Seasonal variations in macrofauna distribution according to the distance from a herbaceous strip in a Mediterranean alley cropping plot

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2   **herbaceous strip in a Mediterranean alley cropping plot**

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Abstract

In alley cropping systems, herbaceous strips planted with trees are added into cropped fields. These systems could provide a favorable habitat for many organisms, producing shade in summer and litter in autumn, but their impacts on soil macrofauna are poorly studied, with most samplings limited to one date and therefore one season only. This study investigated the seasonal variations in the spatial distribution of soil macrofauna in a Mediterranean alley cropping plot. We sampled three different shading treatments: a strong shading treatment with lightly-pruned trees, a light shading treatment with pollarded trees, and a no-shade treatment without trees in the herbaceous strips. We measured the density of macrofauna individuals classified into different taxonomic and functional groups by the extraction of 25*25*25 cm soil cores in the herbaceous strip (with or without trees), and in the crop alley at 1 m and 2.5 m from the herbaceous strip. We found a similar response of macrofauna density to the distance from the herbaceous strip for all shading treatments. However, this response was strongly dependent on the season. In spring, just after soil tillage, most soil macrofauna presented higher densities in the herbaceous strip than in the crop alley, except for plant feeders. In early summer, this trend was mostly visible for earthworms, Diplopoda and adult Coleoptera. In autumn, higher densities in the herbaceous strip than in the crop alley were only significant for anecic and endogeic earthworms, while other macrofauna categories presented similar densities at all distances from the herbaceous strip, due to an increase in macrofauna density in the crop alley. These large seasonal variations in spatial distribution of soil macrofauna highlight the importance of temporal repetitions when studying community responses to agroforestry system design and management. The mechanisms by which population densities increased in the crop alley a few months after soil perturbation require further investigation.

1. **Introduction**

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We are facing a global decrease in biodiversity, partly associated with landscape simplification and with current dominant agricultural practices (Habel et al., 2019; McLaughlin and Mineau, 1995). Agroforestry, or the association of trees and agriculture, can promote an increase in animal biodiversity in cropped fields, because of the addition of new and more diverse habitats (Jose, 2009). For example, agroforestry has been shown to impact positively the presence of birds and of pollinator insects compared to simplified agricultural areas (Udawatta et al., 2019). Its impacts on soil dwelling fauna have been far less studied, especially in the Mediterranean zone, and so far work has focused mainly on earthworms or organisms of the soil surface such as adult carabids (Marsden et al., 2020).

In addition to its purely conservational value, soil-dwelling fauna performs essential roles in ecosystem functions, and increases in its diversity and density could improve the multifunctionality of agricultural land (Bender et al., 2016). Among them, litter-feeders (mainly earthworms, woodlice and millipedes) contribute to the decomposition of organic matter and to associated nutrient turnover through litter comminution and modification of microbial activity in their guts and fresh feces (Barrios, 2007; David, 2014; Frouz, 2018), and

76 to carbon storage in old feces (Frouz, 2018; Wolters, 2000). Some earthworms incorporate

77 fresh organic matter into the mineral soil, where it can be protected from degradation and

78 contribute to carbon storage (Bossuyt et al., 2005), which has also been shown for millipedes

79 (Filser et al., 2016). Conversely, earthworms feeding on stabilized particulate organic matter

80 contribute to mineralize it (Lavelle et al., 2006). Earthworm activity can also improve soil

81 structure by the creation of burrows, soil decompaction and the creation of an organo-mineral

82 horizon (Lavelle et al., 1998). In addition, the regulation of the invertebrate community by

83 animal feeders is increasingly studied for pest control (Holland et al., 2017), and is performed

84 by organisms living in the soil (Chilopoda, most larvae of Coleoptera, etc) or dependent on

85 the soil for their larval stage or overwintering (insect predators at their adult stage) (Geiger et

86 al., 2009). The soil can also host larvae of future crop pollinators. Practices promoting soil

87 fauna abundance and diversity should thus be favored in a context of reduction of chemical

88 inputs.

89 In alley cropping agroforestry, trees are organized in rows in order to facilitate crop

90 management practices along alleys. A strip of around 2 m wide is uncultivated below the

91 trees, and an associated understory vegetation is initially sown or appears spontaneously,

92 mostly herbaceous. The lack of disturbance, permanent soil cover and source of additional

93 food resources from this non-exported vegetation creates a specific habitat in these

94 herbaceous strips. They are an overwintering site for most surface-dwelling invertebrates

95 living in alley cropping systems (Boinot et al., 2019), present higher earthworm densities than

96 crop fields and crop alleys (Cardinael et al., 2019), and host high densities of woodlice

97 (D'Hervilly et al., 2020; Pardon et al., 2019, 2020) and millipedes (Pardon et al., 2019). This

98 herbaceous strip could particularly be a reservoir of macrofauna, i.e. soil fauna larger than 2

99 mm, then able to disperse towards the crop alley over wider distances than mites or

100 springtails, for example. However, proof is still lacking that the herbaceous strip really

101 benefits the establishment of diverse and abundant communities in the crop alley, as studies

102 of spatial heterogeneity in alley cropping systems are scarce and show contrasting results. For

103 example, no variation in earthworm density was found in crop alleys with the distance from

104 the herbaceous strip by Cardinael et al. (2019), while earthworm density was negatively

105 impacted by herbaceous strip proximity in some sites (D'Hervilly et al., 2020; Rivest et al.,

106 2020). In addition, the effect of the distance from the herbaceous strip is also probably

107 influenced by tree properties, as they determine the quantity and quality of litter added in the

108 crop alley and the strength and range of the microclimate effect. For example, woodlice and

109 millipede activity were positively affected by old but not young tree row proximity in one

110 study (Pardon et al., 2019), and a gradient in earthworm density from the tree to the center of

111 the crop alley was found in a Canadian system for some tree species only (Price and Gordon,

112 1999). Finally, most studies rely on one single sampling season, while conceptually, dynamic

113 exchanges between herbaceous strips and crop alleys can be expected to depend on the period

114 of the year, driven by adverse or favorable properties of the crop alley or by soil organism life

115 cycles affected by climatic conditions.

116 This study aims to determine the seasonal variations of soil macrofauna distribution according

117 to the distance from a herbaceous strip (HS) for different shade treatments in an alley

118 cropping plot. We made the hypotheses that (i) HS can be a specific shelter for macrofauna,

119 leading to higher faunal densities in HS than in crop alleys; (ii) this bimodal density

120 distribution is particularly visible during certain seasons when conditions are not favorable for

121 soil fauna in the crop alley, and replaced by a gradient-type distribution during other seasons

122 due to soil macrofauna dispersal from the HS towards the crop alley; and (iii) this HS shelter

123 effect is less visible in the presence of strongly-shading trees than in the presence of lightly-

124 shading trees or without trees, as tree-induced changes in microclimate and in litter and soil

125 organic matter presence may render crop alley conditions more favorable for soil macrofauna.

2. Methods

2.1. Site description

The study took place in an experimental alley cropping plot of 2,250 m^2 located near Vézénobres in the South of France (44.058 N, 4.111 E), included in a wider alley cropping site. The climate is Mediterranean (temperature of 15.2 °C and 760 mm of precipitation per year on average, French Institute of Meteorology Météo France). The soil is on recent alluvial deposits, loamy in the first 25 cm depth with a pH of around 8, 16 % of $CaCO_3$ and a CEC of 9.5 cmol$^+$ kg^{-1} on average (basic soil properties are detailed in Table 1 and Table S1 in the Supplementary resources). These soil properties were determined at the beginning of the sampling in 2018. Texture was analyzed on decarbonated soil sieved at 2 mm. Bulk density was determined with the cylinder method (dry weight of soil sampled in 100 cm^3 cylinders). Soil organic carbon content was measured with an elemental analyser (Thermo Fisher Scientific Flash 2000) on soil sieved at 2 mm and ground at 250 µm.

Hybrid walnut trees (*Juglans nigra* L. x *Juglans regia* L.) spaced 10 m apart were planted in 1995. Cereal and legume crops were grown with conventional practices between the tree rows until 2010 (mostly wheat and rapeseed, with some fallow periods). Since then, the plot has been converted to organic farming.

The experiment was set up in 2015, after a year of sunflower cultivation with reduced tillage and no fertilizer, followed by a cover crop. The experimental plot was divided into three shade treatments of 750 m^2 each (Fig. 1a). In the strong shade treatment (shade ++), trees were only lightly pruned as usually done in agroforestry systems (mean diameter at breast height of 0.37 ± 0.07 m in 2017). In the light shade treatment (shade +), trees were strongly pruned to obtain pollarded trees (mean diameter at breast height of 0.33 ± 0.08 m in 2017).

151   Pruning took place in February 2015 and again in February 2018. In the last treatment, no tree

152   was present (no shade), as trees initially present had been removed in 2011. Each treatment

153   consists of three 8 m-wide and 25 m-long crop alleys and their adjacent herbaceous strips

154   (HS), planted with trees in treatments shade ++ and shade + (Fig. 1a). HS are 2 m wide,

155   covered with spontaneous vegetation and mowed about twice a year in each treatment

156   (characteristics of the vegetation are detailed in Table 1 and Table S1 in the Supplementary

157   materials). Each crop alley is made up of 5 mulched raised beds (with rameal chipped wood

158   on 2 to 3 cm width or hay on 7 cm width alternating each year), about 90 cm wide and 20 cm

159   high, parallel to the HS (Fig. 1b). In each shade treatment, the three crop alleys are managed

160   as a rotation of 3 vegetable crops (carrots, tomatoes and lettuces), meaning that in a given

161   year only one crop alley in each shade treatment is planted with tomatoes. The order of

162   rotation of the different vegetables between the different crop alleys is the same for all shade

163   treatments. Soil was rotary tilled and beds formed at the beginning of the experiment in 2015,

164   and then soil was always rotary tilled after carrot production. After tomatoes or lettuces, beds

165   were only consolidated with a hoe. Fertilization was similar for all productions, with the

166   addition of ricinus oil cakes in 2015 (250 kg ha^{-1}), and 2016 (400 kg ha^{-1}), and of poultry

167   manure in 2017 and 2018 (560 kg ha^{-1} each year). No pesticides were used except in 2018

168   when slug pellets were applied (Ferramol at 0.08 kg ha^{-1}). Crops are drip irrigated during

169   summer.

