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► To cite this version:

Julien Jacquet, Emile Benizri, Guillaume Echevarria, Catherine Sirguyey. New insights on glass industry wasteland ecosystems. *Environmental Pollution*, 2022, 315, pp.120431. 10.1016/j.envpol.2022.120431 . hal-03874428

HAL Id: hal-03874428

<https://hal.science/hal-03874428v1>

Submitted on 19 Apr 2024

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1 **Environmental Pollution**

2 **New insights into wasteland ecosystems of the glass industry**

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6 *Keywords:* Industrial brownfield, Slag heap, Lead contamination, Soil fertility, Biodiversity

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9 **Abstract**

10 Over the past two centuries, large amounts of waste have been released into ecosystems from
11 glass manufacturing operations. Nowadays, an increasing number of abandoned slag heaps
12 results from the cessation of these activities and may be contaminated with metallic trace
13 elements (MTE) formerly used in glass and crystal products. However, recent findings
14 emphasized that derelict lands should also be recognized as a resource delivering a wide range
15 of ecosystem services. Therefore, the major purpose of this research was to provide new insights
16 into wasteland ecosystems of the glass industry; while assessing the relative influence of
17 edaphic factors on biotic communities, which is still poorly understood although closely related
18 to the potential re-use of these sites. To that end, we investigated (i) Orthoptera, diurnal
19 Lepidoptera, plant communities, and (ii) abiotic parameters in the topsoils.

20 Multivariate analysis revealed strong correlations between potentially phytoavailable and
21 pseudo-total lead (Pb), cadmium (Cd) and zinc (Zn). Arsenic (As) also co-varied significantly
22 with DTPA-extractable Pb. Despite the severe Pb contamination reported in the topsoils,
23 wastelands of the glass industry were able to support a substantially higher than expected
24 biodiversity with orthopteran assemblages similar to grasslands and a diurnal Lepidoptera
25 diversity comparable to urban parks. We also noted a positive influence of plant richness on the
26 diurnal Lepidoptera community structure. Hence, Pb contamination does not appear to be a
27 primary limiting factor for the establishment of these communities regarding the low mobility
28 of Pb due to the high amount of organic matter recorded in the topsoils under subalkaline
29 conditions. We assumed that wasteland ecosystems of the glass industry were more affected by
30 soil fertility or land management practices. To conclude, these sites are able to provide
31 biodiversity ecosystem services, acting as wildlife sanctuaries for Orthoptera and diurnal
32 Lepidoptera, and strategic metals by phytoextraction in a circular economy model.

33 **1. Introduction**

34 With the closure of various industrial activities during the last century, many lands have
35 been left abandoned throughout the world (Bradshaw and Wong, 2003). In France, large
36 surfaces of brownfield sites have been actually recorded, which represents about 100,000 ha
37 (ADEME, 2016). These derelict soils may be contaminated in metallic trace elements (MTE)
38 due to former industrial activities (Tarradellas et al., 1996) like metal ore mining and smelting,
39 coking plants, glass factories (Dudka and Adriano, 1997). In France, 322,400 potentially
40 polluted areas were identified in the BASIAS database in July 2020 (BASIAS, 2021). These
41 areas can be enriched in various MTE such as arsenic (As), cadmium (Cd), copper (Co), lead
42 (Pb) and zinc (Zn) (Dudka et al., 1996; Jennings et al., 2002; Markiewicz-Patkowska et al.,
43 2005; Gallagher et al., 2008; Hartley et al., 2009; Luo et al., 2012). Vacant lands are also
44 characterized by strong spatial heterogeneities regarding their soil physico-chemical properties
45 (Morel et al., 2005). Nonetheless, industrial wastelands share common edaphic features such as
46 elevated pH values ($\text{pH} > 7$), high coarse fractions in soils ($> 2 \text{ mm}$) and may have high C/N
47 ratios due to a lack of N in organic matter (Morel et al., 2005). Indeed, these industrial substrates
48 can be depleted in nutrients (Dickinson, 2003).

49 Wastelands, like urban soils, are predominantly considered as surface areas to be
50 converted or a potential threat to ecosystems due to their properties and pollution levels
51 (Blanchart et al., 2019). In fact, the dispersal of soil particles in the atmosphere may directly
52 affect human health through inhalation of dust and skin absorption. MTE can also indirectly
53 damage organisms by contaminating the food through transfer to plants or metal leaching (Su
54 et al., 2014). However, brownfields are not only contaminated lands and they should be
55 recognized as a resource for land management (Morel et al., 2005; Lehmann and Stahr, 2007;
56 Anderson and Minor, 2017). Indeed these abandoned sites could deliver various ecosystem
57 services (ES) such as provisioning of biomass for the production of sustainable energy (Lord,

58 2015) or bio-sourced metals through the cultivation of various herbaceous or ligneous
59 hyperaccumulating plants (van der Ent et al., 2015; Corzo Remigio et al., 2020). On
60 contaminated derelict lands, phytoremediation coupled with biomass production could provide
61 new urban environments (e.g. green areas) that are attractive from a sustainable development
62 perspective (Wilschut et al., 2013).

