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Quentin Vincent, Corinne Leyval, Thierry Beguiristain, Apolline Auclerc. Functional structure and composition of Collembola and soil macrofauna communities depend on abiotic parameters in derelict soils. Applied Soil Ecology, 2018, 130, pp.259-270. 10.1016/j.apsoil.2018.07.002. hal-01930921

HAL Id: hal-01930921

https://hal.science/hal-01930921

Submitted on 8 Mar 2019

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Functional structure and composition of Collembola and soil

macrofauna communities depend on abiotic parameters in

3 derelict soils

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ABSTRACT

In the last decades, anthropogenic disturbances have altered the ability of soils to provide diverse functions. Certain anthropogenic soils, with a low fertility level and often contaminated, ended up underused and derelict. Although derelict for humans, these soils may be refuges for biodiversity, but their biological functioning remains poorly understood. To this end, a trait-based approach of soil invertebrate communities might be an effective predictor of ecosystem state. The present work aims to highlight the *in situ* links between the abiotic characteristics of derelict soils and the taxonomic and functional structure and composition (through a trait-based approach) of macrofauna and Collembola communities inhabiting these soils. We studied 6 different derelict soils: two soils from coking plants, one soil from a settling pond, two constructed soils, and an inert waste storage soil. We measured fifteen abiotic soil parameters that inform on fertility and contamination. We took into account sixteen traits and ecological preferences to characterize the functional structure and composition of Collembola and macrofauna communities. Soil fertility (organic matter content, C:N ratio, P, Ca and

Mg concentrations, cation-exchange capacity, and clay content) and moderate contamination (Pb, Cd, Zn, and PAH concentrations) altered the taxonomic and functional composition of Collembola and macrofauna communities by selecting traits such as body length, pigmentation, vertical distribution, diet type, and habitat preference. Compost-amended constructed soil properties selected taxonomic and functional community composition of slightly disturbed soil. In contrast, metal-contaminated constructed soil harbored a higher proportion of Collembola displaying the traits and ecological preferences of instable ecosystems. The study of functional profiles of Collembola and macrofauna communities in the derelict soils evidenced that they support different communities with more or less wide functional potential. It underlines the interest of multiple biotic component studies to reach a better ecosystem description.

HIGHLIGHTS

- 35 Collembola and macrofauna communities from derelict soils were characterized.
- 36 Links between fauna community composition and soil characteristics were studied.
- 37 Fertility and contamination altered invertebrate community structure and composition.
- 38 Compost-amended constructed soil selected typical slightly disturbed soil species.
- 39 Collembola species traits in contaminated and constructed soil were typical of instable ecosystems.

KEYWORDS

42 Derelict soils, Functional diversity, Traits and ecological preferences, Soil invertebrates, Technosol

1. INTRODUCTION

Soils are complex ecosystems, described as multifunctional systems because many components interact inside them (Kibblewhite et al., 2008; Briones, 2014). Nevertheless, over the last decades growing evidence has emerged about the negative impact of anthropogenic disturbances on the ability of soils to provide functions (Levin et al., 2017) and occasionally to host biodiversity (Orgiazzi et al., 2016). In this context, the closing down of steel, shipbuilding or metal manufacturing industries because of economic issues and the closing down of mining sites in different European countries have resulted in land abandonment and altered ecosystem functioning (Wong and Bradshaw, 2002). Sometimes called greenfields, wastelands, uncultivated/vacant/abandoned lands, the soils of these derelict lands can be disturbed, i.e. characterized by a low fertility level (Dickinson, 2003) and/or sometimes by contamination (Morel et al., 2005). These specific abiotic parameters can alter soil functions likely to be important for agriculture and lead to surfaces being unmanaged and underused (Cundy et al., 2016). In Vincent et al. (2018), we showed that derelict soils could have low organic matter and nitrogen contents, a low cation exchange capacity, alkaline conditions with cations occasionally lacking or in excess, and moderate organic and inorganic contamination linked to their history. Biotic parameters (i.e. plant, fauna and microorganism density and richness) co-varied more with soil fertility proxies than with soil contamination parameters in moderately contaminated derelict soils. Once soils are underused and abandoned by humans, they may turn into refuges for biodiversity provided that abiotic parameters are not extreme for life (Baranova et al., 2014). Derelict soils have been reported to harbor a rich invertebrate biodiversity; insects (Bonthoux et al., 2014), beetles (Eyre et al., 2003), carabids (Small et al., 2006), earthworms and Collembola (Butt and Briones, 2017). However, the biological functioning of derelict soils remains poorly understood as compared to forest or agricultural lands. In this vein, trait-based approaches focusing on the characteristics of individuals provide an interesting tool to assess soil functioning (Verberk et al., 2013). Pey et al. (2014) defined traits in soil invertebrate studies as "morphological, physiological, phenological or behavioral features measurable at the individual level, from the cell to the whole-organism level, without reference to the environment or any other level of organization". Traits could be considered as functional because they impact

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organism fitness directly and indirectly (Violle et al., 2007). Finally, functional traits can help to understand the effect of environmental stressors on soil communities (Auclerc et al., 2009; Vandewalle et al., 2010; Hedde et al., 2012; Salmon and Ponge, 2012). In recent years, traits have been recognized as effective predictors of exposure to disturbances (metal contamination in Hedde et al., 2012; restoration practices in Rosenfield and Müller, 2017), or of management strategies such as intensive agriculture, polyculture and monoculture (Wood et al., 2015; Sechi et al., 2017) to better understand the soil functional community structure and composition.

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The functional community composition of the soil fauna is linked to soil abiotic parameters. For example, in urban soils, the proportion of Collembola with a sexual reproduction type was positively correlated with total Cu and Ni concentrations, and the proportion of pigmented Collembola was positively correlated with total Cr, Pb and Zn concentrations (Santorufo et al., 2015). The proportion of Collembola living belowground (with the following trait attributes: small size, reduced or absent sensorial organs and furca, non-sexual reproduction type) in coal mine spoil tip soils was found associated to a coarser soil texture and higher nitrogen and organic matter concentrations (Vanhee et al., 2017). In parallel, the proportion of macroinvertebrates with a soft body decreased when metal contamination increased (Hedde et al. (2012), and the body size of ants was found driven by soil granulometry (Costa-Milanez et al. (2017). Moreover, other authors showed that soil age played a key role in the functional community composition by selecting certain traits such as moisture or light requirement in carabid communities (Aubin et al., 2013) or the feeding group for macrofauna communities (Frouz et al., 2013). To our knowledge, existing studies dealing with trait-based approaches in anthropogenic sites focussed on only one group of soil invertebrates (Collembola or carabids or macrofauna) and were carried out on highly contaminated and/or low fertile soils. Furthermore, there are too few studies considering the links between soil chemistry parameters and trait-based invertebrate community composition (e.g. Salmon and Ponge, 2012; Santorufo et al., 2014, Santorufo et al., 2015; Martins da Silva et al., 2016).

