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Predation on epigeic, endogeic and anecic earthworms by carabids active in spring and autumn

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Abstract

Background and purpose: *Generalist predatory carabid beetles can control the abundance of a range of prey species within ecosystems, including certain pests. In terms of bio-control, these unspecialised predators may be sustained in the field when pest populations are low by predating on other animals such as earthworms. The aim of this study was to reveal patterns in predation by a community of carabids in the field on different earthworm species with respect to anecic, endogeic and epigeic earthworm ecotypes.*

Materials and methods: *We utilised DNA extracted from the gut content of 23 carabid species to reveal predation on earthworms directly in the field, comparing spring and autumn active species. The DNA was then screened using PCR with five earthworm species-specific primers.*

Results and discussion: *Our results show that 20 species, which accounted for 53% of all tested individual beetles, were positive for earthworms, with similar proportions in the spring and autumn samples and between the sexes. Earthworms from all three ecotypes were confirmed within the predator guts and were widely consumed within the carabid community.*

Conclusions: *These results suggest that predation on earthworms might be an important mechanism sustaining populations of generalist predatory carabids in the field, which can be advantageous for biological control. Therefore, management systems that maintain a healthy soil with all three ecotypes of earthworm present is likely to be beneficial for carabids and indirectly for control of plant pests.*

INTRODUCTION

Previous studies on gut content analyses and field observations have confirmed that predatory carabid beetles feed mainly on soil invertebrates such as earthworms, slugs, snails, woodlice, springtails and various insects *(e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, etc.)*. As such, carabids can in theory contribute to the control of pests and/or invasive alien species within ecosystems by predation *(e.g., 10, 11, 12, 13, 14, 15, 16, 17* etc.). Still we lack some more precise information about their consumption of pests but also of other animals that help to sustain beneficial predatory carabids in the field when pest populations are low.

Many adult carabid beetles, as well as their prey, are active in the upper soil layers, the soil surface and the litter layer, where they feed. Although some carabid species can dig (i.e., *Scarites* spp.) or use holes that are already present in the soil to move deeper into the substrate *(18)*, most of them search for food on the soil surface.

Earthworms, however, exhibit some degree of vertical stratification within the soil and can be categorized into epigeic, endogeic and anecic

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ecological groups. Epigeic species live on the soil surface, where they feed on leaf litter and usually do not make burrows (e.g., *Lumbricus rubellus, Lumbricus castaneus, Satchellius mammalis*) *(19)*. Endogeic species make horizontal burrows through the upper part of the soil (e.g., Allolobophora chlorotica) *(19)*. Anecic earthworms live deeper, but make vertical burrows in the soil, where they feed on leaves that they drag into their burrows from the soil surface (e.g., *Lumbricus terrestris* and *Aporrectodea longa*) *(19)*. In European soils, the earthworm population is characterised by a mean density of ~100 individuals per m^2 and a mean biomass of 5 g dry weight per m^2 and therefore contributes significantly to soil biomass *(20)*.

Although, earthworms compose the main component of the diet of many carabid species *(8, 9)* and may improves fitness parameters in some carabid species *(6)*, the patterns in predation by a community of carabids in forest ecosystem on different earthworm species with respect to anecic, endogeic and epigeic earthworm ecotypes have not been studied jet.

Carabids differ in morphological (e.g. procerisation, cychrisation) and ecological traits (e.g. olfactory or visual hunters, winged or wingless specimens) and seasonal breeding and feeding activity *(2, 21)*, and these differences may affect their diets. Members of the same species may even change their diet through the year. For example, it was confirmed that *Carabus violaceus* may switch from a mainly earthworm diet in the spring to a slug-based diet in the autumn *(22)*.

To overcome the shortcomings of classical methods (e.g., direct observations in the field, microscopic analysis of the gut contents) we utilised molecular methods to detect with high precision the semi-digested DNA from different earthworm species in the carabid gut *(e.g., 7, 15, 23, 24, 25)*.

Taking into account differences in the traits of carabid species within a community and predation on the three ecological types of earthworms, we screened a gut content of a range carabid species collected in the field in late spring/early summer and in autumn in two PCR multiplex assays *(7)* for carabid diet analysis. Our assays comprises already designed and optimised nine primer pairs *(7)* to target five earthworm species, each species belonging to one of the three ecological groups of earthworms.