63 At the same time, brownfields may record high levels of biodiversity (Bonthoux et al.,
64 2014; Vincent et al., 2018). Following a certain time of abandonment (since the last
65 anthropogenic actions), several vacant lands have shown a high degree of biodiversity with the
66 early colonization of spontaneous flora and fauna communities on a short time scale. In an
67 industrial settling pond managed by natural attenuation in Pompey (Lorraine, North-East
68 France), 20 to 50 years were required to recreate a well-forested area (Lucisine et al., 2015;
69 Huot et al., 2018). In the six derelict lands investigated by Vincent et al. (2018), 2 to 40 years
70 were necessary to initiate recolonization by different biotic communities (e.g. microbial,
71 mesofauna, macrofauna and plant components) that harbor a great biological diversity.
72 According to Bonthoux et al. (2014), species-rich habitats can spontaneously establish
73 themselves within a year to several decades in urban wastelands. In fact, some abandoned lands
74 could play a crucial role in biodiversity conservation for a wide range of biotic components. In
75 particular, several wastelands have demonstrated their ability to provide new habitats for
76 endangered terrestrial arthropod species in Europe (Gardiner et al., 2013), while at the same
77 time, a massive decline in terrestrial arthropod biodiversity was recorded in natural ecosystems
78 throughout all compartments of the food web (Hallmann et al., 2017; Seibold et al., 2019).
79 Further studies reported that some brownfields have the capacity to support a rich biodiversity
80 and to host rare species such as Coleoptera (Eyre et al., 2003; Small et al., 2006), soil mesofauna
81 (Joimel et al., 2017) and vascular flora (Robinson and Lundholm, 2012).

82 In order to give indications on the potential re-use of wastelands, recent studies have
83 focused on assessing the overall quality of these sites, not only in terms of pollution or soil
84 fertility, but also in terms of their biological diversity. Godefroid et al. (2007) examined the
85 influence of edaphic and microclimatic parameters on the community structure of vascular
86 plants in 22 wasteland sites (in Brussels, Belgium). The results revealed that plant species
87 assemblages were mostly governed by soil humidity, water pH, soil nutrient content and other
88 environmental variables such as light intensity. On the other hand, Dazy et al. (2009) showed
89 that Cd and Hg contaminations in soils were the main factors controlling the community
90 structure of the spontaneous vascular flora in a former coking plant site (Homécourt, France).
91 However, for other biotic components of brownfield ecosystems, Joimel et al. (2016, 2017)
92 suggested that land management practices had probably a stronger effect on soil mesofauna
93 diversity (e.g. Acari and Collembola) than edaphic parameters, such as MTE contamination or
94 soil fertility. For instance, these authors reported that most industrial wastelands harboured a
95 greater soil mesofauna diversity than in agricultural soils, despite the high concentrations in
96 MTE found in some of them. In a recent work performed on six derelict lands, Vincent et al.
97 (2018) revealed that the structure of microbial, soil fauna (e.g. mesofauna and macrofauna),
98 and plant communities were more closely related to soil fertility proxies than to other edaphic
99 parameters such as contamination in MTE or organic pollutants. Hence, the relative influence
100 of environmental factors (e.g. soil contaminations, soil fertility, land management practices) on
101 wasteland biodiversity is still poorly understood.

102 The aims of this study are to provide new insights into wasteland ecosystems of the glass
103 industry, which to our knowledge have never been studied. These derelict sites may include for
104 instance Pb and Zn oxides as they are respectively used to induce transparency and elasticity in
105 crystal products. Other elements can also enter into the composition of these materials for glass
106 coloring (e.g. cobalt (Co), erbium (Er), gold (Au), manganese (Mn), neodymium (Nd), nickel

107 (Ni), silver (Ag)). One sub-objective of this study is to assess the relative influence of soil
108 fertility and trace metal contamination on wasteland biodiversity of the glass industry. For that
109 purpose, several ecological inventories were performed on three distinct components of
110 brownfield ecosystems: (i) vascular plant, (ii) Orthoptera, and (iii) diurnal Lepidoptera
111 communities. According to the structure of fauna and flora communities, we also studied
112 edaphic parameters (fertility and trace metal contamination) and we explored the links between
113 abiotic variables. Then, we examined the relationship between plant assemblages, Orthoptera
114 and diurnal Lepidoptera communities using ecological indices for an integrated description of
115 spatial biodiversity patterns. Finally, we assessed the relative influence of soil fertility and trace
116 metal contamination on wasteland biodiversity of the glass industry.