The present work aims to characterize which environmental factors shape macrofauna and Collembola invertebrate communities and their functional structure and composition in derelict soils characterized

by moderate contamination and/or low fertility. We hypothesized that the soil characteristics related to past industrial activities and/or to the materials used for construction drove the composition of invertebrate communities by selecting certain traits and ecological preferences. We studied 6 different derelict soils: two soils from coking plants, one soil from a settling pond, two constructed soils, and an inert waste storage soil (Vincent et al., 2018). We took into account sixteen traits and ecological preferences to characterize the functional structure and composition of the soil invertebrate communities.

2. MATERIALS AND METHODS

2.1. Derelict soils and fauna sampling methods

We studied six soils from open industrial derelict sites located in north-eastern France in a historically industrial region, with many former coking plant sites and steel factories. The climate in this region is continental with annual mean temperature of 10.8°C. For the sampling year (2015), monthly mean temperatures ranged between 1.8 and 22.1°C and cumulated annual rainfall was around 500 mm. Soil and fauna were sampled between the 8th and 29th of April 2015 (Vincent et al., 2018), during the period of highest invertebrate activity.

Soil named "CS" (for constructed soil) was a 1.13 ha experimental constructed soil set up on the site of a former coking plant (Séré et al., 2008). Soil named "WL" (for waste landfill) was from a 0.7 ha non-hazardous waste landfill. Soils named "CP1" and "CP2" (for coking plant site 1 and 2, respectively) were from former coking plant sites. These two soils differed in metal trace element concentrations. Soil named "SP" (for settling pond) was from a settling pond site filled with steel sludge. Lastly, soil named "CCS" (for constructed and contaminated soil), was an experimental 0.09-ha constructed soil. See Table 1 for more details.

We measured eleven abiotic variables that describe soil fertility: organic matter (OM) content, water holding capacity (WHC), clay content, exchangeable Ca, K, Mg and Na cation concentrations, Olsen phosphorus content, pH, cation-exchange capacity (CEC), C:N ratio. We also measured four chemical variables to quantify the contaminant levels in the soils, i.e. PAH and available Cd, Zn and Pb concentrations. We also took into account the time span since the last anthropogenic action on each site. The abiotic characteristics are summarized in Table 1 (for more details see Vincent et al. 2018)). We took five soil samples from each derelict site (except for CCS where four samples were taken), along a 20-m transect with 5 m distance between each sample, in accordance with the Tropical Soil Biology and Fertility (TSBF) method (Anderson and Ingram 1993). We characterized communities in open areas (i.e. not forested areas) and adjusted our transect strategy to avoid potential edge effects for one given site. Collembola communities were sampled using soil cores and MacFadyen method, and macrofauna communities were sampled by soil blocks and hand-sorting method. Collembola were sampled on the 6 plots, on 10 cm depth, using two 5-cm deep corers (according to the ISO 23611-2 standard), fitted one above the other. The microarthropods were extracted using a high-temperature gradient extractor (MacFadyen, 1961) and identified to the species level. The soil macrofauna communities were studied on 5 sites by excavating 0.25×0.25×0.30 m soil blocks, according to the ISO 23611-5 standard. Macrofauna community from CCS was not studied because the destructive method would have caused high disturbances on the experimental plots. Macrofauna organisms were hand-sorted in laboratory, and kept in a 70% (v/v) ethanol solution. They were all identified down to the family level, and further down to the species level for rove beetles, ground beetles and earthworms. Ants were removed from all analyses because these social insects are highly aggregated in space (Nahmani and Rossi 2003).

2.2. Functional study (trait-based approach) of Collembola and macrofauna

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We selected 7 traits for the trait-based approach of the Collembola study: body length (BLN), body shape (BSH), number of ocelli (OCE), presence of a post antennal organ (PAO), pigmentation (PIG),

motion strategy (MS), reproduction type (REP), and 3 ecological preferences: vertical distribution (DIST), micro-habitat preference (MHABI), and habitat preference (HABI). As for the macrofauna, we selected 3 traits: body length (BLN), presence of an integument (INT), and mouth part type (MthT), and 3 ecological preferences: diet type (DIET), micro-habitat preference (MHABI), and habitat preference (HABI). These traits were chosen because they are commonly used to characterize the dispersal abilities of organisms in a given ecosystem, or to describe habitats and environmental stressors (Makkonen et al., 2011).

We gathered values (*e.g.* attributes) of the traits and ecological preferences of species from various sources (aggregated data, books, literature, *etc.*) but mainly from the BETSI (Biological and Ecological Traits of Soil Invertebrates - http://betsi.cesab.org) database. We used 8,200 Collembola data and 1,400 macrofauna data. The different traits/ecological preferences and attributes we assessed are listed in Table 2.

To analyze attribute data for a given taxon, we performed a fuzzy coding analysis. This method was developed by Chevene et al. (1994) and is widely used for trait approaches. Calculations were performed according to the method of Hedde et al. (2012). The information obtained from each source was coded by an affinity score for a given taxon and for a given trait attribute ranging from 0 to 4 (from no (0) to very high affinity (4) of the taxon to a trait attribute, respectively). Subsequently, the affinity scores obtained from all the sources were added up and transformed into a percentage to build the trait profile of each species/taxon. Finally, for each trait attribute, the mean affinity for the entire Collembola or macrofauna community, called Community Weighted Mean (CWM), was calculated according to Garnier et al. (2004). The CWM calculation was performed using equation 1,

169 (Eq. 1) $CWM = \sum_{i=1}^{n} P_i \times T_i$,

where Pi is the relative abundance of species/taxon i, n is the number of Collembola or macrofauna taxa, and T_i is the relative trait/ecological preference attribute affinity of species/taxon i.

CWMs were calculated separately for Collembola and macrofauna communities, for each trait attribute in each sample, using the vegan package (Oksanen et al., 2017) in R software (version 3.1.3,

2.3. Taxonomic and functional diversity indices of Collembola and

macrofauna communities

We measured different ecological indices that describe taxonomic or functional community structure because they are commonly used to assess ecosystem stability and functioning (Tilman, 2001; Tilman et al., 2014; Bukvareva, 2017). For a taxonomic description of the communities sampled in each derelict soil, we estimated taxonomic richness (TRic) and taxonomic evenness (TEve) using the vegan package (Oksanen et al., 2017) in R software. TRic was the number of taxa composing the Collembola or macrofauna communities sampled in each derelict soil, and TEve was the partitioning of individuals among the different taxa across each community.

For a functional description of Collembola and macrofauna community structure, we calculated functional richness (FRic) and functional evenness (FEve) according to (Villéger et al., 2008), using trait values (Laliberté et al., 2014). FRic was the volume created by attribute values in a multidimensional trait space. FEve was the evenness with which attributes were distributed in the multidimensional functional space. These functional indices were calculated using R software and the FD package (Laliberté et al., 2014).