Considering carabids' generalist feeding, we hypothesise that in addition to the epigeic and endogeic earthworms located in the upper soil, adult carabids may also predate on anecic earthworms when these earthworms collect leaves from the soil surface.

MATERIALS AND METHODS

Material used for molecular analyses

All invertebrates were collected from five sites located in deciduous woodlands (two sites in Croatia, in Mt. Medvednica near Zagreb, and three sites located in woodland patches surrounded by arable fields in Wales, UK, one in Llantrisant and two in Rudry. Earthworms were sampled from the topsoil layer in three visits in spring and three in autumn in both 2007 and 2010 by digging and hand collection (using five 50 cm^2 quadrats per site to a depth of 10 cm). All collected earthworms were maintained in separate Petri dishes containing moist filter paper to empty their guts for 24 to 48 hours. They were then killed at –80 °C. Pitfall trapping and hand sampling were used to collect adult carabid beetles. As described in Šerić Jelaska et al. *(8)*, five traps (0.5 L plastic cups containing no preservative) at each site were deployed for approximately three weeks from May until the end of June and again for three weeks from mid-September until the end of October. Trapping was conducted in 2007 in Croatia and in 2010 in the UK. The traps were emptied every morning, and each individual beetle was placed into a plastic tube and killed at –80 °C. All carabid beetles and adult earthworm specimens were identified to the species level based on their morphological characteristics using identification keys *(19, 26, 27, 28)*. Beetles were also sorted by sex.

DNA from collected earthworm species was extracted using the DNeasy Blood & Tissue Kit (Qiagen), and used as a positive control during gut content analyses. Additionally, we extracted the DNA from another 35 soil invertebrate species representing non-target potential prey from the field (the same set of animals was used in Šerić Jelaska et al. 2014 a, b, cf Supporting Information). All primer pairs were thus tested for cross-amplification against carabid predator DNA as well as against the DNA of other soil invertebrate taxa. The non-target organisms were tested individually, and cross-amplifications were not found, confirming that the primers were specific to the prey species for which they were designed.

Prior to molecular analyses, carabid foreguts were dissected as described in Symondson et al. *(5)*. Carabid specimens collected in UK were weighed before and after the dissection. DNA from foreguts was extracted using DNeasy Blood & Tissue Kit (Qiagen).

All extractions were tested for the presence of DNA in the previous work of Šerić Jelaska et al. *(8)* by PCR using general invertebrate primers for a 710 bp fragment of the mitochondrial cytochrome oxidase I (COI) gene *(29)*, and all conditions were described in detail in Šerić Jelaska et al. *(8)*.

Screening of field-caught predators to identify earthworms in predator guts

To screen the gut contents of the field-caught carabids, we used earthworm species-specific primer pairs *(7)* for five common species belonging to the three ecological groupes: epigeic *Lumbricus castaneus* and *L. rubellus,* endogeic *Allolobophora chlorotica,* and anecic *Aporrectodea longa* and *L. terrestris*. For *Allolobophora chlorotica* we used primer pairs designed for five lineages of the species (Table 1).

Table 1. *Details of primer sequences (5'-3') used in two PCR multiplex assays (assay A with primers for multiple* A. chlorotica *lineages and assay B for other four earthworm species) (King et al. 2010).*