117 **2. Methods**

118 *2.1. Site description*

119 According to the BASIAS database (BASIAS, 2021), from the 18th to the middle of the
120 20th century, the studied site was an industrial slag heap used as a dumping site for the waste of
121 the glass industry (including glass fragments, refractory materials, and coke combustion
122 residues) leading to the formation of a 10 m thick landfill. Until its closure in 1990, it received
123 Pb₃O₄, acid waste, Pb-rich neutralization sludge for acid effluents and crystal fragments from
124 the art glass industry. Over the last thirty years, a spontaneous vascular flora and a particular
125 entomofauna have colonized this wasteland, which contributed to recreate a well-vegetated
126 ecosystem (supplementary Fig. S1). Nowadays, this small artificial hill is covered by a 1200-
127 m² herbaceous layer at its top (about 60 m long and 20 m wide), surrounded by forested slopes.
128 Since the 90's, this environment has been maintained semi-open by basic land management
129 practices: some trees were pruned and a small path was partially mowed once a year in order to
130 access to the top of the hill. The dumping site lies on clayed-marly formations of the Dogger
131 (Middle Jurassic marly limestones) overlaid by a superficial layer of alluvia made of calcareous
132 sands and gravels. The studied site is localized in the North-East of France (Lorraine), a region
133 with a climate under semi-continental influence. The mean annual temperature is 10.1°C with
134 an average temperature range of 1.8°C in January to 18.8°C in July. The mean annual
135 precipitation is 713 mm per year.

136 *2.2. Sampling design for biotic components*

137 Several inventories were carried out on the herbaceous layer, located at the top of the
138 hill. For vegetation sampling, a field study was conducted on July 2019. The identification of
139 plant taxa was carried out down to the species level. Each species was classified according to
140 its habitat preferences (inventory in supplementary Tab. S1). For the entomofauna sampling,

141 an inventory of Orthoptera and diurnal Lepidoptera communities was performed during two
142 days in July and August 2019 (inventory in supplementary Tab. S2). The month of July was
143 particularly dry with an average temperature of 21.1°C (temperature range: 8.4°C - 39.6°C) and
144 a cumulative rainfall of 20.8 mm. The month of August was slightly more humid with an
145 average temperature of 20.6°C (temperature range: 8.7°C - 34°C) and a cumulative rainfall of
146 58.4 mm. Orthoptera and diurnal Lepidoptera were collected with nets for species identification
147 and abundances were assessed by semi-abundance coefficients for all species.

148 *2.3. Topsoil sampling and analyses*

149 Fifteen soil samples were taken from the topsoil layer (0-10 cm without the plant litter),
150 in July 2019. Soil chemical analyses were conducted using French standard methods at the
151 Laboratoire Sols et Environnement (Université de Lorraine, Vandoeuvre-lès-Nancy) on 2 mm-
152 sieved and air-dried samples. Pseudo-total concentrations of MTE were evaluated after aqua
153 regia soil digestion (NF ISO 11464 standard). Five hundred mg of crushed soil (at 250 µm)
154 were digested for 16 h in 6 mL HCl and 2 mL HNO₃ at room temperature. Then, mineralization
155 took place in a DigiPREP system during 3 hours at 105°C. After cooling down, the solutions
156 were diluted to 50 mL with ultrapure water and then filtered under vacuum at 0.45 µm before
157 analysis on Inductively Coupled Plasma with Optical Emission Spectroscopy (ICP-OES, iCAP
158 6000 series, Thermo Scientific, Cambridge, UK). To study soil chemical fertility, several
159 parameters were determined. Total C and N contents were measured by dry combustion of
160 crushed soil (at 250 µm) and quantification of the emitted volume of CO₂ and N₂ (NF ISO
161 10694). Organic carbon (Org C) was assessed by dry combustion after carbonate dissolution
162 according to NF ISO 10694. To measure water pH, 5.0 mL of soils were added to 25 mL of
163 ultrapure water according to NF ISO 10390. Electrical conductivity was also determined in a
164 soil/water ratio 1:5 (v/v) according to NF ISO 11265. The effective cation exchange capacity
165 (CEC) and the extraction of the exchangeable cations were implemented according to the NF

166 ISO 31-130 standard. Exchangeable cations were then analyzed by ICP-OES, according to the
167 NF ISO 22036 standard. Available P was assessed by the Olsen method (NF ISO 11263). To
168 evaluate the potentially bio-available fraction of MTE, 5.0 g of soil were mixed with a 10 mL
169 DTPA (Diethylene Triamine Pentaacetic Acid)-based extraction solution at pH 7.3, according
170 to the NF ISO 13651 standard (solution prepared according to the NF ISO 31121 standard).
171 DTPA-extractable major and trace elements were quantified by ICP-OES. Quality control
172 assessment was done by inserting certified control soils as samples. To verify the validity of
173 the ICP-OES results, a blank and a control sample, as well as a certified solution (EU-H3, SCP
174 Science, Courtaboeuf, France) were inserted regularly into the batches to be analysed.