2.4. Statistical and multivariate analyses

To test the differences in functional community composition among soils, we independently performed univariate tests on CWMs for each attribute of a given trait. We also performed univariate tests on TRic, TEve, FRic and FEve to compare taxonomic and functional diversity levels among soils. As no dataset followed the conditions of parametric tests (we checked normality with Shapiro Wilk's test and homogeneity of variances with Bartlett's test), we performed Kruskal-Wallis (K-W) tests followed by the Dunn's test of multiple comparisons with Bonferroni adjustment to control the experimentwise error rates (Dinno, 2017). We used R Software for data processing, and performed all univariate tests with a 95% significance level.

To highlight the relationships between the trait/ecological preferences and abiotic parameters of the derelict soils, we performed RLQ analyses (Dolédec et al., 1996). RLQ analysis is a generalization of co-inertia analysis (Dray et al., 2003) that couples multiple data sets and identifies co-relationships between them. RLQ analysis performs a double inertia analysis of two arrays (tables R and Q) with a link expressed by a contingency table (table L). RLQ analysis is a method to highlight how the environment filters certain species traits (Dray et al., 2014). We used it to provide simultaneous ordination, and to analyze the joint structure of three datasets: the abiotic parameter dataset of each sample (table R), the relative abundance of taxa in each sample (table L), and the attribute trait values for each taxon (table Q). We tested the significance of the relationship between taxon traits and abiotic parameters using a Monte Carlo test (999 permutations). We performed RLQ analysis for Collembola and macrofauna communities separately, using the ade4 package (Dray and Dufour, 2007) in R software. This approach is one of the most integrated methods to analyze trait-environment relationships (Dray and Dufour, 2007; Kleyer et al., 2012). We tested linear regressions with the lmtest package in R, and correlations between abiotic and biotic parameters with the Pearson correlation coefficient (Hothorn et al., 2015).

3. RESULTS

3.1. Collembola communities from all 6 derelict soils

The density of Collembola ranged between 17,000 and 91,000 individuals *per* m² on average, and their taxonomic richness between 3.4 and 7 per sample (Table 3). As regards the taxonomic approach, Collembola species richness was the highest in SP, CP2 and CCS, and significantly different from CS (K-W test; P=0.005). The evenness of Collembola species did not significantly differ among soils, with mean values ranging from 0.46 to 0.74. As for the trait-based approach, Collembola functional richness (between 0.02 and 0.16) was the highest in CP1, SP and CP2, and significantly different from WL and CCS (K-W test; P<0.001). No difference in the functional evenness of Collembola communities was found among soils, with mean values ranging between 0.62 and 0.71.

Figure 1 shows the CWM of each attribute measured for each of the ten traits/ecological preferences of the Collembola communities sampled in each of the six derelict soils. For habitat preference, the Collembola collected through the study are forest and grassland taxa (81% of the community on average for the 6 soils) and preferred soil as a micro-habitat (64% of the community on average for the 6 soils). CS harbored a higher proportion of Collembola that have a furca and inhabiting decaying woods (ca. 24%) than in CP1, SP and CP2. CCS was characterized by a higher proportion of Collembola with the biggest size (2-3 mm; ca. 49%) compared to WL, SP and CP2. The percentage of Collembola with a PAO was higher in CCS (ca. 88%) than in WL. Collembola in soil CCS have a higher number of ocelli (5, 6, 7 or 8 pairs; ca. 100%), they are more pigmented (ca. 88%) compared to soils CP1 and SP, and have a sexual reproduction type (ca. 88%), with an epiedaphic ecological preference (ca. 93%, a significant higher value compared to CS, CP1 and SP). CCS harbored the lowest proportion of Collembola inhabiting forests and grasslands (62% of the community) but the highest proportion of Collembola inhabiting agricultural (22% of the CCS community) and wetland habitats (14% of the community). According to Figure 2A, the first two RLQ analysis axes accounted for 88% of total Collembola community variance (69 and 19%, respectively). The Monte-Carlo permutation test evidenced a global link between physico-chemical soil variables and the traits/ecological preferences of Collembola (P value: 0.052). The projection of the centroid calculated for each soil separated them into 4 groups: CCS; WL-CP1-CP2; CS; SP. Species composition is given in Figure 2B. Isotomurus fucicolus, Isotomurus antennalis, Desoria violacea, Proisotoma minuta and Entomobrya lanuginosa were in the highest density in CCS as compared to the other soils. *I.fucicolus*, *I.antennalis* and *P.minuta* were only sampled in this soil. Likewise, CP2 hosted Smithurinus schoetti, Sphaeridia pumilis and Sminthurinus elegans in a higher abundance than the other soils. S.schoetti was only sampled in CP2. Metaphorura affinis, Cyphoderus albinus and Paratullbergia callipygos had the highest density in SP. The other soils (CS, WL, CP1, and CCS) did not host exclusive Collembola species (i.e. sampled in only one of the six soils).

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Figure 2C shows that the highest proportion of Collembola inhabiting mineral micro-habitats was associated with the highest C:N ratio, pH and Ca concentration and a small quantity of clay, which were the abiotic characteristics of SP. The higher proportions of forest- and grassland-inhabiting Collembola were associated with high OM contents and low trace elements (Cd, Zn, Pb) concentrations in soils. These trait-abiotic parameter relationships were characteristic of CS, and to a lesser extent of WL, CP1 and CP2. CCS had the highest Cd, Zn and Pb concentrations and the lowest OM content. It hosted the highest proportion of Collembola with a high number of pairs of ocelli (*i.e.* 5, 6, 7 or 8), and the highest proportion of Collembola inhabiting wetland habitats. The highest proportion of Collembola inhabiting in root and soil micro-habitats was associated with the highest soil CEC values in CS and to a lesser extent in WL, CP1 and CP2. Finally, the highest proportion of Collembola with a low number of ocelli (*i.e.* 1, 2, 3 or 4) was associated with the highest soil CEC and PAH concentrations, such as measured in CS. The other traits/ecological preferences were not clearly linked to measured physico-chemical parameters. Moreover, the time span since the last anthropogenic action on each site (*i.e.* "age") did not seem to influence trait selection across the sites.

3.2. Macrofauna communities in 5 derelict soils

The average macrofauna density ranged between 118 and 668 individuals *per* m², and the average taxonomic richness of the macrofauna community ranged between 3.0 and 11.6 per sample (Table 3). Taxon richness (between 6.0 and 16.2) was significantly higher in CS and WL than in SP and CP2 (K-W test; P=0.001). No significant difference in the evenness of macrofauna taxa was measured among soils, with mean values ranging from 0.53 to 0.71. Functional richness (between 0.06 and 0.14) was significantly higher in CS than in SP, CP1 and CP2 (K-W test; P=0.041). As for Collembola community results, no difference in functional evenness was measured among soils, with mean values ranging from 0.74 to 0.78.