Species	Multiplex	Primer name	Primer sequence	Amplicon size (bp)
Allolobophora chlorotica	A	COI AchL1F4	AAATTGATTACTACCYCTG	231
		COI AchL1R2	GAAGCACCTGCTAGRTGG	
		COI-AchL2A-F5	TGCAGTAGAAAAGGGTGCG	151
		COI-AchL2A-R3	AGTAATAAAATTAATGGCA	
		COI-AchL2B-F3	CATCACTAATCCTTCTAGTG	126
		COI-AchL2B-R3	AGAAGATAGCTAAGTCTACG	
		COI-AchL3-F2	TGGAAATTGACTATTACCAC	261
		COI-AchL3-R2	ATGAAATTAATTGCCCCGAG	
		COI-AchL4-F2	CCAACTATATAATACTATCGTT	152
		COI-AchL4-R2	ATCTCATGTTATTGAGTCGA	
Aporrectodea longa	B	COI-Al-F2	TGGCTTCTACCTCTAATACT	213
		COI-Al-R2	ATGAAGGGAGAAGATGGCCA	
Lumbricus castaneus	B	COI-Lc-F2	AACTGACTCCTCCCACTAAT	189
		COI-Lc-R2	AGAAGGTCCTGCGTGAGCT	
L. rubellus	B	COII-Lr-F3	AGACGGTAATCTCCTGGAAGT	164
		COII-Lr-R2	CTTCGTATTCTCTATATCACA	
L. terrestris	B	COII-Lt	GAATCTATTTCYACATTTAAGAA	256
		COII-Lt-R2	CGGCTATGCTCTYCTAGCAC	

Two diagnostic multiplex PCRs were used to screen the gut of each field-caught beetle for the presence of multiple earthworm species. The earthworm primers *(7)*, combined in 2 multiplex reactions, are listed in Table 1. The multiplex PCR reactions were performed in a 10 µL reaction mix containing 5 µL of Multiplex PCR Master Mix (Qiagen), 0.2 µM each primer, 10 µg bovine serum albumin (New England Biolabs), dH_2O (Qiagen) and 1.2 µL extracted DNA. After the initial denaturing step at 95 °C for 15 min, amplification proceeded for 35 cycles at 94 °C for 30 s, 56 °C (for multiplex A) or 56.5 °C (for multiplex B) for 1 min 30 s, 72 °C for 1 min 30 s and a final extension at 72 °C for 10 min.

All PCRs included positive (target prey) and negative (sterile water instead of DNA) controls. The PCR products were separated and visualised on a 2% agarose gel stained with ethidium bromide (concentration 0.0750 µg/mL) after 40 min at 120 V. A PCR was considered positive by the presence of a band of the target size on the gel.

All samples were screened twice for the target prey using the same PCR conditions, and presence of bands in each reaction was counted as positive.

Data analyses

Statistical analyses were carried out using Statistica 9.1 (Statsoft Inc., 2010) and PRIMER 6 (2006). Cluster analyses with Bray-Curtis similarity index was employed for comparison of carabid species with respect to consumed earthworms from different ecological groups. Kruskal-Wallis ANOVA (comparison among multiple independent samples) as well as accompanying post hoc multiple comparison tests were used to analyse the relationship of beetle gut and body mass with prey type (three earthworm ecotypes), and check whether presence of earthworm DNA had any effect on gut mass. Spearman rank-order correlations between the body and gut weights were calculated only for the UK populations.

The predation data were presented as the number of beetles testing positive for earthworm consumption in the two seasons (late spring/early summer and autumn). Due to potential differences in prey DNA detectability among the different combinations of predator and prey species *(e.g. 30)*, only the raw predation event data obtained from the positive PCR results are presented without further statistical analysis.

RESULTS

Earthworm consumption by carabids in the field

The foregut content of the field-caught carabid species was positive for earthworm species from each of the three ecological groups. Of all tested carabids (N=317), 168

Table 2. *Field-caught carabid species screened for three earthworm ecotypes; number of individual beetles tested and number of beetles testing positive for each prey group*
Numbers in the brackets indicate number of individuals according to their sex (♀ female and ♂ males).