175 *2.4. Biodiversity assessment method*

176 Alpha diversity was determined on vascular plant, Orthoptera and diurnal Lepidoptera
177 communities with species richness (S), whereas species abundances were evaluated only for
178 Orthoptera and diurnal Lepidoptera communities. The following biodiversity indexes were
179 determined for diurnal Lepidoptera and Orthoptera communities.

180 The Shannon-Weaver diversity index (H') was computed according to the following
181 relation (Shannon and Weaver, 1963):

$$182 \quad H' = - \sum_i^S p_i \ln(p_i) \quad (1)$$

$$183 \quad \text{with } p_i = \frac{n_i}{N} \quad (2)$$

184 where \ln is the natural logarithm, n_i is the species abundance of the species “i” obtained from
185 the median value of the semi-abundance coefficients assigned for each species, N corresponds
186 to the species abundance of each community and S is the species richness index. Thus, H'
187 considers the species abundance in the assessment of biodiversity. The maximum value of H'

188 is reached when all individuals of each species are equally distributed. It is represented as:
189 $H'_{\max} = \ln(S)$. Hence H' is in the range $[0; \ln(S)]$. The Simpson diversity index (\tilde{D}) was
190 calculated according to the following formula (Grall and Coïc, 2006):

$$191 \quad \tilde{D} = 1 - \sum_i^S \frac{[n_i(n_i - 1)]}{[N(N-1)]} \quad (3)$$

192 This index may be interpreted as the probability that two randomly selected individuals belong
193 to different species (Grall and Coïc, 2006). Thus, \tilde{D} is in the range $[0; 1[$ and its value increases
194 with evenness given by the Pielou index (J) which is computed from the Shannon-Weaver
195 diversity index as following (Pielou, 1966):

$$196 \quad J = \frac{H'}{H'_{\max}} \quad (4)$$

197 Therefore, J is in the range $[0; 1]$ (from dominance of one of the species to even distribution).

198 2.5. Chemical risk assessment method

199 To assess the level of MTE contamination in the topsoils (0-10 cm), an integrated
200 chemical risk index was calculated using a Triad-like approach (Jensen and Mesman, 2006). In
201 this method, MTE pseudo-total concentration is compared to its respective soil screening level
202 (SSL). For each MTE, SSL was calculated as three times the local anomalous threshold (vibriss)
203 coming from Saby et al. (2018) dataset (Tab. S3). Only pseudo-total As, Cd, Cu, Pb and Zn
204 contents were included into this index because their values were in the range or above the local
205 threshold values. The effect of a global toxic pressure (PAF – Potentially Affected Fraction)
206 was computed using SSL as environmental quality criteria. This chemical risk index is in the
207 range $[0; 1]$ (1 was assigned for a maximum risk).

208

209

210 2.6. *Statistical analysis*

211 All tests were performed with R software v 4.1.1 (R Development Core Team, 2015).
212 In order to evaluate significant differences ($p < 0.05$) among edaphic parameters between the
213 three zones, one-way ANOVA was performed, followed by Tukey's HSD test from the
214 “multcomp” package in parametric conditions. The adequacy of the data with respect to the
215 ANOVA models was verified by examining the residuals for independence (Durbin-Watson
216 test, “car” package), normality (Shapiro test) and homogeneity (Levene test). If the latter
217 assumptions were not validated after “sqrt” or “log” transformation of the data, a Wilcoxon
218 pairwise test with a Holm correction of the p-value was used. The relationships between the
219 variables describing i) the soil fertility, ii) total soil element content and iii) available soil
220 element content were assessed through a multiple-factor analysis (MFA) followed by an
221 ascendant hierarchical classification (HCPC) with the “FactomineR” package. For the three
222 zones together, Spearman's correlation coefficients were calculated to determine the
223 relationships between the studied parameters. Results are presented as a heatmap with a
224 dendrogram added.

225 **3. Results**

226 *3.1. Vascular plant assemblages*

227 The vascular plant inventory was conducted on July 11th, 2019. The 1200-m² studied
228 area was delimited into three zones (Fig. 1) for several reasons: (i) the vegetation was rather
229 distinct between these areas with a denser plant cover observed in zone 3, the southern part of
230 the plot, (ii) zone 2 differed from zone 1 and zone 3 in terms of light intensity as it was shaded
231 by forested slopes, and (iii) a 200-m² mown area separated zone 1 from zone 2. Hence, the size
232 of the inventoried surface was reduced to 1000 m²: zone 1 covered an area of 350 m²; zone 2
233 represented 320 m²; and zone 3 was 330 m² in size (Fig. 1).