Figure 3 shows the CWM of the six traits/ecological preferences of the macrofauna communities sampled in the five soils. For habitat preference, the macro invertebrates of the 5 soils are forest and

grassland habitat taxa (around 50% of the community in the 5 soils, without significant difference among sites).

CS was characterized by a significantly higher proportion of invertebrates without a specific mouthpart (mainly earthworms; *ca.* 40%) compared to other soils; CS harboured also a higher proportion of geophageous trophic group (*ca.* 24%) compared to WL, SP and CP2 and a higher proportion of invertebrates inhabiting a mineral micro-habitat (*ca.* 58%). SP was characterized by a higher proportion of small invertebrates (2.5-5 and 5-10 mm; *ca.* 40%) than in the other soils, and a higher proportion of invertebrates with a chewing mouthpart type (*ca.* 92%). SP was also characterized by higher proportion of zoophagous trophic groups (*ca.* 58%) and of wetland habitat taxa (*ca.* 37%).

The first two RLQ analysis axes accounted for 87% of total macrofauna community variance (71 and 16%, respectively) (Figure 4A). The Monte-Carlo permutation test indicated a highly significant (P=0.01) link between physico-chemical variables and traits/ecological preferences of soil macroinvertebrates. The projection of the centroid calculated for each soil delineated 3 groups: CS; WL-CP2; CP1-SP. Taxon composition is presented in Figure 4B. *Athous* click-beetle, Dermaptera, *Allolobophora chlorotica*, *Allolobophora oculata* and *Aporrectodea longa* earthworms had a higher density in CS than in the other soils. The Miridae group were in a higher density in WL and CP2 than in the other soils. Limacidae, the spider family Linyphiidae, the centipede family Geophilidae and the millipede order Julida (with especially the species *Ommatoiulus sabulosus*) had a higher density in CP2 than in the other soils. Finally, Steninae and Staphylinidae were in a higher density in SP than in the other soils.

Figure 4C shows that the highest proportions of macrofauna without a mouthpart, geophagous invertebrates and animals with a preference for agricultural habitats were associated with higher soil CEC and WHC, which were characteristic of CS. A weak but significant positive correlation was observed between earthworm density and soil CEC values (R²=0.40; T-test; P<0.001) and between earthworm density and OM content (R²=0.26; T-test; P=0.01). The highest proportion of invertebrates

inhabiting mineral micro-habitats was associated with the highest soil PAH, phosphorous, and available Cd concentrations and the highest OM content. The highest proportion of invertebrates with a wetland habitat preference was associated with the highest soil Mg, Ca and Pb concentrations, the highest C:N ratio and the lowest soil K concentration, characteristic of CP1 and SP. The highest proportion of individuals with a sclerotized integument was associated with the highest available soil Zn concentrations and lowest clay contents. The time span since the last anthropogenic action on each site (*i.e.* "age") did not seem to have any impact on functional community composition.

4. DISCUSSION

The use of functional traits of edaphic fauna highlighted differences in Collembola and macrofauna communities structures among the studied derelict soils. CS (a constructed compost amended soil) and WL (a non-hazardous waste landfill soil) had the highest taxonomic and functional macrofauna richness, but the lowest taxonomic and functional Collembola richness of the studied sites. These soils probably contained a high quantity of food resources and many potential different niches possibly linked to a high plant density, with no metal contamination. Focusing on earthworms, CS hosted the highest density of the studied sites. The presence of earthworms is currently considered as a good indicator of the absence of disturbance such as metal contamination and land-use intensification (Nahmani and Lavelle, 2002; Pérès et al., 2011; Cluzeau et al., 2012). We can suggest that competition for resources between meso- and macro-organisms can occur at one given time. For example, Eisenhauer (2010) showed negative impacts of endogeic earthworms due to competition for food resources with microarthropods such as Collembola. Earthworms can also affect the Collembola community by modifying, maintaining, creating or destroying habitats (Eisenhauer, 2010; Wright et al., 2012). At the opposite, CCS (an experimental constructed and contaminated soil) with the lowest fertility level and the highest metal concentration of the study, harbored the highest taxonomic

diversity but a low functional diversity of Collembola. This might suggest functional redundancy of species within the community, defined as several species contributing in equivalent ways to an ecosystem function such that one species may substitute for another (Bruno et al., 2016). Species that are functionally redundant act as 'spare wheels' during a disturbance to maintain ecosystem functions (Cole et al., 2006). Functional redundancy of species in a given community might be interesting in disturbed habitats, and then a community composed of species with similar functional traits might be more resilient to a new disturbance (Gerisch, 2014; Hodecek et al., 2016).

Following the RLQ analyses, the differences in fertility levels and contaminant concentrations among the 6 anthropogenic derelict soils appeared to be associated with different composition of Collembola and macrofauna communities at the taxonomic and functional levels, with difference of response for both communities.

Firstly, the OM content was associated with the presence of Collembola Lepidocyrtus lanuginosus and Isotomiella minor, found at high densities in soils with high OM contents (Santorufo et al., 2014) and in coal mine spoil tip soils (Vanhee et al., 2017). These species are considered as forest-inhabiting species (Heiniger et al., 2015). They have been showed to prefer soils with dense vegetation (high plant biomass) and rich in OM (Gillet and Ponge, 2004; Fjellberg, 2007; Dunger and Schlitt, 2011). The OM content, which was the highest in CS owing to compost addition, appeared to positively influence the density of forest- and grassland-inhabiting Collembola in our study, but had no effect on the macrofauna. Collembola communities could be more driven by fine OM (only OM smaller that 2 mm was assessed in this study) than macrofauna because of the size of the food they eat. Secondly, the highest soil C:N ratio was linked to the highest proportion of wetland-inhabiting macrofauna sampled in SP and CP1 (settling pond and coking plant soils, respectively). Wetlands harbor plant communities with a high C:N ratio that are decomposed slowly (Plum, 2005) and can in turn result in a high soil C:N ratio. Thirdly, the soil CEC value was linked to the proportion of invertebrates without a mouthpart, mainly represented by earthworms, in WL. The selection of earthworm populations (geophageous invertebrates without a mouthpart) by high soil CEC has already been observed in soils of oil palm plantations and agricultural areas (Sabrina et al., 2009; Hedde et al., 2012). The authors underlined that the effects of CEC on earthworm populations was similar to the effects of organic carbon. In our study, a weak correlation was observed between earthworm density and OM content or CEC values. The soil CEC value also seemed to be an important factor on Collembola species characterized by trait attributes linked to a high dependence on soil as a habitat (*i.e.* "soil-inhabiting", "inhabiting root micro-habitats" and "low number of ocelli"). Finally, Ca and Mg concentrations in the soils also appeared to alter the proportion of chewing mouth-part type macrofauna that we assumed to use herbaceous litter as food. An explanation of this abiotic-biotic link could be that the Ca concentrations in plants and consequently in litter originate from the soil. Thus, in the studied soils, litter calcium might have come from the materials used for constructing soils, and may have influenced the functional composition of soil invertebrates by selecting certain species with a specific mouth-part type.