170

171   **2.2. Sampling design and plot management during sampling**

172   Macrofauna samplings took place in April (16th to 23rd) i.e. spring, June (18th to 22nd) i.e.

173   early summer and November (14th to 21st) i.e. late autumn of 2018 to take into account

174   seasonal variability. Rainfall during the year of the experiment was of 166 mm (January), 59

175   mm (February), 72 mm (March), 90 mm (April, first sampling), 101 mm (May), 4 mm (June,

176   second sampling), 20 mm (July), 56 mm (August), 19 mm (September), 257 mm (October)

177   and 226 mm (November, last sampling, Bellegarde weather station, monitoring data from the

178   INRAE CLIMATIK platform managed by the AgroClim laboratory of Avignon, Francedata).

179   No sampling was carried out during late summer because the rainfall was very low, thus

180   limiting the chance of finding soil macrofauna. Only the crop alleys planted with tomatoes

181   were sampled in order to ensure homogeneous management practices. In these crop alleys,

182   raised beds were rotary tilled in March and covered with a 2 to 3 cm-thick layer of ramial

183   chipped wood obtained from walnut prunings. Different varieties of tomato plants previously

184   grown in the nursery were planted a week after the first sampling (1.6 plants per m²). Each

185   crop alley received two top dressings of poultry manure in March and in April after tomato

186   plantation, for a total of 560 kg ha^{-1}. Tomato plants were drip-irrigated from June (before our

187   sampling date) to mid-September. The previous crops in these crop alleys were lettuces

188   (2015), tomatoes (2016) and carrots (2017). HS were mowed after the June sampling date and

189   at the end of summer. In tomato beds, manual weeding was regularly performed until the end

190   of September, when tomato harvesting stopped.

191   Sampling was designed in order to investigate the effect of the distance from the HS in

192   different shading conditions. Macroinvertebrates were sampled along transects perpendicular

193   to the HS with three distances from the HS (0 m i.e. in the middle of the HS, 1 m from the HS

194   border i.e. in the closest tomato bed to the HS, and 2.5 m from the HS border i.e. in the next

195   tomato bed) (Fig. 1b). For each shade treatment (shade++, shade + and no shade) and at each

196   sampling season, each of the three distances from the HS was sampled eight times (eight

197   transects were made), meaning 24 samples were taken for each shade treatment for a given

198   season (72 samples per season all treatments combined, thus 216 samples in total). All

199   samples were taken in the same crop alley for a given shade treatment as only one crop alley

200   was planted with tomatoes in each shade treatment, and as no replication of the shade

treatments was available on this experimental plot. In the treatments with trees (shade ++ and shade +), four of these transects started at an HS sampling position located 1m from the nearest tree, and four others started at an HS sampling position located 5m from the nearest tree (Fig. 1b).

2.3. Macroinvertebrate sampling

At each sampling point, macroinvertebrates were hand sorted from soil blocks of 25*25*25 cm (adapted from Anderson and Ingram, 1993). Earthworms were kept alive in water, and then counted and determined at species level under laboratory conditions, and classified into the three main ecological categories anecic, endogeic and epigeic following Bouché (1977). Other macroinvertebrates were kept in ethanol, and then counted and determined under laboratory conditions. Chilopoda were determined at order level, while insects and Diplopoda were determined at family level when possible. Count values obtained were expressed as densities (individuals per m^2). As ants presented a wide variation in their densities (due to their properties as social insects), ant density was separated in three density classes (no individuals found, between one and 240 individuals per m^2, and more than 240 individuals per m^2). This classification was chosen to make a distinction between no ants present, few ants presents (240 individuals per m^2 refers to 15 individuals found in a soil block, abundance after which ant counts always reached high numbers of individuals which could not be counted precisely), and probable ant hill. A feeding group was attributed to each category of macrofauna when possible. Diplopoda, Isopoda and larvae of Diptera were classified as litter-feeders. Araneae, Chilopoda, Staphylinidae and larvae of Carabidae, Staphylinidae and Cantharidae were classified as animal-feeders. Elateridae, Curculionidae and larvae of Lepidoptera, Elateridae, Curculionidae and Melolonthinae were classified as plant-feeders, although they consume different parts of the plants. Anecic earthworms were classified at 50

226 % as soil-feeders, and at 50 % as litter-feeders based on anecic earthworm gut analyses by

227 Bernier (1998), while endogeic earthworms were all classified as soil-feeders and epigeic

228 earthworms were all classified as litter-feeders.

229

230 **2.4. Statistical analyses**

231 To investigate the effect of the distance from the HS within the different seasons and shade

232 treatments, macroinvertebrate density (grouped in taxonomic and feeding groups, as well as

233 ecological categories anecic, endogeic and epigeic for earthworms) was analyzed using

234 generalized linear models fitted by quasi Poisson law (with a log link function). These

235 analyses were not run for each earthworm species individually, as most species did not

236 account for enough individuals. Distance from the HS (0, 1 or 2.5 m), shade treatment (shade

237 ++, shade + or no shade) and season (April, June or November) and their interactions were

238 used as fixed factors. Significant effects were determined by type II F tests and followed by

239 pairwise comparisons (Tukey). Significance level was set to 5 %. As no significant difference

240 was found for macrofauna density between transects distant 1 m and 5 m from the tree in the

241 shade treatments with trees (shade ++ and shade +), all transects were treated as simple

242 replicates, whatever their distance from the tree (data not shown). Analyses were run using R

243 statistical software (version 3.6.2) (R Core Team, 2019) with packages *lme4* (Bates et al.,

244 2015), *car* (Fox and Weisberg, 2019) and *emmeans* (Lenth, 2019). Ant density classes were

245 analyzed with a cumulative link model, using the *ordinal* R package (Christensen, 2019).

246 Species richness (i.e. number of different species) and the Shannon diversity index were

247 calculated for earthworms at species level (*vegan* package in R, Oksanen et al., 2019), and

248 analyzed using generalized linear models (species richness) or analyses of variance (Shannon

249 index) with the same fixed factors season, distance from the HS and shade treatment.

250 Variations in macrofauna community composition between distances from the HS and shade

251 treatments were evaluated with a principal component analysis using densities in taxonomic

252 groups at order or family level when possible (packages *FactoMineR* and *ggplot2* in R) (Lê et

253 al., 2008; Wickham, 2016) for each season. Groups with too few individuals (median equal to

254 0) were excluded from the analysis. Earthworms were separated into the ecological groups

255 anecic, epigeic and endogeic to account for their different behaviors. In order to account for

256 large differences in density between taxonomic groups, data were standardized to the

257 maximal density measured for each taxonomic group (maximal margin method of function

258 decostand, *vegan* package in R, Oksanen et al., 2019).

259

260 **3. Results**

261

262   **3.1. Macrofauna community composition**

263 Identified macrofauna were mostly earthworms (17 ± 12 % standard deviation, 32 ± 17 % and

264 29 ± 15 % of macrofauna individuals in the no shade, shade + and shade ++ treatments

265 respectively), Chilopoda (9 ± 9 %, 10 ± 10 % and 20 ± 16 % respectively), Diplopoda (21 ±

266 12 %, 13 ± 10 % and 4 ± 6 % respectively) and Coleoptera (26 ± 13 %, 17 ± 16 % and 21 ±

267 18 % respectively). Formicidae represented up to an estimated 82 % of all individuals in some

268 samples (while their mean contribution was estimated to 12 ± 19 %, 10 ± 16 % and 7 ± 14 %

269 in the no shade, shade + and shade ++ treatments respectively) (Table S2 in the

270 Supplementary materials).

271 The principal component analysis based on macrofauna density in the different taxonomic

272 groups shows variable patterns with the season. In April and June, the principal component

273 analysis mostly isolates sampling points of the HS from those of the tomato beds along the

274 first axis (representing 23 % of the variance in April and 25 % of the variance in June) (Fig.

11

275   2a and Fig. 2b). In April, it is explained by more epigeic and anecic earthworms, Julidae and

276   Isopoda in the HS than in the tomato beds. In June, it is explained by more epigeic

277   earthworms, Julidae and Staphylinidae in the HS than in the tomato beds. The separation of

278   sampling points from the different shade treatments is much less clear, and takes place along

279   axis 2, which explains a lower part of the variance (16% in April, 15 % in June) (Fig. 2a and

280   Fig. 2b). In the opposite, the separation between the HS and the tomato beds is not clear in

281   November, where it is driven by component 2 (16 % of the variance, Fig. 2c), while the first

282   component (22 % of the variance) clearly isolates the shade treatments from one another,

283   especially the no shade treatment from shade ++. There were particularly more Polydesmidae

284   and Elateridae larvae in the no shade treatment compared to other treatments, while the shade

285   ++ treatment is additionally characterized by lower densities of endogeic earthworms and of

286   Julidae compared to other treatments. The tomato beds at 1 m or 2.5 m from the HS could not

287   be separated at any season.

288

289   **3.2. Macrofauna density in taxonomic groups**

290   The response of macroinvertebrate density to the distance from the HS was similar in all

291   shade treatments (Table S3 in the Supplementary materials, significant interaction between

292   distance from the HS and shade treatment for Diplopoda only, and probably due to large

293   differences in densities between shade treatments, see Fig. S1 in the Supplementary

294   materials). However, macrofauna density was different between shade treatments, with

295   especially lower densities in shade ++ compared to other treatments for most taxonomic

296   groups (except Chilopoda, cf Table S2 in the Supplementary materials).