Species	Sites		Number of beetles testing positive for earthworms			
	(1,2 in Croatia 3-5 in the UK)	No. ind. (♀/♂)	All positives	Anecic (♀/♂)	Epigeic (♀/♂)	Endogeic (♀/♂)
Nebria brevicolis	3,4,5	87 (33/54)	35	23 (10/13)	19 (11/8)	9 (3/6)
Abax parallelus	1,2	68 (39/29)	50	38 (21/17)	22 (8/14)	27 (15/12)
Abax parallelepipedus	1,2,3,4,5	63 (20/43)	34	25 (7/18)	13 (5/8)	13 (4/9)
Carabus nemoralis	1,2	18 (7/11)	6	4 (3/1)	3 (3/0)	0/0
C. ullrichi	2	13 (8/5)	8	7 (5/2)	2 (1/1)	1 (0/1)
C. violaceus	1,2,4,5	9 (6/3)	4	1 (1/0)	1 (1/0)	4 (4/0)
Cychrus attenuatus	1,2	7 (2/5)	5	4 (1/3)	0 (0/0)	3 (2/1)
Pterostichus madidus	3,4,5	7 (6/1)	3	2 (1/1)	2 (2/0)	1 (1/0)
Agonum sp.	5	6 (3/3)	0	0/0	0/0	0/0
C. convex	2	6 (6/0)	3	2 (2/0)	2 (2/0)	0/0
C. coriaceus	2	6 (3/3)	4	4 (3/1)	2 (1/1)	1 (1/0)
C. intricatus	1,2	6 (3/3)	4	2 (2/0)	2 (0/2)	1 (0/1)
Leistus fulvibarbis	3,4	3 (0/3)	1	0/0	0/0	1 (0/1)
P. fasciatopunctatus	2	3 (2/1)	2	1 (1/0)	1 (1/0)	1 (0/1)
C. problematicus	4	2 (1/1)	0	0/0	0/0	0/0
Molops piceus	2	2 (0/2)	2	2 (0/2)	0/0	2 (0/2)
P. melanarius	3,4	2 (1/1)	1	1 (0/1)	0/0	0/0
P. transversalis	1,2	2 (0/2)	2	2 (0/2)	0/0	0/0
Synuchus vivalis	3,4	2 (2/0)	1	1 (1/0)	1 (1/0)	1 (1/0)
Aptinus bombarda	1	1	0	0/0	0/0	0/0
Notiophilus rufipes	2	1	1	0	1	0
Bembidion nigricorne	3	1 (1/0)	1	0/0	0/0	1 (1/0)
B. quadrimaculatum	3	1 (0/1)	1	0/0	1 (0/1)	0/0

(53%) were positive for at least one earthworm species, and the 149 (47%) individuals remaining were negative even after a second PCR screening. Most of the individual beetles (N=101) were positive for only one earthworm species, 48 individuals were positive for two prey species, 16 for three prey species and only three individuals showed positive for four earthworm prey species.

Of the 23 carabid species recorded at the five sites, only three species (*Agonum* sp., *Carabus problematicus* and *Aptinus bombarda*) were negative for earthworm species, while the rest were positive for at least one earthworm species (Table 2). Sixteen of the 23 carabid species were positive for anecic earthworms, 14 for epigeic earthworms and 14 for endogeic earthworms. Ten species were positive for earthworm species from all three ecotypes. Five species were positive for earthworms from two ecotypes (Table 2). When analysed for similarity using positive/negative data after screening the carabid guts for five earthworm species, most of the carabid beetles could be grouped into two sets of species with more than 50% of similarity (Bray-Curtis similarity): *Pterostichus fasciatopunctatus*, *P. madidus*, *Carabus violaceus*, *C. convexus* and *C. coriaceus*, together with two *Abax* species and *Nebria brevicollis* in one group; *Carabus ullrichi*, *C. intricatus*, *Molops piceus*, *Cychrus attenuatus* and *Synuchus vivalis* in another group (Fig. 1).

Of all tested individual beetles, 77 and 41 were positive for *A. longa* and *L. terrestris*, respectively, both belonging to the anecic ecotypes, 66 were positive for *A. chlorotica* belonging to the endogeic ecotypes, and 55 and 18 individual beetles were positive for epigeic *L. castaneus* and *L. rubellus*, respectively. In addition, earthworms from all three ecotypes were predated in both seasons (91 individuals were positive in late spring/early summer and 77 in autumn) (Table 3, Fig. 2).