Metal concentrations seemed also to influence the density of certain species and to alter the functional community composition of the soil biodiversity, especially for Collembola. In this vein, *Proisotoma* minuta was only found in the soil with the highest metal concentration (CCS). P.minuta has already been shown as an indicator of manure or slurry pollution and predominates in decaying sewage sludge (Dunger and Schlitt, 2011). It seems to be a generalist species adapted to different ecosystems likely to be altered. Euedaphic Collembola are considered as the most sensitive living forms to metal contamination according to several studies (Fountain and Hopkin, 2004; Parisi et al., 2005; Joimel, 2015). Epedaphic Collembola, are more resistant to pollutants than euedaphic Collembola because they have a higher movement ability and a lower direct contact with soil particles (Fountain and Hopkin, 2004). This result is consistent with our findings: the derelict soil with the highest metal concentrations (CCS) did not host euedaphic Collembola. In the present study, the metal concentration also appeared to positively drive the proportion of Collembola with a maximum number of ocelli, as observed previously for soil Pb concentrations (Joimel, 2015). The number of ocelli is an indicator of the dispersal ability of Collembola since species with a complete visual apparatus can disperse rapidly (Ponge et al., 2006; Salmon et al., 2014). Several authors underlined that Collembola with a high dispersal ability were present in disturbed environments (Auclerc et al., 2009; Salmon et al., 2014; Joimel, 2015). Thus, the metal concentrations of our soils, though low as compared to other studies on anthropogenic soils, appeared to have selected Collembola with a high dispersal ability linked to specific morphological traits. In the same vein, the highest proportion of Collembola with sexual reproduction was observed in CCS, which contained the highest metal concentrations. A higher proportion of sexual reproduction species as compared to parthenogenetic Collembola species has been reported in copper-polluted environments (Niklasson et al., 2000). The reproduction type of Collembola is associated with survival or colonizing strategies (Salmon et al., 2014). Sexual reproduction favors polymorphism and is an advantage for colonizing disturbed ecosystems like polluted soils on a long term (Austruy et al., 2016). According to the trait-based approach, individuals with a long body, several ocelli, a PAO organ, a sexual reproduction type, an epiedaphic living form and a pigmented body mainly represented the Collembola community in soil F. All of these trait attributes are characteristic of organisms inhabiting in urban environments (Santorufo et al., 2014), and are linked to a good dispersal ability that allows adaptation to disturbed habitats. The age effect was not a major factor explaining the functional composition of communities according to the RLQ. Yet, we may expect links between the fact that CCS was the youngest plot and that it hosted a Collembola community rich in high dispersal capacity species, which are best adapted for site colonization.

Regarding macrofauna community, available soil Pb concentrations seemed to influence earthworm density: the highest earthworm density was found in soils with low metal contamination (CS and WL) as compared to other soils. This impact of Pb concentrations on earthworm communities has been shown by Lévêque et al. (2015), who observed a low earthworm density in soils near a Pb-recycling factory containing a high total Pb concentration (2,010 mg.kg⁻¹ of Pb on average, that is a higher concentration than in our soils). Pižl and Josens (1995) also observed a low earthworm density in soils with a moderate Pb concentration (89 ind.m⁻² in a soil containing 108 mg.kg⁻¹ of total Pb) as compared to a soil with a low Pb concentration (490 ind.m⁻² in a soil containing 25 mg.kg⁻¹ of total Pb). In our study, we found 42 and 48 earthworms.m⁻² on average in the soils with the highest total Pb concentrations (346 and 339 mg.kg⁻¹ for CP1 and SP, respectively) and 112 and 45 earthworms.m⁻² on

average in the soils with the lowest total Pb concentrations (41 and 37 mg.kg⁻¹ for WL and CP2, respectively). Other authors have also shown that earthworms were sensitive to metal pollution (Nahmani and Lavelle, 2002; Nahmani et al., 2005). As Collembola, earthworms abundance and diversity are dependent of soil properties, that highlight their bioindicator potential (Ponge et al., 2003; Pérès et al., 2011; George *et al.*, 2017). Furthermore, a higher available soil Zn concentration was associated with a higher density of sclerotized macrofauna individuals. We can hypothesize that organisms with sclerotized integuments are more protected against pollutants because sclerotized integuments provide defence against chemical attacks (Linz et al., 2016). By contrast, soft-body individuals may be directly exposed to pollution by direct skin contact. A negative correlation has already been observed between the proportion of soft-body individuals and metal soil concentrations (Hedde et al., 2012).

Finally, at a larger spatial scale, the surrounding landscape would have to be taken into account in order to better understand the variability of community composition across sites. Collembola and macrofauna communities can be both affected by surrounding environment and vegetation in derelict soils (Cortet et al., 2013; Baranova et al., 2014; Vanhee et al., 2017). Although no geostatistical analyses were performed in the present study, some hypotheses might be suggested on species traits selection. For instance, we found that communities were dominated by forest and grassland habitat taxa, which can be explained by the surrounding landscapes around the sites. CP1, SP and CP2 were surrounded by forests, while CS, WL and CCS were surrounded by grasslands. This could explain the significant higher proportion of euedaphic, blind and non-pigmented Collembola in CP1 and SP, that are characteristics traits of forest-inhabiting species (Salmon et al., 2014). Moreover, the proportion of macrofauna inhabiting wet habitats was higher in SP than in CS, WL and CP2, which could be explained by the proximity to a river (ca. 70 m away). The proportion of Collembola inhabiting wetland habitats was higher in CCS. The inefficient irrigation system in this experimental constructed soil probably allowed for colonization by wet-habitat species. Moreover, these species may have come from a nearby pond located at 300 m distance and been brought by trucks and human activities during the soil construction process or wind or animals (Costa et al., 2013).