297   The effect of the distance from the HS on macroinvertebrate density varies with the

298   taxonomic group and the season, as shown in Table 2 and Table S3 in the Supplementary

299   materials (significant effect of the interaction between the distance from the HS and the

season for Lumbricidae, Diplopoda, Isopoda, Chilopoda and adult Coleoptera). At all seasons, earthworms presented significantly higher densities in the HS compared to the tomato beds (1 and 2.5 m from the HS), and ants presented higher densities in the HS compared to 2.5 m from the HS (Table 2). Diplopoda presented higher densities in the HS than 2.5 m from the HS in April, and higher densities in the HS compared to all tomato beds (1 and 2.5 m from the HS) in June, but not in November. Isopoda presented higher densities in the HS than in the tomato beds (1 and 2.5 m from the HS) in April only. Adult Coleoptera presented higher densities in the HS compared to 1 m from the HS in April, and compared to both 1 and 2.5 m from the HS in June, while no difference was found in November. In the opposite, Chilopoda presented higher densities at 1 m from the HS than in the HS in November, while no difference was found at other seasons. Dermaptera and Coleoptera larvae did not vary significantly with the distance from the HS at any season (no significant effect of the interaction between distance from the HS and season or of the distance from the HS alone, Table S3 in the Supplementary materials). No significant difference between 1 and 2.5 m from the HS was found for any season or taxonomic group. The attenuation of differences in macrofauna density between the HS and the tomato beds in November is due to a significant increase in macroinvertebrate density at this season compared to April and June in the tomato beds but not in the HS for earthworms, Diplopoda, Isopoda and Chilopoda (Table 2).

3.3. Earthworm diversity

Seven species of earthworms were found in all shade treatments (Table 3). Earthworm diversity varied with the distance from the HS (Table 3). Species richness was higher in the HS than both 1 and 2.5 m from the HS in April and June, and than 2.5 m from the HS in November. The Shannon index presented a significant decreasing gradient from the HS to 2.5 m from the HS for all seasons.

The response of earthworm density to the distance from the HS varies with their ecological category (Fig. 3). Anecic earthworms presented higher densities in the HS compared to 1 and 2.5 m from the HS at all seasons, especially due to *Aporrectodea nocturna* (Table 3). Endogeic earthworms presented higher densities in the HS compared to 1 and 2.5 m from the HS in April and June, but only compared to 2.5 m from the HS in November, especially due to *Allolobophora chlorotica* and to *Aporrectodea rosea*. Epigeic earthworms (all of *Lumbricus castaneus* species) presented higher densities in the HS compared to 2.5 m from the HS in April, and compared to both 1 and 2.5 m from the HS in June, but no significant difference was found anymore in November for epigeic density between distances from the HS, because of a large increase in epigeic density in the tomato beds between June and November.

3.4. Feeding groups

The distribution of macrofauna in the plot varies according to the feeding group considered (Fig. 4). Litter-feeders presented higher densities in the HS compared to tomato beds in April and June, but not in November (Fig. 4a). This result was mostly driven by litter-feeding earthworms and Diplopoda. Soil-feeding earthworms presented higher densities in the HS than in the tomato beds at all seasons (Fig. 4b). Animal-feeders presented higher densities in the HS than in the tomato beds in April only (Fig. 4d). Animal-feeders were mostly Chilopoda, especially in June and November. Plant-feeders presented a different pattern from other groups, with a significantly lower density in the HS compared to the tomato beds in April (Fig. 4c). Plant-feeders were mostly Coleoptera larvae, of which a huge proportion consisted of Elateridae (85 % ± 27 %).

4. Discussion

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4.1. Constant patterns of macrofauna distribution throughout seasons

Throughout all seasons, HS macrofauna communities were characterized by higher densities of litter-feeders but similar Dermaptera and Chilopoda densities, compared to communities found in the tomato beds.

Earthworms presented higher densities in the HS compared to both tomato beds at all seasons, especially due to anecic earthworms. Higher earthworm densities in HS than crop alleys were also found by Cardinael et al. (2019) in France and Rivest et al. (2020) in Québec for different alley cropping plots mostly cultivated with cereals and legume crops, and by Crittenden et al. (2015) in the Netherlands for herbaceous margins of fields cultivated with cereals and tuber crops. Anecic earthworms live in permanent burrows in the soil. HS were probably a favorable habitat to them as they were not subject to soil perturbation, while anecic earthworms were probably negatively impacted by the repeated destruction of their burrows and the lack of adequate food resources and protection against predators at the soil surface in the tomato beds (Chan, 2001). *Aporrectodea nocturna*, the main anecic earthworm species found in this study, is in particular known to reuse its burrows intensively (Capowiez, 2000). In addition, endogeic earthworms always presented higher densities in the HS than 2.5 m from the HS. Endogeic earthworms were probably strongly impacted by the recent rotary tillage as they live and feed inside the soil. Similarly, ants also presented higher densities in the HS than 2.5 m from the HS at all seasons. Anthills were probably repeatedly destroyed by intensive tillage in the tomato beds. In agreement with us, Boinot et al. (2019) found more activity density of ants in the tree row than in the crop alley in mature alley cropping systems cultivated with cereals in the south of France. In spatially structured systems like the one studied here, spatial differences in soil properties (e.g. higher soil organic carbon concentrations and stocks under tree rows such as often found in agroforestry systems

375   (Cardinael et al., 2017)) could contribute to explaining locally higher densities of earthworms.

376   However in our case, basic soil characteristics, including organic matter content, did not vary

377   according to sampled positions (Table 1), therefore differences in macrofauna communities

378   are more likely related to other changes in habitat characteristics caused by vegetation

379   structure and management practices.

380   Dermaptera and Chilopoda density did not vary between HS and crop alleys at any season.

381   Similarly, Smith et al. (2008b) did not find significant differences in Chilopoda abundance

382   between grass margins and the adjacent wheat crop in England. These organisms are able to

383   move quite fast and could have recolonized tomato beds more rapidly than other soil-dwelling

384   organisms such as earthworms and larva after tillage. While adult Coleoptera have a faster

385   dispersal, particularly flying Staphylinidae, they did not present the same pattern, probably

386   because they use different habitats for living and feeding.

387

388   **4.2. Seasonal variations in macrofauna distribution**

389   Despite a constant general trend, this study reveals some seasonal variations in the differences

390   between HS and tomato bed macrofauna communities, which could be due to climatic

391   variations or to crop management. In April, most soil macrofauna was probably negatively

392   impacted by the recent soil perturbations in the tomato beds. Smith et al. (2008b) indeed

393   found that soil scarification reduces macrofauna density, but that populations recovered

394   within a season after soil scarification. In the present study, these macrofauna categories seem

395   to have recovered in November, or even in June for Isopoda. The recovery in November

396   could have been amplified by the interruption of weeding and of other management, allowing

397   the colonization of tomato beds by herbaceous vegetation, probably providing food and

398   habitat usually not found in crop alleys for soil macrofauna. In addition, November 2018 was

399   a particularly rainy month, which could have favored macrofauna activity. More generally,

400 Mediterranean regions are affected by strong climate variability: spring is not always a

401 favorable season as it can sometimes be too dry, summer is generally very dry and hot, and

402 intense rainfall events can occur in autumn, with variable timing and amplitude. Endogeic

403 macrofauna is very sensitive to such variations in rainfall and temperature, with a limited

404 activity in summer, and as found in this study a potential increase in activity in autumn. The

405 herbaceous strip could act as a refuge in unfavorable seasons, by buffering climatic conditions

406 compared to the exposed crop alley. In addition, some soil macrofauna taxa require different

407 habitats at different periods of the year. In spring, macrofauna is probably found in the places

408 where it overwintered, i.e. probably buffered habitats such as the HS. In summer, macrofauna

409 concerned by aestivation (such as epigeic and anecic earthworms) is probably not present in

410 places submitted to high temperatures and fast desiccation such as the crop alley, with little

411 plant cover of the soil, low bulk density and fragile structure due to rotary tillage. The specific

412 effect of the climatic conditions versus agricultural practices, particularly soil tillage, is

413 however difficult to determine as each agricultural practice is strongly associated with the

414 season.

415 Epigeic earthworms and isopods are usually present at very low densities in tilled soils,

416 probably because of the reduction in adequate shelter and food at the soil surface (Hassall and

417 Tuck, 2007; Pelosi et al., 2009). Higher densities of isopods in tree rows compared to crop

418 alleys have already been found in spring similarly to us in both young and old sites with

419 cereals or tuber crops in Belgium and France (D'Hervilly et al., 2020; Pardon et al., 2019),

420 and isopods have been shown to hibernate in tree rows in mature alley cropping plots in the

421 south of France (Boinot et al., 2019). Epigeic earthworms have also been shown to be mostly

422 present in tree rows in spring in France (Cardinael et al., 2019; D'Hervilly et al., 2020) or in

423 field margins in July (Crittenden et al., 2015) compared to the neighboring crop. In addition,

424 higher densities of Diplopoda were also found in tree rows than in adjacent crops by Pardon et

al. (2019). However, our study demonstrates that Isopoda, epigeic earthworms and Diplopoda

can be present in the crop alley at similar densities to those of the HS in autumn, their

establishment being probably favored in our case by the shelter provided by ramial chipped

wood presence at the soil surface of the tomato beds. Tomato bed irrigation in June probably

further favored early colonization by isopods, limiting the usual adverse conditions of the

summer. Surprisingly, it did not favor epigeic earthworms whose presence was mostly

restricted to the HS in June, although they are known to be positively affected by increases in

soil humidity and litter presence at the soil surface (Bouché, 1977). Temperatures were

probably too high for epigeic earthworm activity in summer, and ramial chipped wood was

probably a more interesting resource for Isopoda than for epigeic earthworms.