Overall, 148 female and 168 male beetles were collected from the five sites, of which 127 females and 130 males were positive for earthworms. Furthermore, all three groups of earthworms were predated by both sexes (Table 2, Fig. 3). Because the PCR data on carabid consumption was not adjusted by calculating the time of

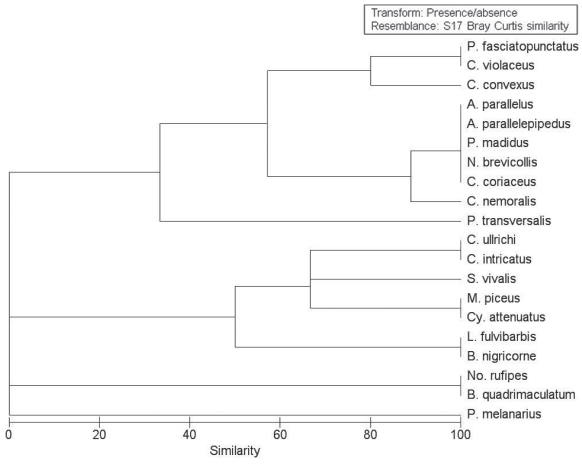

Fig. 1. *Dendrogram depicting results of cluster analysis for similarity in carabid species according to the presence/absence (PCR positive/negative data) of each earthworm species in carabid guts using Bray-Curtis resemblance measure.*

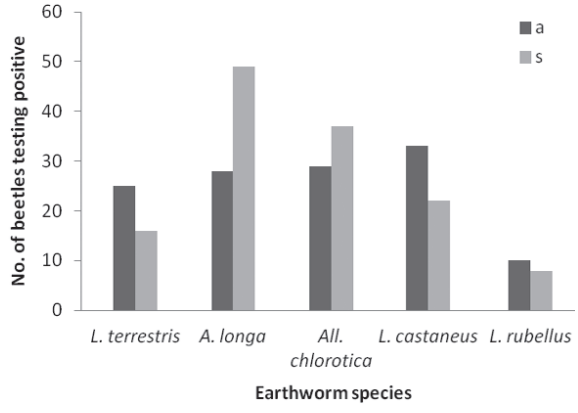

Fig. 2 *Number of beetles testing positive in spring/summer (s) and autumn (a) for each earthworm species are shown as bars.*

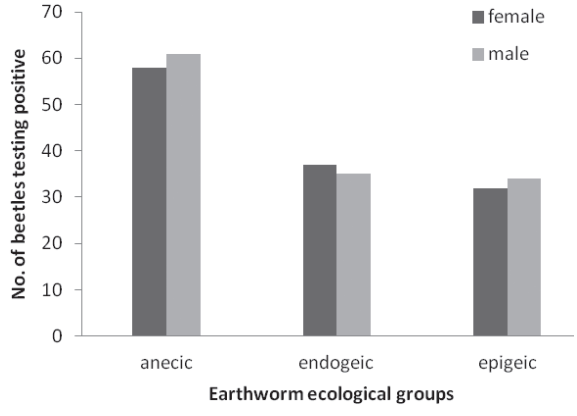

Fig. 3 *Number of female and male beetle individuals testing positive for earthworms classified in three ecological groups.*

digestion for each predator-prey combination in this study, all proportions presented above should be considered only as approximate.

The gut weight was significantly correlated with the body weight of the individual beetles (Spearman correlation for all UK beetles tested: N=132, R=0.48267, and for UK beetles that tested positive: N=55, R=0.555231, at $p<0.05$). Kruskal-Wallis ANOVA did not show significant effect of presence of earthworms from three ecotypes on the gut weight or body weight of PCR-positive individual beetles (H for gut weight (N=55) =8.310491 p=0.5985; H for body weight (N=55) =6.172715 p=0.8005).

DISCUSSION

This study confirm for the first time predation by carabid beetles on earthworms belonging to all three ecological groups (epigeic, endogeic and anecic) at the community level. Despite different spatial distributions of earthworms within the soil substrate, all three ecotypes of earthworms were detected as prey within the carabid assemblages in both seasons. All except three carabid species were positive for at least one earthworm species, demonstrating widespread predation on earthworms within the beetle community. Because the carabid assemblages considered in here were composed mainly of epigeic, nocturnal, and generalist predators, predation on anecic earthworms, which are active at night, when they collect leaves from the soil surface, was also expected and thus confirms our hypothesis. Interestingly, some carabid species such as *P. melanarius* or *C. ullrichi* that can be active during the day *(31)* were positive for anecic earthworms.