234 According to Tab. S1, 54 vascular plant species were identified in the 1000-m² studied
235 zone. The plant species composition was rather homogeneous with 30, 33, and 36 species
236 recorded respectively in zones 1 to 3 (Fig. 2). Half of the species was common to the three
237 zones revealing some uniformity in the assemblage of plant species on a site scale.
238 Nevertheless, more than a third of the species in zone 3 was not observed in zone 1 and zone 2,
239 and more than a quarter of the species in zone 2 was not reported in other parts of the area. By
240 contrast, only *Achillea millefolium*, *Linaria vulgaris* and *Torilis japonica* were endemic to zone
241 1 (Tab. S1). Regarding Fig. 2, zone 1 recorded the highest ratio of plant species (37 %) shared
242 between two zones: six species were shared with zone 2 and five species were shared with zone
243 3 (Tab. S1).

244 In terms of habitat preferences, Tab. 1 shows the distribution patterns of vascular plant
245 species for each zone. It was shown that grassland, wasteland and edge species particularly
246 dominate. The presence of seven bio-indicators of calcareous soils and seven woody plant
247 species should also be noted (Tab. S1). On a site scale, the plant species common to the three
248 zones were the most relevant bio-indicators for habitat characterization. According to Tab. S1,

249 ubiquitous species were: *Arrhenatherum elatius*, *Cirsium arvense*, *Dactylis glomerata*,
250 *Elytrigia repens* subsp. *repens*, *Ervilia hirsuta*, *Galium album*, *Hypericum perforatum* var.
251 *perforatum*, *Origanum vulgare* subsp. *vulgare*, *Pastinaca sativa* subsp. *urens*, *Plantago*
252 *lanceolata*, *Poa pratensis*, *Prunus spinosa*, *Trigonella alba* and *Urtica dioica*. Regarding these
253 14 species, wasteland and grassland species were still dominant followed by an edge species
254 prevalence. Two calcareous plant species (*Galium album* and *Origanum vulgare* subsp.
255 *vulgare*) and one thicket species (*Prunus spinosa*) were identified in the three zones (Tab. S1).

256 For each zone, the order of the main habitats followed the general trend described on a
257 site scale with a prevalence of wasteland, grassland, edge and calcareous plant species (Tab. 1).
258 However, some differences were observed between the three zones. First, zone 1 comprised the
259 highest number of grassland species and the smallest number of edge and thicket species.
260 According to Tab. S1, *Prunus spinosa* was the only shrub species inventoried in zone 1 while
261 zone 2 and zone 3 contained three thicket species each: *Cornus sanguinea*, *Hedera helix*,
262 *Prunus spinosa* and *Crataegus monogyna*, *Prunus spinosa*, *Rosa canina*, respectively. Then,
263 the largest number of calcareous plant species and the smallest number of wasteland species
264 were found in zone 2. Last, on the contrary, zone 3 was characterized by a dominance of
265 wasteland and edge species.

266 3.2. Spatial distribution and biological diversity of Orthoptera and diurnal Lepidoptera 267 communities

268 The first inventory began at 10:00 a.m. on July 16th, 2019. The weather was sunny with
269 no cloud cover. No precipitation fell that day and the wind was light (5 to 7 km h⁻¹).
270 Temperature was 20°C with 40% of moisture during the day. The second inventory began at
271 9:30 a.m. on August 22nd, 2019. The weather was sunny, and the wind was blowing at 8 km h⁻¹.
272 ¹. No precipitation fell that day. Temperature was 14°C with 75% of moisture during the day.
273 Both inventories were combined in Tab. S2.

274 On a site scale, 8 species of Orthoptera and 13 species of diurnal Lepidoptera were
275 recorded (Tab. S2). The number of butterflies was estimated to be at least 50 specimens. The
276 most abundant species were *Pieris rapae* and *Pyronia tithonus*. The number of grasshoppers
277 and crickets was estimated to be at least 35. The most abundant species were *Bicolorana bicolor*
278 and *Gomphocerippus rufus*.