Adult Coleoptera were mostly Staphylinid beetles and Carabids in our study. Andersen

(1997), and Smith et al. (2008b) found respectively more Carabid and Staphylinid beetles or

more Coleoptera in field margins than in the cereal crop fields (in Norway and England

respectively), similarly to what we found in April and June. The higher densities of adult

Coleoptera in the HS than in the tomato beds in April and June could be due to additional

prey for the animal-feeders in the HS as usually described for margin habitats (Landis et al.,

2000). It could also be due to their behavior, as most of them feed at night (Martin-Chave et

al., 2019a), and could use the HS as a shelter during the day.

Contrary to our hypothesis, no significant difference was found between distances 1 m and

2.5 m from the HS for macrofauna density at all seasons. However, we only investigated the

HS close vicinity, due to the limited size of our experimental plot (crop alleys were only 8 m

wide), which could have hidden some effects. For example, Smith et al. (2008a) found higher

macrofauna density up to 3 m from field margins compared to the middle of the field in

England. However, while the HS presented higher endogeic earthworm density than both

tomato beds in April and June, endogeic earthworm density was only significantly higher in

450 the HS than 2.5 m from the HS in November, suggesting a positive effect of HS vicinity on

451 endogeic earthworm density in November. A positive effect of tree row proximity on

452 earthworm density has already been suggested by Price and Gordon (1999). A possible

453 explanation is that most macrofauna dispersed from the HS to at least 2.5 m from the HS in

454 the crop alley in November, but that this dispersion was more limited for endogeic

455 earthworms, whose movements are slowed as they are restricted to the soil.

456 Plant-feeding Coleoptera larvae presented higher densities in the tomato beds than in the HS

457 in April, and no differences between distances from the HS at other seasons. These

458 Coleoptera were mostly wireworms (Elateridae), known to be detrimental to crops, which

459 could explain their preference for the crop alley. However, wireworms are usually destroyed

460 directly or indirectly by tillage (Parker and Howard, 2001), which was performed only a

461 month before the first sampling season. Wireworms could have been located deeper in the soil

462 in March during tillage, or soil cover by ramial chipped wood could have limited their

463 predation after exposure at the soil surface. In addition, the use of rotary tillage could have

464 made it easier for them to dig into the loose soil (particularly low bulk density in the crop

465 alley).

466 Overall, the seasonal and spatial patterns observed in this study reflect taxon-specific

467 responses to a complex combination of variations in climatic conditions and management

468 practices in a spatially-structured agricultural system. The well-known impacts of soil

469 perturbation on soil fauna in agricultural systems (e.g. van Capelle et al., 2012) could be

470 partially alleviated by the inclusion of herbaceous strips, allowing the recovery of some

471 macrofauna taxa in crop alleys at certain seasons.

472

473   **4.3. Variations between shade treatments**

474   We expected to find more homogeneous macrofauna densities relative to the distance from

475   the HS in the strong-shade treatment (shade ++) than in the light shade (shade +) and no shade

476   treatments, as we expected that the more homogeneous tree shading and litter inputs would

477   attenuate the habitat differences between HS and raised beds. However, we found no effect of

478   the shade treatment on the spatial distribution of macrofauna within the plot. Shade treatments

479   most likely influenced all distances from the HS homogeneously, as soil macrofauna density

480   and community composition differed between shade treatments at a general level, especially

481   in November. Increased shade has been found to increase soil macrofauna density in tropical

482   systems (Martius et al., 2004), but has rarely been studied in temperate or Mediterranean

483   systems. Pardon et al. (2019) investigated the effect of herbaceous strips with or without trees

484   at field edges in Belgium, and found a higher woodlice density in the presence of trees and no

485   difference for millipedes, while in our study both were positively impacted in the absence of

486   trees (no shade treatment). In our study, higher shade led to a lower macrofauna density for

487   most macrofauna, Chilopoda excepted. This was particularly visible in November, possibly

488   related to a delayed and cumulative impact on populations of the shade that appeared around

489   the end of May. Similarly, Martin Chave et al. (2019b) found a lower activity density of the

490   main Carabidae in the strong shade treatment compared to lighter shade treatments in

491   September in the same experimental plot. The strong shade treatment (shade ++) was shown

492   to buffer temperature variations in this same study (Martin-Chave et al., 2019b). A strong

493   shade could have lowered temperature too much during the day in November, attenuating the

494   positive impact of autumn on macrofauna activity. The soil environment was also probably

495   strongly modulated by the soil cover by tree leaves and the limited herbaceous vegetation

496   biomass observed in November in the strong shade treatment (shade ++). This would have

497   further created buffered microclimate conditions in this treatment, and induced strong

498   differences in litter quality and quantity between shade treatments, which has been shown to

20

499   modulate soil macrofauna abundance and diversity (Loranger-Merciris et al., 2008; Vohland

500   and Schroth, 1999). Interestingly, earthworms and Dermaptera presented higher densities in

501   the light shade treatment (shade +) than in both strong shade (shade ++) and no shade

502   treatments, meaning the observed trends are not only the results of a variation of shade and of

503   the inputs of tree litter. While these results on the shade treatments seem interesting, it is

504   important to note that there was no replication of the shade treatments in this experiment.

505   Observations of the differences between shade treatments are thus limited to hypotheses, and

506   should be the focus of future investigations. However, the fact that the same effect of the

507   distance from the HS was observed for three different shade treatments is relevant as it

508   indicates that uncropped herbaceous strips are a possible reservoir of soil macrofauna in

509   spring and summer, independently of the presence and management of trees on these strips.

510

511   **5. Conclusions**

512

513   Herbaceous strips were shown to present higher macrofauna density and earthworm diversity

514   compared to nearby tomato beds whatever the shade treatment. This finding confirms the

515   important role of non-perturbed herbaceous strips to preserve dense and diverse macrofauna

516   communities across fields, including in alley cropping systems. However, this study reveals

517   the importance of considering seasonal variations when studying soil macrofauna in alley

518   cropping systems. The maximal seasonal activity was found to be in autumn, while most

519   samplings in crops usually take place in spring, and patterns of macrofauna distribution in the

520   plot could be quite different according to the season. The role of the season itself versus of the

521   management practices (especially soil tillage) in these differences is hard to determine as both

522   vary simultaneously, and requires further investigations with limited soil perturbation, such as

523   zero tillage experiments, or the comparison of different crop managements. A possible

524 recolonization of crop alleys by macrofauna coming from the HS via seasonal movements in

525 soil fauna after soil perturbation, as suggested by the results of this study, should be further

526 investigated.

527

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538 **References**

539 Andersen, A., 1997. Densities of overwintering carabids and staphylinids (Col., Carabidae

540   and Staphylinidae) in cereal and grass fields and their boundaries. J. Appl. Entomol. 121,

541   77–80. https://doi.org/10.1111/j.1439-0418.1997.tb01374.x

542 Anderson, J.M., Ingram, J.S.I., 1993. Tropical soil Biology and Fertility, a handbook of

543   methods - second edition. CAB International, Wallingford, UK.

544 Barrios, E., 2007. Soil biota, ecosystem services and land productivity. Ecol. Econ. 64, 269–

545   285. https://doi.org/10.1016/j.ecolecon.2007.03.004

546 Bates, D., Maechler, M., Bolker, B., Walker, S., 2015. Fitting Linear Mixed-Effects Models

547   Using lme4. J. Stat. Softw. 67, 1–48. https://doi.org/10.18637/jss.v067.i01

548 Bender, S.F., Wagg, C., van der Heijden, M.G.A., 2016. An Underground Revolution:

549   Biodiversity and Soil Ecological Engineering for Agricultural Sustainability. Trends

550   Ecol. Evol. 31, 440–452. https://doi.org/10.1016/j.tree.2016.02.016

551   Bernier, N., 1998. Earthworm feeding activity and development of the humus profile. Biol.

552   Fertil. Soils 26, 215–223. https://doi.org/10.1007/s003740050370

553   Boinot, S., Poulmarc'h, J., Mézière, D., Lauri, P.É., Sarthou, J.P., 2019. Distribution of

554   overwintering invertebrates in temperate agroforestry systems: Implications for

555   biodiversity conservation and biological control of crop pests. Agric. Ecosyst. Environ.

556   285, 1–11. https://doi.org/10.1016/j.agee.2019.106630

557   Bossuyt, H., Six, J., Hendrix, P.F., 2005. Protection of soil carbon by microaggregates within

558   earthworm casts. Soil Biol. Biochem. 37, 251–258.

559   https://doi.org/10.1016/j.soilbio.2004.07.035

560   Bouché, M.B., 1977. Stratégies lombriciennes. Ecol. Bull. 25, 122–132.

561   Briones, M.J.I., Morán, P., Posada, D., 2009. Are the sexual, somatic and genetic characters

562   enough to solve nomenclatural problems in lumbricid taxonomy? Soil Biol. Biochem.

563   41, 2257–2271. https://doi.org/10.1016/j.soilbio.2009.07.008

564   Capowiez, Y., 2000. Differences in burrowing behaviour and spatial interaction between the

565   two earthworm species *Aporrectodea nocturna* and *Allolobophora chlorotica*. Biol.

566   Fertil. Soils 30, 341–346. https://doi.org/10.1007/s003740050013

567   Cardinael, R., Hoeffner, K., Chenu, C., Chevallier, T., Béral, C., Dewisme, A., Cluzeau, D.,

568   2019. Spatial variation of earthworm communities and soil organic carbon in temperate

569   agroforestry. Biol. Fertil. Soils 55, 171–183. https://doi.org/10.1007/s00374-018-1332-3

570   Chan, K.Y., 2001. An overview of some tillage impacts on earthworm population abundance

571   and diversity - implications for functioning in soils. Soil Tillage Res. 57, 179–191.