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5. CONCLUSION

The diversity indices of Collembola and macrofauna communities showed different results attributable to differences in living modes between the two soil invertebrate communities (e.g. feeding groups and niche scales). It shows that the derelict soils support different communities with more or less wide functional potential depending on their physico-chemical characteristics. Our study revealed that derelict soil characteristics have more consequences on taxonomical and functional profiles of Collembola communities than macrofauna. In this sense, we can assume that Collembola are more relevant bioindicators of derelict soil characteristics than macrofauna. For instance, the studied more fertile and less contaminated compost-amended constructed soil exhibited taxonomic and functional community structures and composition of slightly disturbed soil. In contrast, the metal-contaminated constructed soil harbored a higher proportion of Collembola with the traits and ecological preferences of both unstable and scarcely vegetated ecosystems. Our study underlines the interest of trait-based approach and multi traits studies to assess the diversity of derelict soils and to reach a better understanding of soil ecosystem. However, in-depth analysis of direct relations between Collembola and macrofauna in these soils was limited due to different traits and attributes between both fauna groups in existing databases. Since we studied invertebrate communities at one given season, it might be interesting to characterize the community response patterns at different seasons. We previously measured the potential mineralization activity of these derelict soils (Vincent et al., 2018), but in situ measurement of soil functions, such as organic matter decomposition - by litter bag method - or soil respiration, might add complementary information to link functional traits of the fauna with the derelict soil functioning.

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ACKNOWLEDGEMENTS

This work was carried out within the GISFI (Groupement d'intérêt scientifique sur les friches industrielles, www.gisfi.fr), within the framework of the LORVER project (www.lorver.org). It was supported by the French Lorraine Region and the European Regional Development Fund (ERDF). The authors wish to thank Mickaël Hedde and Jodie Thénard for identifying ground beetles and a few other coleoptera, and BETSI project colleagues for providing access to their database. We also thank Sandrine Salmon and Johanne Nahmani for coding the traits of numerous Collembola species, and Sophie Joimel for providing the Collembola trait database. We also thank the technical staff of the GISFI, LIEC and LSE for the sampling campaign, particularly Alain Rakoto.

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- Table 1: Abiotic parameters of the six derelict soils. Means \pm SD (n=5, except for soil F where n=4).
- 2 Soil classification according WRB (2014); localization in the Lambert 93 projection in m; age is the
- 3 time lapse from the last anthropogenic action (years); clay and WHC in %; CEC in cmol⁺.kg⁻¹ dry soil;
- 4 soil pH in water; Olsen Phosphorous in mg.kg⁻¹ dry soil; OM in ‰; Exchangeable Ca, K, Mg, Na in
- 5 mg.kg⁻¹ dry soil, Available and total Cd, Pb, Zn in mg.kg⁻¹ dry soil; and PAH (Polycyclic Aromatic
- 6 Hydrocarbons) concentrations in mg.kg⁻¹ dry soil. For more details, see Vincent et al. (2018).

Soil abbreviation	CS	WL	CP1	SP	CP2	CCS
Soil name correspondence with Vincent et al., 2018	A	В	С	D	E	F
Site type and name	Experimental constructed soil (Biotechnosol)	Non- hazardous waste landfill (Retonfey)	Coking plant site (Homécourt)	Settling pond site (Moyeuvre- Petite)	Coking plant site (Micheville)	Experimental constructed and contaminated soil (Jeandelaincourt)
Soil classification	Spolic Garbic Technosol	Skeletic Technosol	Spolic Technosol	Spolic Technosol	Spolic Technosol	Spolic Technosol
Composition	paper mill waste, thermal desorption- treated PAH- contaminated soil, and green- waste compost	ground construction and demolition wastes	former coking plant site	settling pond site filled with steel sludge	former coking plant site	biopile-treated PAH- contaminated soil mixed with metal- contaminated sludge
Year of construction	2007	2011				2013
Year of abandonment			1980	1981	1975	
Age	8	4	35	34	40	2
Localization	E:918202 N:6905686	E:942352 N:6897630	E:918487 N:6905891	E:920222 N:6911528	E:942262 N:6897475	E:938477 N:6864617
Texture	Silt loam	Silty clay loam	Loamy sand	Sandy loamy	Loam	Silt loam
Clay	11	40	6	4	27	15
WHC	95±7	59±8	63±5	58±4	70±1	64±1
CEC	22.0±2.9	17.2±3.3	17.2 ± 1.8	11.3±0.7	15.5±1.4	10.1 ± 0.4
pH	7.9 ± 0.2	7.9 ± 0.1	7.9 ± 0.1	8.4 ± 0.2	7.2 ± 0.2	8.3±0.4
Olsen Phosphorous	62±5	25±17	39±16	44±10	36±2	44±10
C:N	30±6	28±6	26±5	65±20	20±3	37±16
OM	39.7±5.9	3.2 ± 1.4	16.7±6.3	13.3±2.8	4.8 ± 3.0	2.8 ± 0.8
$Ca_{\text{exchangeable}}$	292±44	713±144	903±405	2031±240	786±84	1123±130
$K_{\text{exchangeable}}$	706±105	491±24	155±36	377±30	131±50	218±36
$Mg_{\text{exchangeable}}$	84±14	152±38	103±6	312±47	47±15	72±34
$Na_{\text{exchangeable}}$	37±19	34 ± 28	11±3	42±11	14±14	55±29
$Cd_{available}$	0.22 ± 0.05	0.03 ± 0.01	0.16 ± 0.05	0.15 ± 0.01	0.10 ± 0.03	$9.07{\pm}1.5$
$Pb_{available} \\$	13±3	14±10	46±14	18±5	14±6	110±60
$Zn_{available}$	29±9	8±6	45±20	25±6	7±2	308±189
Cd_{total}	1.1±0.5	0.1 ± 0.2	0.1 ± 0.7	1.4 ± 0.1	0.1 ± 0.1	23.6±6.9
Pb_{total}	150±53	41±26	346±106	339±50	37±10	460±73
Zn_{total}	345±37	131±2	1162±396	1196±122	116±23	1813±302
Σ16 PAH (US-EPA)	170±49	12±12	179±136	97±54	142±90	10±4

- 8 Table 2: Traits/Ecological preferences of the Collembola and macrofauna sampled in the 6 derelict
- 9 soils. Codes are used in Figures 1 to 4.