279 Spatial patterns of Orthoptera and diurnal Lepidoptera communities are presented in
280 Fig. 3. Regarding Orthoptera (Fig. 3a), twice as many species and individuals were counted in
281 zone 1 and zone 2 compared with zone 3. According to Tab. S2, *Bicolorana bicolor*,
282 *Gomphocerippus rufus* and *Phaneroptera falcata* were common to the three zones. Zone 1 and
283 zone 2 comprised the same ratio of endemic species, whereas no endemic species were found
284 in zone 3. *Chrysochraon dispar* was only shared between zone 1 and zone 2. Regarding diurnal
285 Lepidoptera abundances (Fig. 3b), twice as many specimens were reported in zone 3 compared
286 to zone 1 and zone 2. The distribution of butterfly species was highly heterogeneous with six,
287 three and twelve species identified in zone 1, 2 and 3, respectively. According to Tab. S2,
288 *Melanargia galathea*, *Pieris rapae* and *Pyronia tithonus* were common to the three zones.
289 Almost 60% of the species recorded in zone 3 were not inventoried in zone 1 and zone 2. On
290 the contrary, no endemic species were found in zone 2. Only one species was endemic to zone
291 1: *Cupido argiades* (Tab. S2).

292 Further characterization of Orthoptera and diurnal Lepidoptera communities was done
293 with the calculation of several ecological indices to assess biological diversity (Tab. 2). On a
294 site scale, the Shannon-Weaver diversity (H') index was 1.70 for Orthoptera. The Simpson
295 diversity (\tilde{D}) and the Pielou evenness (J) indices reached elevated numbers with 0.79 and 0.82,
296 respectively. Regarding the Orthoptera diversity in the three zones, all ecological indices were
297 higher in zone 2. In particular, the Pielou index reached a high value in zone 2 (0.91) in
298 comparison with the one reported in zone 1 (0.68). Shannon-Weaver diversity (H') index

299 increased in the following order: zone 3 (0.81) < zone 1 (1.22) < zone 2 (1.62). A similar pattern
300 was observed for the Simpson diversity (\tilde{D}) index: zone 3 (0.52) < zone 1 (0.65) < zone 2 (0.81).
301 On a site scale, the Shannon-Weaver diversity (H') index was 2.01 for diurnal Lepidoptera. The
302 Simpson diversity (\tilde{D}) and the Pielou evenness (J) indices reached high values with 0.84 and
303 0.79, respectively. Regarding the diurnal Lepidoptera diversity in the three zones, all ecological
304 indices were higher in zone 3. A lower Pielou evenness (J) was computed in zone 1 (0.66)
305 compared to zone 3 (0.79). Zone 1 displayed an intermediate Shannon-Weaver diversity (H')
306 number (1.18) but much closer to the one determined in zone 2 (0.83) than in zone 3 (1.95). For
307 the Simpson diversity (\tilde{D}) index, a similar result was found with close values in zone 1 (0.59)
308 and in zone 2 (0.55), that were substantially lower than the one calculated in zone 3 (0.83).

309 *3.3. Abiotic parameters in the topsoils*

310 Each zone was characterized by five soil samples taken from the topsoil layer (0-10 cm).
311 Soil pseudo-total element composition is presented in Tab. 3. It was dominated by Ca, Fe, Al,
312 K, Mg and Pb (content exceeding 1 g kg⁻¹ dry soil). Significant differences between zones have
313 been only observed for Ca and Fe, with zone 2 having respectively highest and lowest values.
314 All the topsoils collected were highly contaminated with Pb and Cd, which pseudo-total
315 concentrations that were respectively at least 30 times and 2.5 to 4 times greater than the local
316 geochemical background (Tab. S3). In comparison with the local average soil composition, all
317 samples contained large amount of pseudo-total As, Cu and Zn. By contrast, all the topsoils
318 were strongly depleted in Co, Cr and Ni. Hence, the trace metal pollution in the topsoils was
319 largely dominated by Pb and Cd then, by a lower degree, by Cu, As and Zn. The concentration
320 of Pb in the topsoils was rather distinct between the three zones with values in the following
321 order: zone 3 (3.1 g kg⁻¹) > zone 1 (2.6 g kg⁻¹) > zone 2 (1.8 g kg⁻¹); even if values were not
322 significantly different. Zone 3 also displayed the most severe contamination in As (95.1 mg kg⁻¹;
323 ¹; $p < 0.05$) and Cd (5.7 mg kg⁻¹), these two elements behaving like Pb. On the contrary, the

324 highest Zn level was measured in zone 2 with 276 mg kg⁻¹ of dry soil. In zone 1, Cu
325 concentrations were close to the SSL limit value. Regarding the chemical risk index, the values
326 for the toxic pressure increased in the following order: zone 2 (0.940) < zone 3 (0.969) < zone
327 1 (0.973).

328 Regarding the parameters of soil fertility, no statistical differences have been observed
329 between the three zones (Tab. 4). Topsoils present a water-pH slightly alkaline (7.6-7.7) and a
330 mean electrical conductivity of 0.153 dS m⁻¹. The cation exchange capacity (CEC) ranges from
331 24.2 cmol⁺ kg⁻¹ in zone 3 to 27.1 cmol⁺ kg⁻¹ in zone 1, and is always saturated by Ca²⁺ with
332 Ca/CEC ranging from 0.83 to 0.90. Both K⁺ and Mg²⁺ represent each about 3.5 % of the CEC
333 with thus a K/Mg ratio near 1. The content of topsoils in organic carbon increases in the
334 following order: 6.6% in zone 3 < 7.7% in zone 2 < 8.1% in zone 1. As total nitrogen is rather
335 homogenous among the three zones, the C/N ratio decreased from 27.5 in zone 3 to 24.5 in
336 zone 1. Finally, the bio-available fraction of phosphorus is rather low with a mean value of 17.8
337 mg kg⁻¹.