572   https://doi.org/10.1016/S0167-1987(00)00173-2

573   Christensen, R.H.B., 2019. ordinal - Regression Models for Ordinal Data. R package version

574   2019. 4-25.

575   Crittenden, S.J., Huerta, E., de Goede, R.G.M., Pulleman, M.M., 2015. Earthworm

576   assemblages as affected by field margin strips and tillage intensity: An on-farm

577   approach. Eur. J. Soil Biol. 66, 49–56. https://doi.org/10.1016/j.ejsobi.2014.11.007

578   D'Hervilly, C., Marsden, C., Hedde, M., Bertrand, I., 2020. Sown understory vegetation strips

579   impact soil chemical fertility, associated microorganisms and macro-invertebrates in two

580   temperate alley cropping systems. Agrofor. Syst. 94, 1851–1864.

581   https://doi.org/10.1007/s10457-020-00501-w

582   David, J.F., 2014. The role of litter-feeding macroarthropods in decomposition processes: A

583   reappraisal of common views. Soil Biol. Biochem. 76, 109–118.

584   https://doi.org/10.1016/j.soilbio.2014.05.009

585   Filser, J., Faber, J.H., Tiunov, A. V., Brussaard, L., Frouz, J., De Deyn, G., Uvarov, A. V.,

586   Berg, M.P., Lavelle, P., Loreau, M., Wall, D.H., Querner, P., Eijsackers, H., Jiménez,

587   J.J., 2016. Soil fauna: Key to new carbon models. Soil 2, 565–582.

588   https://doi.org/10.5194/soil-2-565-2016

589   Fox, J., Weisberg, S., 2019. An R Companion to Applied Regression, Third. ed. Sage,

590   Thousand Oaks CA.

591   Frouz, J., 2018. Effects of soil macro- and mesofauna on litter decomposition and soil organic

592   matter stabilization. Geoderma 332, 161–172.

593   https://doi.org/10.1016/j.geoderma.2017.08.039

594   Geiger, F., Wäckers, F.L., Bianchi, F.J.J.A., 2009. Hibernation of predatory arthropods in

595   semi-natural habitats. BioControl 54, 529–535. https://doi.org/10.1007/s10526-008-

596   9206-5

597   Habel, J.C., Samways, M.J., Schmitt, T., 2019. Mitigating the precipitous decline of terrestrial

598   European insects: Requirements for a new strategy. Biodivers. Conserv. 28, 1343–1360.

599   https://doi.org/10.1007/s10531-019-01741-8

600 Hassall, M., Tuck, J.M., 2007. Sheltering behavior of terrestrial isopods in grasslands.

601   Invertebr. Biol. 126, 46–56. https://doi.org/10.1111/j.1744-7410.2007.00075.x

602 Holland, J.M., Douma, J.C., Crowley, L., James, L., Kor, L., Stevenson, D.R.W., Smith,

603   B.M., 2017. Semi-natural habitats support biological control, pollination and soil

604   conservation in Europe. A review. Agron. Sustain. Dev. 37.

605   https://doi.org/10.1007/s13593-017-0434-x

606 Jose, S., 2009. Agroforestry for ecosystem services and environmental benefits: an overview.

607   Agrofor. Syst. 76, 1–10. https://doi.org/10.1007/s10457-009-9229-7

608 Landis, D.A., Wratten, S.D., Gurr, G.M., 2000. Habitat management to conserve natural

609   enemies of arthropod pests in agriculture. Annu. Rev. Entomol. 45, 175–201.

610 Lavelle, P., Decaëns, T., Aubert, M., Barot, S., Blouin, M., Bureau, F., Margerie, P., Mora, P.,

611   Rossi, J.P., 2006. Soil invertebrates and ecosystem services. Eur. J. Soil Biol. 42.

612   https://doi.org/10.1016/j.ejsobi.2006.10.002

613 Lavelle, P., Pashanasi, B., Charpentier, F., Gilot, C., Rossi, J.-P., Derouard, L., André, J.,

614   Ponge, J.-F., Bernier, N., 1998. Large-scale effects of earthworms on soil organic matter

615   and nutrient dynamics To cite this version : Large-Scale Effects of Earthworms on Soil

616   Organic Matter and Nutrient Dynamics, in: Edwards, C.A. (Ed.), Earthworm Ecology.

617   pp. 103–122.

618 Lê, S., Josse, J., Husson, F., 2008. FactoMineR: An R Package for Multivariate Analysis. J.

619   Stat. Softw. 25, 1–18. https://doi.org/10.18637/jss.v025.i01

620 Lenth, R., 2019. emmeans: Estimated Marginal Means, aka Least-Squares Means. R package

621   version 1.4.1.

622 Loranger-Merciris, G., Imbert, D., Bernhard-Reversat, F., Lavelle, P., Ponge, J.F., 2008.

623   Litter N-content influences soil millipede abundance, species richness and feeding

624   preferences in a semi-evergreen dry forest of Guadeloupe (Lesser Antilles). Biol. Fertil.

625   Soils 45, 93–98. https://doi.org/10.1007/s00374-008-0321-3

626   Marsden, C., Martin-Chave, A., Cortet, J., Hedde, M., Capowiez, Y., 2020. How agroforestry

627   systems influence soil fauna and their functions - a review. Plant Soil 453, 29–44.

628   https://doi.org/10.1007/s11104-019-04322-4

629   Martin-Chave, A., Béral, C., Capowiez, Y., 2019a. Agroforestry has an impact on nocturnal

630   predation by ground beetles and Opiliones in a temperate organic alley cropping system.

631   Biol. Control 129, 128–135. https://doi.org/10.1016/j.biocontrol.2018.10.009

632   Martin-Chave, A., Béral, C., Mazzia, C., Capowiez, Y., 2019b. Agroforestry impacts the

633   seasonal and diurnal activity of dominant predatory arthropods in organic vegetable

634   crops. Agrofor. Syst. 93, 2067–2083. https://doi.org/10.1007/s10457-018-0309-4

635   Martius, C., Höfer, H., Garcia, M.V.B., Römbke, J., Förster, B., Hanagarth, W., 2004.

636   Microclimate in agroforestry systems in central Amazonia: Does canopy closure matter

637   to soil organisms? Agrofor. Syst. 60, 291–304.

638   https://doi.org/10.1023/B:AGFO.0000024419.20709.6c

639   McLaughlin, A., Mineau, P., 1995. The impact of agricultural practices on biodiversity.

640   Agric. Ecosyst. Environ. 55, 201–212. https://doi.org/10.1016/0167-8809(95)00609-V

641   Oksanen, J., Blanchet, F.G., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin,

642   P.R., O'Hara, R.B., Simpson, G.L., Solymos, P., Stevens, M.H.H., Szoecs, E., Wagner,

643   H., 2019. Vegan: ecological diversity. R Package Version 2.4-4.

644   https://doi.org/10.1029/2006JF000545

645   Pardon, P., Mertens, J., Reubens, B., Reheul, D., Coussement, T., Elsen, A., Nelissen, V.,

646   Verheyen, K., 2020. *Juglans regia* (walnut) in temperate arable agroforestry systems:

647   effects on soil characteristics, arthropod diversity and crop yield. Renew. Agric. Food

648   Syst. 35, 533–549. https://doi.org/10.1017/S1742170519000176

649   Pardon, P., Reheul, D., Mertens, J., Reubens, B., De Frenne, P., De Smedt, P., Proesmans, W.,

650   Van Vooren, L., Verheyen, K., 2019. Gradients in abundance and diversity of ground

651   dwelling arthropods as a function of distance to tree rows in temperate arable

652   agroforestry systems. Agric. Ecosyst. Environ. 270–271, 114–128.

653   https://doi.org/10.1016/j.agee.2018.10.017

654   Parker, W.E., Howard, J.J., 2001. The biology and management of wireworms (*Agriotes* spp.)

655   on potato with particular reference to the U.K. Agric. For. Entomol. 3, 85–98.

656   https://doi.org/10.1046/j.1461-9563.2001.00094.x

657   Pelosi, C., Bertrand, M., Roger-Estrade, J., 2009. Earthworm community in conventional,

658   organic and direct seeding with living mulch cropping systems. Agron. Sustain. Dev. 29,

659   287–295. https://doi.org/10.1051/agro/2008069

660   Price, G.W., Gordon, A.M., 1999. Spatial and temporal distribution of earthworms in a

661   temperate intercropping system in southern Ontario, Canada. Agrofor. Syst. 44, 141–

662   149. https://doi.org/10.1023/A:1006213603150

663   R Core Team, 2019. R: A language and environment for statistical computing.

664   Rivest, M., Whalen, J.K., Rivest, D., 2020. Variation of soil microbial and earthworm

665   communities along an agricultural transect with tree windbreak. Agrofor. Syst. 94, 1639–

666   1649. https://doi.org/10.1007/s10457-019-00476-3

667   Smith, J., Potts, S., Eggleton, P., 2008a. The value of sown grass margins for enhancing soil

668   macrofaunal biodiversity in arable systems. Agric. Ecosyst. Environ. 127, 119–125.

669   https://doi.org/10.1016/j.agee.2008.03.008

670   Smith, J., Potts, S.G., Woodcock, B.A., Eggleton, P., 2008b. Can arable field margins be

671   managed to enhance their biodiversity , conservation and functional value for soil

672   macrofauna ? J. Appl. Ecol. 45, 269–278. https://doi.org/10.1111/j.1365-

673   2664.2007.01433.x

674   Udawatta, R.P., Rankoth, L.M., Jose, S., 2019. Agroforestry and biodiversity. Sustainability

675   11, 2879. https://doi.org/10.3390/su11102879

676   van Capelle, C., Schrader, S., Brunotte, J., 2012. Tillage-induced changes in the functional

677   diversity of soil biota - A review with a focus on German data. Eur. J. Soil Biol. 50, 165–181.