The presence of anecic earthworms in the gut has also been confirmed by King et al. *(7)* for *P. melanarius* species. Our study further contributes to previous studies by screening guts from the entire community of carabid beetles for the presence of earthworms from the three ecological groups. In addition, we screened carabids from woodland communities, while the studies by Symondson et al. *(24)* and King et al. *(7)* were in arable fields.

An anecic *A. chlorotica*, was confirmed in the guts of the tested carabids but was not collected in the field. The reason this species was not observed in the field could be due to the digging and hand collecting method used in the topsoil layer, which can underestimate the abundance of the fauna located in the deeper soil layers. The results of King et al. *(7)* suggested that earthworms were predated proportionally to their field abundances. Similar findings were reported by Scheller *(10)*, who detected aphid remains in proportion to the aphid field density for *Bembidion lampros*, *Agonum dorsale*, *Pterostichus melanarius* and *Loricera pilicornis*. Because all five earthworm species were detected in the carabid guts, we can indirectly confirm their presence in the field as well. Although indirect method, our PCR multiplex system could be a sensitive for detecting cryptic species via the gut content analysis of their predators *(32)*.

The substantial number of individual beetles positive for *A. chlorotica* despite its strong defensive secretions *(7)*

Table 3. *Number of earthworms sampled in each field, in spring and autumn and their proportion (%) within seasonal samples; and number of beetles tested positive for each prey species at each site and portion of (%) carabid beetles testing positive in both seasons.*

		Late Spring/Early Summer				Autumn			
Earth-worm species	Plots	Prey abundance in the field	% of prey in spring	N (beetles tested positive)	% of beetles positive in spring	Prey abundance in the field	% of prey in autumn	N (beetles tested positive)	% of beetles positive in autumn
Lumbricus rubellus									
	1	4	4.88	–	–	9	0.12	2	2.60
	2	–	–	1	1.10	13	0.17	–	–
	3	3	3.66	–	–	3	0.04	–	–
	4	–	–	2	2.20	–	–	1	1.30
	5	–	–	5	5.49	1	0.01	7	9.09
L. castanea									
	1	2	2.44	1	1.10	–	–	13	16.88
	2	–	–	–	–	–	–	–	–
	3	–	–	2	2.20	1	0.01	8	10.39
	4	–	–	3	3.30	–	–	1	1.30
	5	3	3.66	16	17.58	–	–	11	14.29
L. terrestris									
	1	1	1.22	3	3.30	3	0.04	7	9.09
	2	–	–	1	1.10	1	0.01	3	3.90
	3	1	1.22	3	3.30	3	0.04	1	1.30
	4	–	–	–	–	–	–	–	–
	5	–	–	9	9.89	–	–	14	18.18
Allolobophora chlorotica									
	1	–	–	2	2.20	–	–	9	11.69
	2	–	–	1	1.10	–	–	–	–
	3	–	–	5	5.49	–	–	1	1.30
	4	–	–	4	4.40	–	–	–	0.00
	5	–	–	25	27.47	–	–	19	24.68
Aporrectodea longa									
	1	–	–	3	3.30	–	–	8	10.39
	2	–	–	5	5.49	–	–	1	1.30
	3	–	–	2	2.20	–	–	1	1.30
	4	–	–	5	5.49	–	–	4	5.19
	5	–	–	34	37.36	–	–	14	18.18
Satchellius mammalis									
	1	–	–	–	–	1	0.01	–	–
Eisenia fetida									
	1	1	1.22	–	–	–	–	–	–
	4	–	–	–	–	1	0.01	–	–
	5	1	1.22	–	–	1	0.01	–	–
Ap. smaragdina									
	4	20	24.39	–	–	8	0.10	–	–
	5	5	6.10	–	–	3	0.04	–	–
Juveniles – unidentified									
	1	8	9.76	–	–	6	0.08	–	–
	2	10	12.20	–	–	6	0.08	–	–
	3	8	9.76	–	–	4	0.05	–	–
	4	2	2.44	–	–	1	0.01	–	–
	5	13	15.85	–	–	12	0.16	–	–
Total abundance of earthworms in the filed		82				77			
Number of individual beetles tested positive				91				77	

may demonstrate that not only *P. melanarius* but also some other carabid species have no aversion to consuming this species, as is the case with the secretions of some slugs *(33, 34)*.