338 Cobaltihexammine (CEC-exchangeable) and DTPA extractions were used for
339 respectively assess the actual and potential MTE availability (Tab. 4). Regardless of the zone
340 and the element considered, CEC-exchangeable concentrations were all below the analytical
341 detection limits. Concerning the potential bioavailability of MTE, significant differences
342 between the three zones were only found for Fe. For Pb and Cd, DTPA-extractable content in
343 zone 2 was respectively about and 1.5 and 1.3 times lower than that in zone 1 and zone 3. For
344 Fe, Mn and Zn, an opposite pattern was found with DTPA concentrations about 1.3 (Mn) and
345 1.5 (Fe, Zn) times higher in zone 2 compared to zones 1 and 3. Cu and Ni have a similar pattern
346 with the highest DTPA-extractable content in zone 1. At the site level, the percentage of the
347 pseudo-total content that was extracted by DTPA decreased in the following order: 45% (Pb),
348 27% (Cd), 14% (Cu), 9.2% (Zn), 2.3% (Ni), 1.9% (Mn), 0.1% (Fe). It should be noted that the

349 distribution of DTPA-extractable Cd, Pb and Zn contents in the topsoils of the three zones (Tab.
350 4) followed the same trend as that described by their pseudo-total concentrations (Tab. 3).

351 *3.4. Relationships between abiotic parameters in the topsoils*

352 The first MFA axis explained 29.1% of the variance and was mainly explained by both
353 groups of variables determined as available and total elements soil contents (Fig S2). Thus, this
354 axis was positively correlated with total As and Pb and DTPA-extractable Pb for the element
355 content parameters and CEC and exchangeable K and Mg for the fertility parameters (Fig. 4A).
356 Only element content parameters (DTPA-extractable Cu, Fe and Mn, and total Mg and Mn)
357 were negatively correlated to the first axis. This axis allowed a separation of soil samples along
358 a gradient of both As and Pb pollution and soil exchange properties. This is confirmed by the
359 ascendant hierarchical classification that allowed to separate the soil samples in four clusters of
360 which clusters 1 and 4 are the most correlated with axis 1 (Fig. 4C). Indeed, cluster 1 was
361 characterised by lower average levels of total Pb (1471 mg kg^{-1}) and DTPA-extractable Pb (629
362 mg kg^{-1}) and total As (50.2 mg kg^{-1}) than the overall average. Similarly, DTPA-extractable Cd
363 had a lower average content than the general average (0.654 mg kg^{-1}). On the other hand, cluster
364 3 was characterised by average contents above the general average of total Pb (3248 mg kg^{-1})
365 and DTPA-extractable Pb (1456 mg kg^{-1}) and total As (101.7 mg kg^{-1}). Cluster 4 was
366 characterised by higher levels of total Cu. The second MFA axis explained 18.6% of variance.
367 It was positively correlated with both soil fertility (exchangeable P and Ca and total N) and
368 element content (total S, Na, Mg, Zn, and B and DTPA-extractable Mn and Zn) parameters
369 (Fig. S2 and Fig. 4A). This axis allowed a separation of soil samples along a gradient of Zn
370 pollution and P and N fertility. Thus, cluster 2 was characterised by higher average contents of
371 total Zn (364 mg kg^{-1}), DTPA-extractable Zn (39 mg kg^{-1}), and P Olsen (24.59 mg kg^{-1}) than
372 the general average (Fig. 4C). When comparing the representation of individuals on the factorial
373 map according to their zone or cluster of origin (Fig. 4B and 4C) we did not observe a full

374 match. However, zone 3 was mostly characterized by cluster 3, whereas zone 2 was mainly
375 characterized by both clusters 1 and 2.

376 The heatmap of the Spearman correlation coefficients between soil parameters (Fig. S3)
377 highlighted that DTPA-extractable metal concentrations were significantly positively
378 correlated to their respective pseudo-total concentration. Pseudo-total As and Pb and DTPA-
379 extractable Pb presented high significant positive correlations with each other ($R > 0.9$) but
380 significant negative correlations with DTPA-extractable Fe and Mn and total Ca ($-0.71 < R < -$
381 0.5). DTPA-exchangeable Pb was also significantly positively correlated with CEC-
382 exchangeable K and Mg and total K ($0.5 < R < 0.58$). Pseudo-total and DTPA-exchangeable
383 Zn were significantly positively correlated with P Olsen ($0.57 < R < 0.64$) and total P ($0.35 <$
384 $R < 0.49$), whereas pseudo-total and DTPA-exchangeable Cd were significantly negatively
385 correlated with DTPA-extractable Fe ($-0.62 < R < -0.56$).