678   https://doi.org/10.1016/j.ejsobi.2012.02.005

679   Vohland, K., Schroth, G., 1999. Distribution patterns of the litter macrofauna in agroforestry

680   and monoculture plantations in central Amazonia as affected by plant species and

681   management. Appl. Soil Ecol. 13, 57–68. https://doi.org/10.1016/S0929-1393(99)00021-

682   9

683   Wickham, H., 2016. ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag, New

684   York.

685   Wolters, V., 2000. Invertebrate control of soil organic matter stability. Biol. Fertil. Soils 31,

686   1–19. https://doi.org/10.1007/s003740050618

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Figure captions

699 **Fig. 1.** Experimental plot (a) and sampling design (b). This sampling design was repeated for

700 each shade treatment (no shade = no trees, shade + = strongly-pruned trees, shade ++ =

701 lightly-pruned trees), and for each season (April, June and November of 2018). HS stands for

702 herbaceous strip.

703

704 **Fig. 2.** Graphical representations of principal component analyses computed with

705 macroinvertebrates at order or family level (when possible), for (a) April data, (b) June data

706 and (c) November data. Earthworms were additionally separated by ecological categories.

707 Individuals are identified by their distance from the herbaceous strip in the representations on

708 the left, with red circles for 0 m, orange triangles for 1 m and yellow squares for 2.5 m.

709 Individuals are identified by their shade treatment in the representations on the right with dark

710 blue circles for the no shade treatment, light blue squares for shade + treatment, and medium

711 blue triangles for shade ++ treatment. Ellipses represent confidence intervals at 95 % around

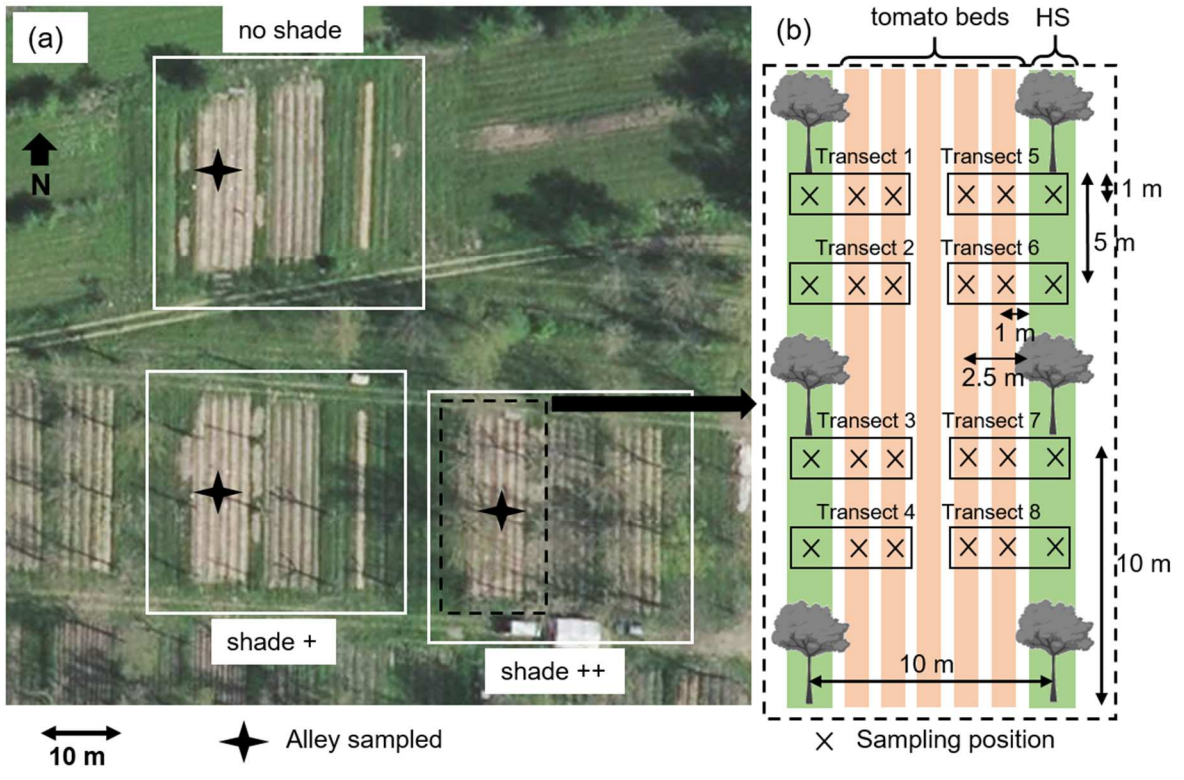712 barycenters. Only variables with a $\cos^2 > 0.5$ are represented.
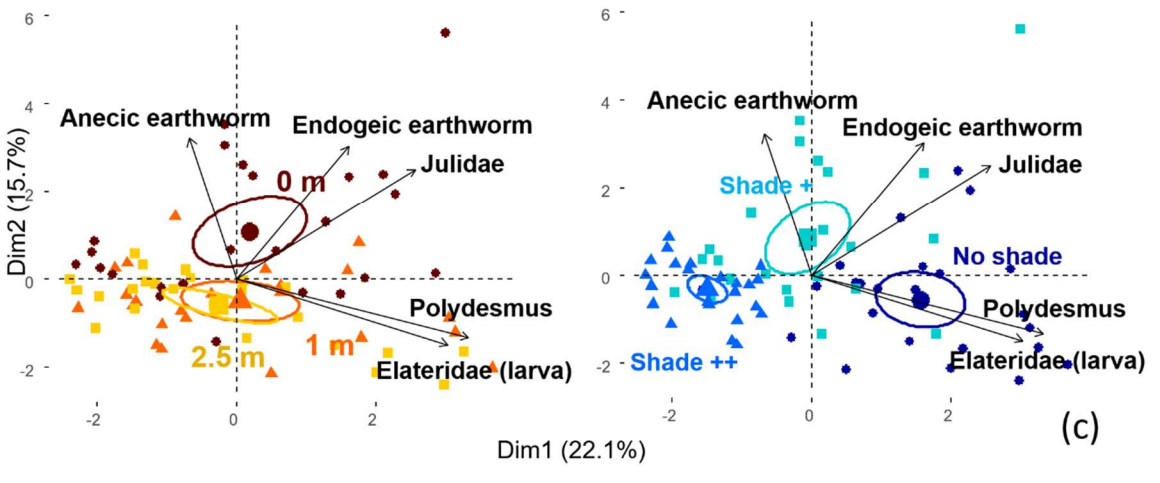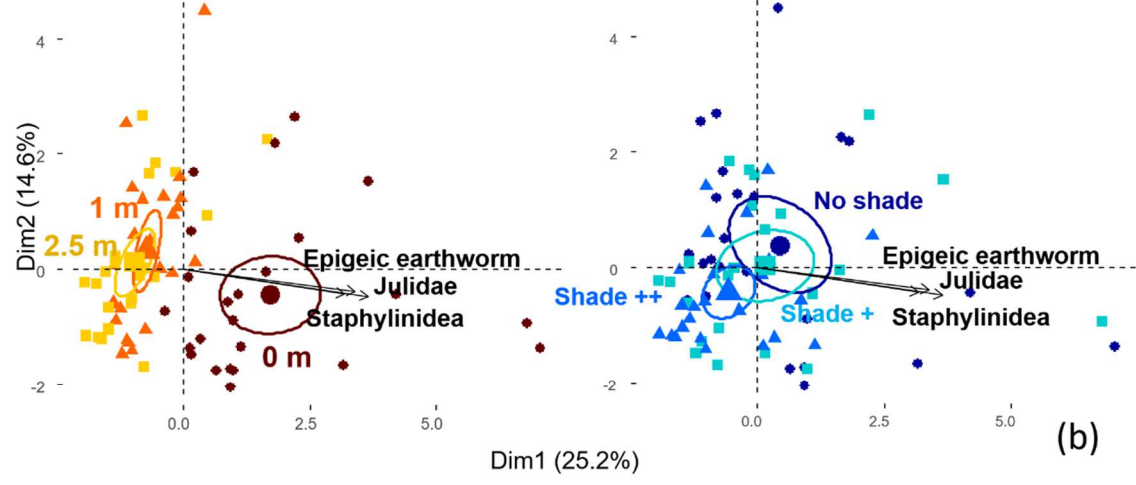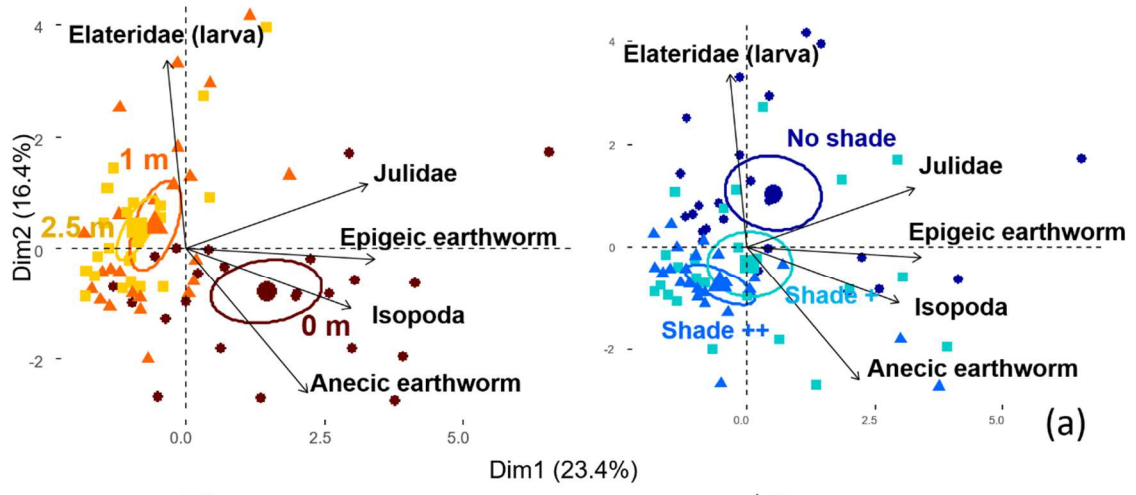
713

714 **Fig. 3.** Mean values of earthworm density for each earthworm ecological category at each

715 season and each distance from the herbaceous strip (HS). Different letters indicate significant

716 differences between distances from the HS for a given earthworm category and season (Tukey

717 test, p-value < 0.05) after generalized linear models fitted with a quasi-Poisson law. Error bars

718 are the standard error of the mean for each earthworm ecological category.

