellata*, *Annelida*), *Soil Org.* 87 (2015) 71–83.
- [49] A. Beylich, U. Graefe, Investigations of annelids at soil monitoring sites in Northern Germany: reference ranges and time-series data, *Soil Org.* 81 (2009) 175–196.
- [50] J. Schlaghamerský, K. Kobetičová, The impact of cattle pasturage on small annelids (Annelida: Enchytraeidae, Tubificidae, Aeolosomatidae) in grasslands of the White Carpathians (Czech Republic), *Eur. J. Soil Biol.* 42 (2006) S305–S309.
- [51] J. Singh, S. Singh, A.P. Vig, Extraction of earthworm from soil by different sampling methods: a review, *Environ. Dev. Sustain.* 18 (2016) 1521–1539.
- [52] M.A. Callahan, P.F. Hendrix, Relative abundance and seasonal activity of earthworms (Lumbricidae and Megascolecidae) as determined by hand-sorting and formalin extraction in forest soils on the southern Appalachian Piedmont, *Soil Biol. Biochem.* 29 (1997) 317–321.
- [53] R.J. Blakemore, Critical decline of earthworms from organic origins under intensive, humic SOM-depleting agriculture, in: *Soil Systems*, 2018.
- [54] J. Römbke, R.M. Schmelz, C. Pelosi, Effects of organic pesticides on enchytraeids (*Oligochaeta*) in agroecosystems: laboratory and higher-tier tests, *Front. Environ. Sci.* 5 (2017).