Some seasonality in the predation rates of different earthworm species were observed between the spring/summer and autumn samples. *A. longa* and *A. chlorotica* were detected in carabid guts at higher rates in spring/early summer than in autumn, which could be due to lower ambient temperatures that could slow the activity of carabids. On the other hand, *Lumbricus* species were detected in higher numbers in carabid guts in autumn than in the spring samples. Those species were also more abundant in the field in autumn, except for *L. terrestris*. As has already been noted, the sampling by hand sorting could also underestimate the populations of *L. terrestris*, an anecic earthworm species, in the field. Then again, chemical extraction methods also have biases and anecic species may be over-represented as the chemical going directly down their vertical burrows *(35)*.

Among the carabids, the most abundant species in spring was *A. parallelepipedus*, with more individual beetles being positive for anecic *A. longa* than for endogeic or epigeic earthworms. In autumn, the most dominant species was *N. brevicollis*, with more individuals positive for epigeic *L. castanea* and for anecic *L. terrestris*.

The cluster analysis (Fig 1) grouped carabids according to their seasonal activities into spring active and autumn active carabids or biseasonal species groups. King et al. *(7)* observed significantly higher predation by *P. melanarius* on *Aporrectodea caliginosa* in July and on *A. chlorotica* in August than was expected *(7)*. In the study by King et al. *(7)*, *P. melanarius* showed the lowest predation rate on *A. chlorotica* in September but higher predation on *L. rubellus* and *L. terrestris* in September than in July. Because we did not perform feeding trials for each trophic combination, we could not adjust the raw PCR data to conduct preference analyses; our data are therefore indicative, reinforcing previous studies *(e.g., 7)* noting the need for further research to reveal if such patterns indicate selective predation or are a result of random feeding.

Specialisation on specific prey is infrequent among carabids *(36)*. Therefore, as primarily generalist predators, carabids can be used in integrated pest management in agriculture and forestry and management systems that help to sustain the beetles in the field when pest numbers are low may be beneficial, such as ensuring healthy populations of non-pest alternative prey. A similar study confirmed collembolans as alternative prey that helps to sustain spiders in the field *(37)*.

Three carabid species, *Agonum* sp., *C. problematicus* and *Aptinus bombarda*, were not positive for any of the five earthworm species, which may be due to only a few individuals being tested (6, 2 and 1, respectively).

Carabid beetles, as well as their prey (earthworms, slugs, springtails, woodlice, etc.), are mainly soil fauna and occupy different soil layers, from the deeper soil to the soil surface and the litter layer, where they feed or can be eaten by other animals. Therefore, different toxic compounds, primarily derivatives from various insecticides that end up in the soil, are taken up by earthworms or plants and then by herbivores, such as slugs, and may be transferred from the soil to higher trophic levels *(e.g., 8, 38, 39)*. According to our study, earthworms from all three eco-types, appear as an important food source for carabids and thus can greatly contribute to the transfer of pollutants from the soil to other ecosystems *(e.g., 8, 39, 41)*.

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Appendix 1. *Field-caught carabid species screened for five earthworm species and number of individual species testing positive for each prey species.*

	L. terrestris	*A. longa*	*L. castaneus*	*L. rubellus*	*A. chlorotica*
A. parallelepipedus	7	18	10	3	13
A. parallelus	15	23	17	5	27
B. nigricorne	0	0	0	0	1
B. quadrimaculatum	0	0	1	0	0
C. convexus	0	2	0	2	0
C. coriaceus	1	3	1	1	1
C. intricatus	0	2	2	0	1
C. nemoralis	2	2	1	2	0
C. ullrichi	0	7	2	0	1
C. violaceus	0	1	0	1	4
Cy. attenuatus	1	3	0	0	3
L. fulvibarbis	0	0	0	0	1
M. piceus	1	1	0	0	2
No. rufipes	0	0	1	0	0
N. brevicollis	11	12	17	2	9
P. transversalis	0	2	0	0	0
P. fasciatopunctatus	0	1	0	1	1
P. madidus	1	1	1	1	1
P. melanarius	1	0	0	0	0
S. vivalis	1	0	1	0	1