719

720 **Fig. 4.** Mean values of densities of litter-feeders (a), soil-feeders (b), plant feeders (c) and

721 animal-feeders (d) at each season and distance from the herbaceous strip (HS). Different

722 letters indicate significant differences between distances from the HS for the given season and

723   feeding group (Tukey test, p-value < 0.05) based on generalized linear models fitted with a

724   quasi-Poisson law. Error bars are the standard error of the mean for each category of

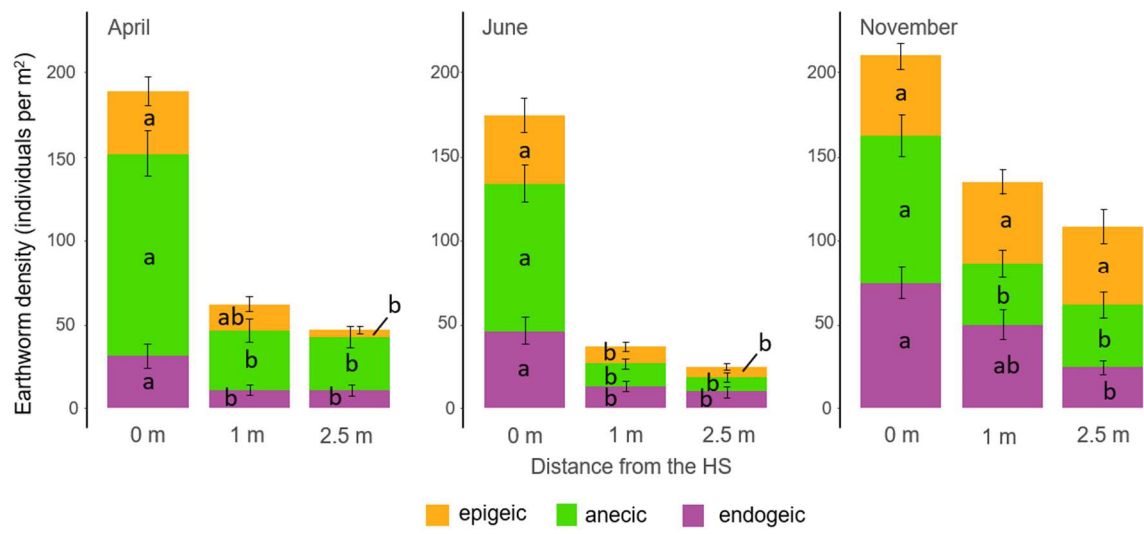725   macrofauna included in the feeding groups.

(a) no shade; shade +; shade ++; N; 10 m; Alley sampled

(b) tomato beds; HS; Transect 1; Transect 5; Transect 2; Transect 6; Transect 3; Transect 7; Transect 4; Transect 8; 1 m; 5 m; 1 m; 2.5 m; 10 m; 10 m; X Sampling position
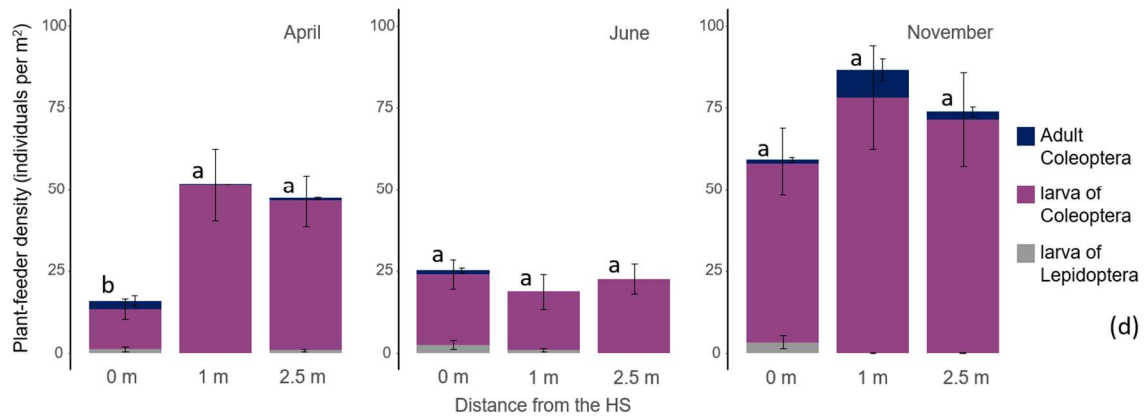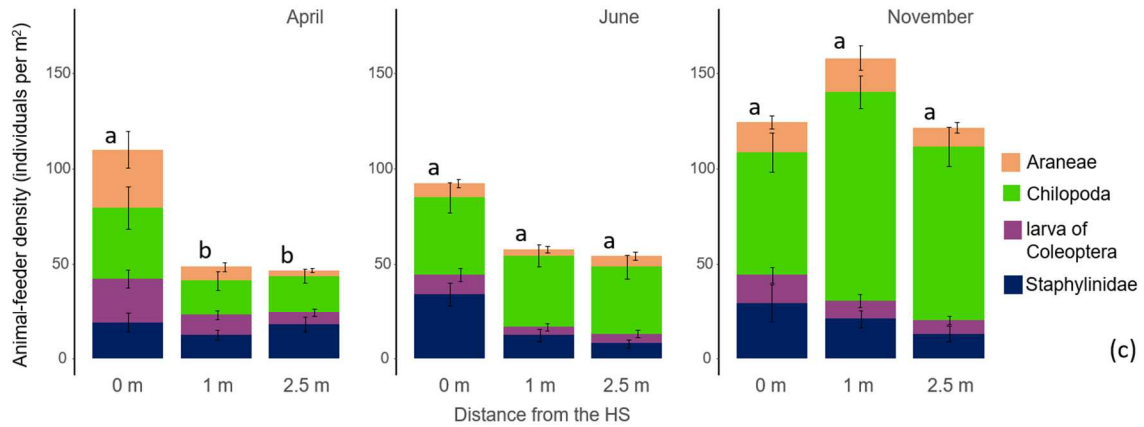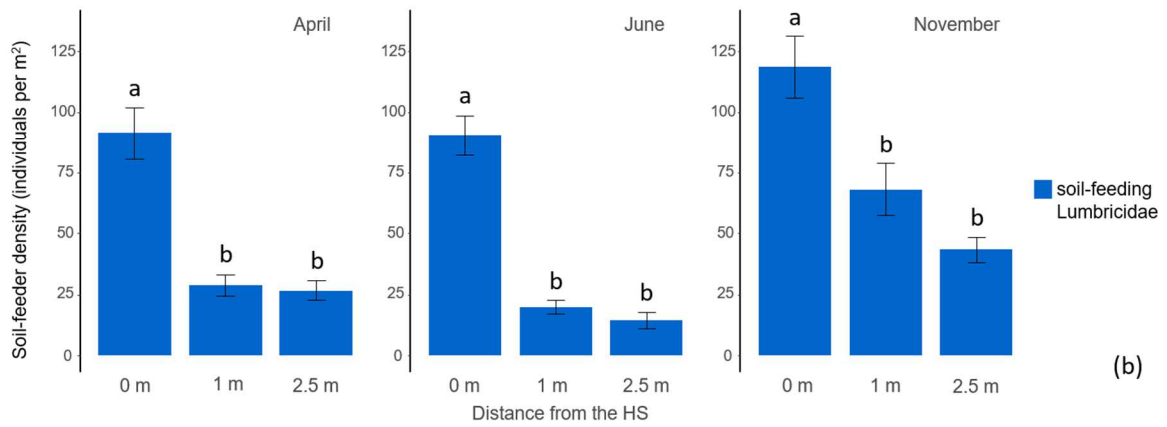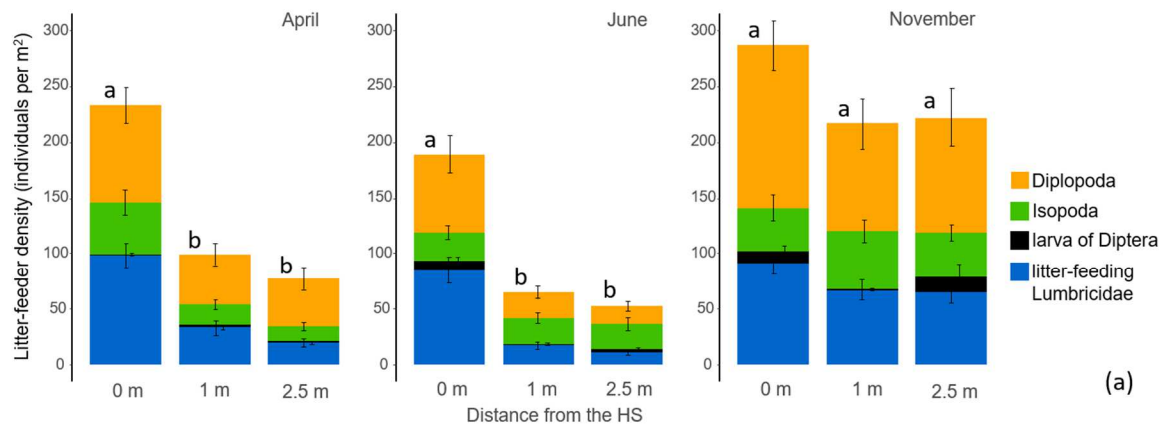
(a)

(b)

(c)

(d)

Table 1 Vegetation and soil properties (of the 0-25 cm soil layer) in each shade treatment, at different distances from the herbaceous strip (HS) (mean ± standard deviation). Vegetation was collected in squares of 50*50 cm and oven-dried at 40 °C during a week for dry biomass determination.