	Trait/Ecological preference	Code of Trait/Ecological preference	Attribute	Code of attribute
	D - d l d		<1 mm	_<1
		BLN	[1:2] mm	_1_2
	Body length	DLN	[2:3] mm	_2_3
			>3 mm	_>3
	Body shape	BSH	Spherical	_sph
	Dody shape	D511	Cylindrical	_cyl
		OCE	No ocellus	_0
	Ocelli (visual organ)		1 or 2 pair of ocelli	_1_2
	ovem (visual organi)		3 or 4 pair of ocelli	_3_4
			5, 6, 7 or 8 pair of ocelli	_5_8
	Post Antennal Organ*	PAO	PAO present	_present
			PAO absent	_absent
_	Pigmentation	PIG	Pigmented	_present
Collembola			Non-pigmented	_absent
nb	Motion strategy	MS	Furca present	_furca
ler			Furca absent	_no.furca
Jo.	Reproduction	REP	Sexual reproduction type	_sex
\cup	<u> </u>		Asexual reproduction type	_asex
	37 41 1 11 4 11 41	DIST	Epigenic Hemiedaphic	_epi
	Vertical distribution		Euedaphic	_hemi
			1	_eu
	Micro-habitat preference	МНАВІ	Inhabiting in decaying micro-habitat Inhabiting in mineral micro-habitat	_dec _min
			Inhabiting in root micro-habitat	_mm _root
			Inhabiting in soil micro-habitat	_soil
	preference		Inhabiting in vegetal micro-habitat	_son _veg
			Inhabiting in wet micro-habitat	_wet
	<u></u>	HABI	Inhabiting in forest and grassland	_forest_grassland
			Inhabiting in agricultural area	_agri
	Habitat preference		Inhabiting in artificial area	_arti
			Inhabiting in wetland	_ _wet
		BLN	<2.5 mm	_<2.5
	Body length		[2.5:5] mm	_2.5_5
			[5:10] mm	_5_10
	•		[10:20] mm	_10_20
			>20 mm	_>20
	Intogument	INT	Sclerotized	_scl
	Integument	INT	Non-sclerotized	_no.scl
			Absent	_absent
		MthT	Piercing-sucking	_piercing.sucking
na	Mouth-part type		Swallowing	_swallowing
an			Sucking	_sucking
rof			Chewing	_chewing
Macrofauna		DIET	Zoophagous	_Z00
\geq	Diet		Phytophagous	_phyto
	- ***		Geophagous	_geo
		MILEDI	Detritivore	detri
	Micro-habitat		Inhabiting in decaying micro-habitat	_dec
	preference	MHABI	Inhabiting in mineral micro-habitat	_min
			Inhabiting in vegetal micro-habitat	_veg
			Inhabiting in forest and grassland	_forest_grassland
	Habitat preference	HABI	Inhabiting in agricultural habitat	_agri
			Inhabiting in artificial habitat	_arti
			Inhabiting in wetland	_wet

^{*} The Post Antennal Organ (PAO) is an organ composed of thermo-, hygro-, or chemo-sensitive receptors.

Table 3: Taxonomic and functional indices for Collembola communities in the six derelict soils and for macrofauna communities in 5 derelict soils. Means ± SD (n=5, except for CCS where n=4). Different letters indicate significant differences among soils (P< 0.05; Kruskal-Wallis test followed by multiple comparisons of rank distribution with Fisher's LSD test). TRic=Taxonomic richness; TEve=Taxonomic evenness; FRic=Functional richness; FEve=Functional evenness. CS=constructed soil; WL=waste landfill soil; CP1=coking plant soil 1; SP=settling pond soil; CP2=coking plant soil 2; CCS=constructed and contaminated soil.

	_	Soils								
	_	CS	WL	CP1	SP	CP2	CCS			
		Indices for Collembola community								
Taxonomic	TRic	3.4±1.3 b	4.2±0.8 ab	6±1.4 ab	7±0.8 a	7±2.2 a	5.8±1 a			
	TEve	0.67 ± 0.23	0.63 ± 0.18	0.46 ± 0.12	0.52 ± 0.1	0.52 ± 0.12	0.74 ± 0.12			
Functional	FRic	0.07±0.04 ab	0.04±0.03 b	0.13±0.04 a	0.14±0.03 a	0.16±0.04 a	0.02±0.01 b			
	FEve	0.62 ± 0.10	0.56 ± 0.22	0.68 ± 0.15	0.65 ± 0.08	0.71 ± 0.04	0.68 ± 0.15			
			Indices for macrofauna community							
Taxonomic	TRich	16.2±4.4 a	13.8±4.9 a	8.7±3.1 ab	6.6±3.2 b	6.0±3.8 b	NA			
	TEve	0.53±0.06	0.62±0.11	0.65 ± 0.27	0.71±0.14	0.71±0.23	NA			
Functional	FRich	0.14±0.01 a	0.10±0.05 ab	0.06±0.05 b	0.07±0.05 b	0.07±0.05 b	NA			
	FEve	0.76±0.03	0.76±0.07	0.78±0.12	0.77±0.10	0.74 ± 0.14	NA			

FIGURE CAPTIONS:

1

2 Figure 1: Community Weighted Mean (CWM) values of Collembola communities sampled in each 3 derelict soil, calculated for the following traits: number of ocelli, body length, vertical distribution, presence of furca, body shape, presence of PAO, pigmentation, reproduction type, habitat preference, 4 5 and micro-habitat preference. Means (n=5, except for CCS where n=4). Different letters indicate 6 significant differences among the 6 soils (P< 0.05) using Kruskal-Wallis tests followed by the Dunn's 7 test of multiple comparisons with Bonferroni adjustment. CS=constructed soil; WL=waste landfill 8 soil; CP1=coking plant soil 1; SP=settling pond soil; CP2=coking plant soil 2; CCS=constructed and 9 contaminated soil. 10 11 Figure 2: Projection of the coordinates of each soil replicate on the first two main components RLQ1 12 and RLQ2 (A), mapping of Collembola species density coordinates (B), and plot of attributes of each 13 trait linked to abiotic soil parameters (C). PAH=sum of the 16 PAH concentrations; Clay=proportion of clay; P= available phosphorous (Olsen method); OM=Organic Matter; WHC=Water Holding 14 15 Capacity; CEC=Cation Exchange Capacity; C:N=carbon/nitrogen ratio; [Exchangeable cations by 16 cobaltihexamine extraction] Ca=calcium; Na=sodium; K=potassium; Mg=magnesium. [Available elements] Pb=lead; Zn=zinc; Cd=cadmium; Age= time since the last anthropogenic action. 17 18 CS=constructed soil; WL=waste landfill soil; CP1=coking plant 1 soil; SP=settling pond soil; 19 CP2=coking plant 2 soil; CCS=constructed and contaminated soil. See Table 2 for the codes of 20 traits/ecological preferences and see Supplementary materials, Appendix A for Collembola species codes. 21 22 Figure 3: Community Weighted Mean (CWM) values of macrofauna communities sampled in each 23 derelict soil, calculated for the following traits: body length, diet type, integument type, mouth-part 24 25 type, habitat preference, and micro-habitat preference. Means (n=5). Different letters indicate significant differences among the 5 soils (P< 0.05) using Kruskal-Wallis tests followed by the Dunn's test of multiple comparisons with Bonferroni adjustment. CS=constructed soil; WL=waste landfill soil; CP1=coking plant 1 soil; SP=settling pond soil; CP2=coking plant 2 soil.