	No shade			Shade +			Shade ++		
Distance from the HS	0 m	1 m	2.5 m	0 m	1 m	2.5 m	0 m	1 m	2.5 m
Texture (clay /	103 ± 2 /	105 ± 2 /	104 ± 4 /	114 ± 3 /	112 ± 6 /	110 ± 7 /	112 ± 4 /	110 ± 5 /	107 ± 2 /
silt /	410 ± 3 /	410 ± 11 /	405 ± 12 /	383 ± 15 /	379 ± 18 /	377 ± 22 /	386 ± 7 /	379 ± 14 /	373 ± 7 /
sand, g kg^{-1})	471 ± 6	469 ± 8	475 ± 8	490 ± 11	496 ± 24	500 ± 30	484 ± 7	491 ± 18	502 ± 9
Bulk density (g cm^{-3})	1.26 ± 0.08	1.07 ± 0.04	1.11 ± 0.04	1.27 ± 0.09	1.10 ± 0.05	1.13 ± 0.05	1.32 ± 0.06	1.11 ± 0.07	1.17 ± 0.04
pH (in water)	8.2 ± 0.0	8.2 ± 0.0	8.2 ± 0.1	8.1 ± 0.1	8.2 ± 0.0	8.2 ± 0.0	8.1 ± 0.1	8.3 ± 0.1	8.2 ± 0.0
Soil organic carbon (%)	1.5 ± 0.2	1.7 ± 0.3	1.6 ± 0.2	1.9 ± 0.1	2.0 ± 0.2	1.7 ± 0.1	2.0 ± 0.2	1.9 ± 0.1	1.8 ± 0.2
Gravimetric water content	27 ±3 /	30 ± 3 /	28 ± 1 /	28 ± 1 /	29 ± 1 /	27 ± 0 /	24 ± 4 /	26 ± 1 /	25 ± 2 /
(April/ June/	24 ± 2 /	32 ± 4 /	30 ± 3 /	23 ± 3 /	29 ± 3 /	26 ± 3 /	25 ± 2 /	26 ± 2 /	24 ± 1 /
November, %)	29 ± 2	33 ± 3	32 ± 2	27 ± 1	28 ± 1	27 ± 1	25 ± 1	26 ± 1	25 ± 1
Vegetation biomass (April /	120 ± 94 /	2 ± 6 /	2 ± 4 /	42 ± 37 /	0 ± 0 /	0 ± 0 /	64 ± 62 /	0 ± 0 /	0 ± 0 /
June /	629 ± 304 /	77 ± 90 /	62 ± 56 /	426 ± 75 /	191 ± 282 /	79 ± 72 /	404 ± 220 /	14 ± 20 /	24 ± 19 /
November, g m^{-2})	391 ± 243	125 ± 150	27 ± 30	233 ± 135	22 ± 27	13 ± 16	65 ± 31	6 ± 9	1 ± 3

Table 2 Mean values of macro-invertebrate densities (individuals per m^2) for each distance from the herbaceous strip (HS) and each season for the three shade treatments combined. Mean values were obtained from linear generalized models fitted with a quasi-Poisson law. Values are back-transformed from the logarithmic scale used for analyses. Different lower case letters indicate significantly different values between the means for a given taxonomic group (Tukey test, p-value < 0.05). Different upper case letters indicate significantly different values between the different distance from the HS for all seasons combined when the answer was similar for all seasons (no significant effect of the interaction between season and distance from the HS in Table S3 of the Supplementary materials) (Tukey test, p-value < 0.05). Bold values and letters underline significant differences between distances from the HS for the taxonomic group and season concerned. For Formicidae, a cumulative link model was used.

	Distance from the HS	April	June	November
Lumbricidae	0 m	**186.56 ab**	**174.03 ab**	**204.31 a**
	1 m	**59.48 d**	**36.04 de**	**129.13 bc**
	2.5 m	**45.45 de**	**25.17 e**	**106.55 c**
Diplopoda	0 m	**66.10 ab**	**60.32 abc**	108.47 a
	1 m	**29.55 bcd**	**18.65 d**	62.87 abc
	2.5 m	**25.43 cd**	**11.2 d**	61.33 ab
Isopoda	0 m	**46.30 a**	23.21 ab	31.19 ab
	1 m	**16.29 b**	23.21 ab	47.38 a
	2.5 m	**12.47 b**	23.91 ab	35.74 ab
Chilopoda	0 m	31.02 cd	39.86 cd	**62.76 bc**
	1 m	16.66 d	39.93 cd	**108.96 a**
	2.5 m	17.66 d	35.74 cd	**91.09 ab**
Coleoptera (adults)	0 m	**51.33 ab**	**61.74 a**	35.69 abc
	1 m	**19.25 c**	**28.38 bc**	35.83 abc
	2.5 m	**30.19 abc**	**21.85 bc**	21.37 c
Coleoptera (larva)	0 m A	35.65	28.01	58.95
	1 m A	58.91	20.09	72.69
	1.5 m A	50.52	23.41	64.25
Dermaptera	0 m A	10.69	17.86	35.67
	1 m A	12.01	18.78	41.69
	2.5 m A	4.37	11.52	38.64
Formicidae [1]	0 m **A**	0 - 240	0 - 240	0 - 240
	1 m **AB**	0	0 - 240	0 - 240
	2.5 m **B**	0	0	0

[1] category to which the median value belongs (between 0, 0 to 240 and more than 240 individuals m^{-2})

Table 3 Mean values ± standard deviation of the density of each earthworm species (individuals per m^2), and mean values of earthworm diversity indices from generalized linear models (fitted by quasi Poisson law for species richness, and normal law for Shannon index). Different lower case letters indicate significantly different values between the means (Tukey test, p-value < 0.05). Different upper case letters indicate significantly different values between the different distance from the HS for all seasons combined when the answer was similar for all seasons (no significant effect of the interaction between season and distance from the HS in **Table S3** of the **Supplementary materials**) (Tukey test, p-value < 0.05). Bold values and letters underline significant differences between distances from the HS and seasons for index concerned.

	Distance from the HS	April	June	November
Lumbricus terrestris L. (anecic)	0 m	5.33 ± 9.03	4.00 ± 7.08	2.67 ± 6.09
	1 m	2.67 ± 7.70	0.67 ± 3.27	2.67 ± 6.09
	2.5 m	0.00 ± 0.00	0.67 ± 3.27	1.33 ± 4.52
Lumbricus castaneus S. (epigeic)	0 m	38.00 ± 42.15	40.67 ± 47.65	46 ± 36.64
	1 m	15.33 ± 21.87	9.33 ± 11.48	48 ± 34.99
	2.5 m	4.00 ± 11.80	6.00 ± 9.21	46 ± 47.49
Allolobophora chlorotica S. (endogeic)	0 m	17.33 ± 27.07	30.00 ± 35.72	35.33 ± 36.54
	1 m	8.00 ± 10.55	8.00 ± 14.92	31.33 ± 34.18
	2.5 m	5.33 ± 9.03	8.00 ± 14.15	17.33 ± 20.52
Aporrectodea nocturna E.[1] (anecic)	0 m	114.67 ± 57.33	83.33 ± 52.96	85.33 ± 58.3
	1 m	32.00 ± 29.08	12.67 ± 14.14	33.33 ± 35.60
	2.5 m	32.00 ± 31.65	8.00 ± 12.48	36.00 ± 36.32
Aporrectodea rosea S. (endogeic)	0 m	14.00 ± 16.56	12.67 ± 14.90	28.00 ± 26.38
	1 m	3.33 ± 8.14	3.33 ± 6.64	15.33 ± 15.99
	2.5 m	5.33 ± 10.19	1.33 ± 4.52	4.67 ± 7.43
Aporrectodea icterica S. (endogeic)	0 m	0.00 ± 0.00	4.00 ± 4.00	8.00 ± 8.00
	1 m	0.00 ± 0.00	2.00 ± 2.00	2.00 ± 2.00
	2.5 m	0.00 ± 0.00	0.00 ± 0.00	1.33 ± 1.33
Aporrectodea caliginosa S. (endogeic)	0 m	0.00 ± 0.00	0.00 ± 0.00	3.33 ± 3.33
	1 m	0.00 ± 0.00	0.00 ± 0.00	1.33 ± 1.33
	2.5 m	0.00 ± 0.00	0.00 ± 0.00	1.33 ± 1.33
Species richness	0 m	**2.9 ab**	**3.3 ab**	**3.9 a**
	1 m	**1.9 cd**	**1.7 cde**	**3.2 ab**
	2.5 m	**1.4 de**	**1.1 e**	**2.3 bc**
Shannon index	0 m **A**	2.10	2.52	3.15
	1 m **B**	1.72	1.52	2.57
	2.5 m **C**	1.36	1.20	2.01

[1] synonymy *Nicodrilus nocturnus* Bouché, 1972 (Briones et al., 2009)

Spring   **Summer**   **Autumn**

Density of:

Litter feeders
(earthworms, woodlice, millipedes)

Animal feeders
(centipedes, staphylinid
beetles, carabid beetles larvae)

Plant feeders
(wireworms)

Herbaceous strip

Crop alley
(tomatoes)