Figure 4: Projection of the coordinates of each soil replicate on the first two main components RLQ1 and RLQ2 (A), mapping of macrofauna taxon density coordinates (B), and plot of attributes of each trait linked to abiotic soil parameters (C). PAH=sum of the 16 PAH concentrations; Clay=proportion of clay; P= available phosphorous (Olsen method); OM=Organic Matter; WHC=Water Holding Capacity; CEC=Cation Exchange Capacity; C:N=carbon/nitrogen ratio; [Exchangeable cations by cobaltihexamine extraction] Ca=calcium; Na=sodium; K=potassium; Mg=magnesium. [Available elements] Pb=lead; Zn=zinc; Cd=cadmium; Age= time since the last anthropogenic action. CS=constructed soil; WL=waste landfill soil; CP1=coking plant 1 soil; SP=settling pond soil; CP2=coking plant 2 soil. See Table 2 for the codes of traits/ecological preferences and see Supplementary materials, Appendix B for macrofauna taxon codes.

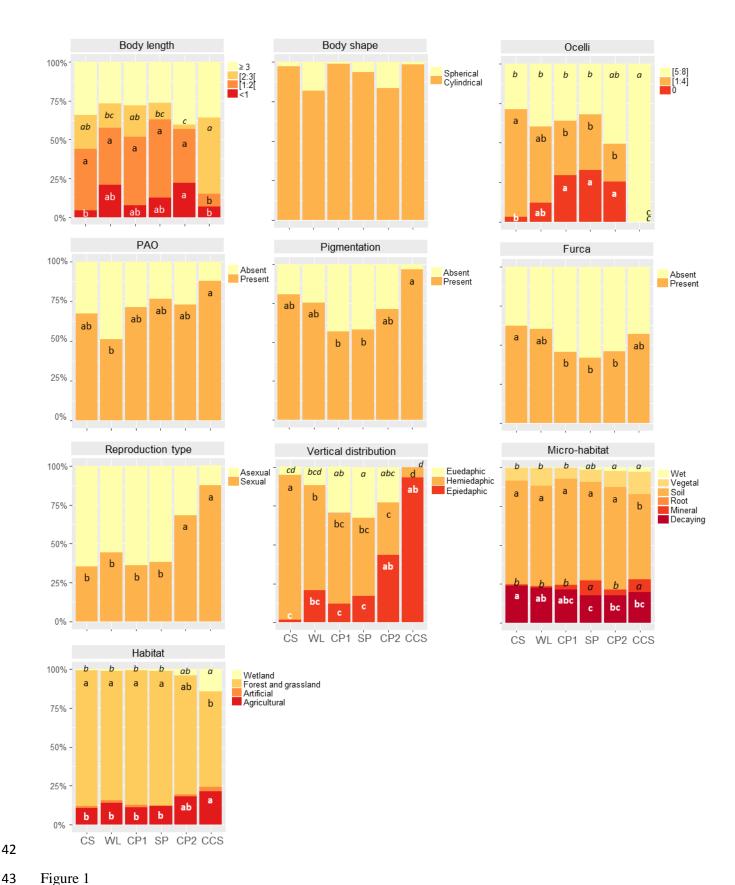
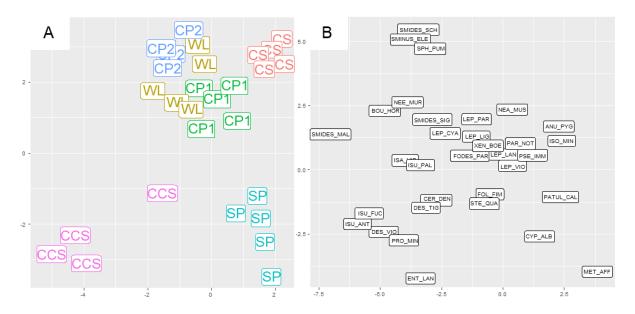
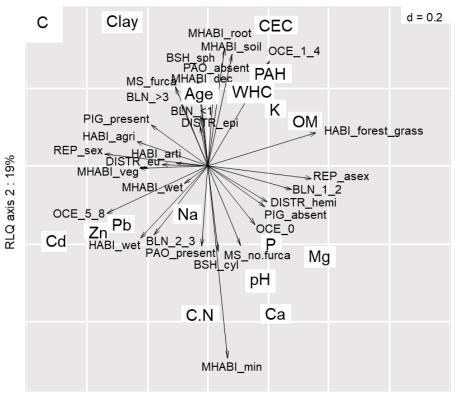


Figure 1





RLQ axis 1:69%

47 Figure 2

48

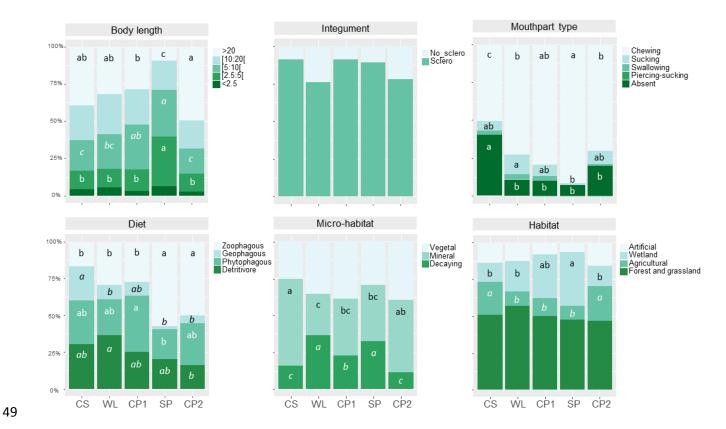
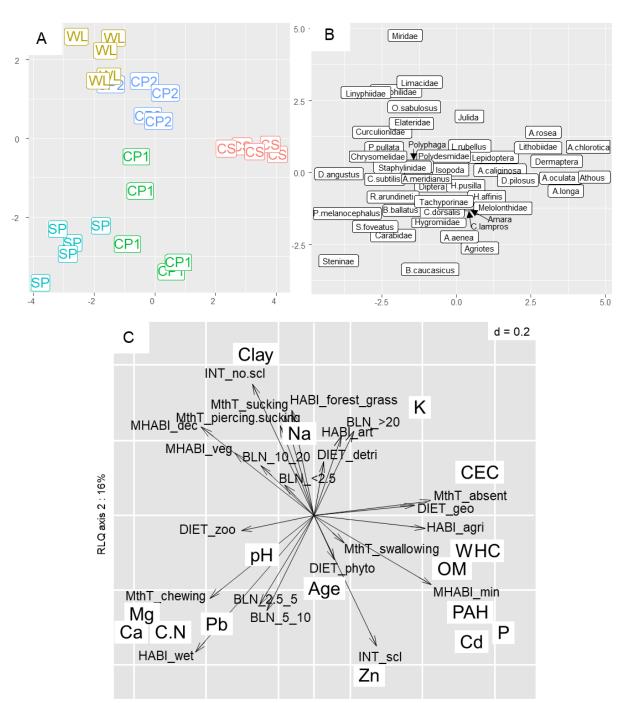


Figure 3



RLQ axis 1:71%

53

Figure 4