386

387 **4. Discussion**

388 *4.1. Biodiversity assessment of vascular plant, Orthoptera and diurnal Lepidoptera*
389 *communities in wastelands of the glass industry*

390 In the 1000-m² studied site, vascular plant assemblages were dominated by wasteland,
391 grassland and edge species. On the one hand, with 54 identified species, we observed about half
392 the vascular plant species inventoried in three calcareous grasslands (between 96 and 116)
393 located in the northwestern Swiss Jura mountains (Baur et al., 1996). However, these sites were
394 10 to 20 times larger (1-2 ha) and according to Bonthoux et al. (2014), the species richness
395 index increases with the size of a delimited area. Nonetheless, it should be noted that Melliger
396 et al. (2017) found that plant species richness did not increased with the size of grasslands
397 located in the urban region of Basel (Switzerland). Hence the plant diversity was considered to

398 be rather low compared to natural ecosystems such as calcareous meadows. On the other hand,
399 the species richness of vascular plants in the studied site was typical of the one recorded in
400 small clearings growing on a former settling pond managed by natural attenuation in Pompey
401 (Lorraine, North-East France) with more than 30 species identified (Lucisine et al., 2015).
402 According to Dazy et al. (2009), our results (54 species identified) were even slightly higher
403 than the ones reported in the vegetation of coking plants sites (27 to 46 species detected in seven
404 zones of the Homécourt wasteland).

405 Regarding orthopteran assemblages, our results (8 species identified) were quite
406 comparable to the species richness of grasshoppers and crickets harboured by 1-2 ha dry
407 grasslands in the Swiss Jura mountains (10 to 16 species; Baur et al., 1996) and 9-10 ha
408 meadows (10 to 14 species; Báldi and Kisbenedek, 1997). Melliger et al. (2017) reported that
409 Orthoptera species richness increased with habitat size in meadows (in the urban region of
410 Basel, Switzerland). Hence, the number of Orthopteran species recorded in the 1000-m² studied
411 was high given the size of the area. On the contrary, the species abundances found in the plot
412 (35 individuals) were much lower than the ones recorded (64 to 175 individuals) in Báldi and
413 Kisbenedek (1997). Nonetheless, the Shannon-Weaver diversity index (H') measured in our
414 study (1.70) was in the range of the ones calculated by Baur et al. (1996) for the three calcareous
415 grasslands (1.41 to 1.81). According to Báldi and Kisbenedek (1997), the Shannon index in the
416 wasteland site (1.70) was higher compared to those determined for one meadow (0.73 to 1.02)
417 and similar to the ones reported in the other meadow (1.71 to 2.07). In the studied site, the
418 Simpson diversity (\tilde{D}) and the Pielou evenness (J) indices (0.79 and 0.82, respectively) tend to
419 show a great heterogeneity in the community structure of Orthoptera. In addition, both
420 ecological indices were more elevated than the ones measured for the two meadows (0.17 to
421 0.72 for the Simpson diversity index and 0.32 to 0.79 for the Pielou evenness) in Báldi and
422 Kisbenedek (1997). The Pielou index (0.82) was in the range of the ones computed by Baur et

423 al. (1996) for the three 1-2 ha calcareous grasslands (0.70 to 0.86). Therefore, this wasteland
424 ecosystem must be considered as attractive for grasshoppers and crickets.

425 With 13 identified species, the assemblages of diurnal Lepidoptera remain different
426 from the ones found in less disturbed ecosystems such as typical calcareous grasslands wherein
427 the butterfly diversity reached 32 to 46 species in three sites of 1 to 2 ha in size (Baur et al.,
428 1996). In comparison with urban areas, our results (13 species identified) were in the range of
429 the species richness observed in eight parks of New York (7 to 22 species reported between
430 June and September 2002; Giuliano et al., 2004). The number of day-active butterfly species
431 was also more elevated than that observed in a business district and an office park near Palo
432 Alto, in California (3 and 7 species, respectively), but was similar to those identified in a
433 residential neighborhood (11 species), a golf course (12 species) and an open-space recreational
434 area in the same city (14 species; Blair and Launer, 1997). However, in the studied site, diurnal
435 Lepidoptera abundances (50 individuals) were below the ones measured in the residential
436 neighborhood (65 individuals), the golf course (60 individuals) and the open-space recreational
437 area (95 individuals) but considerably higher than that counted in the business district (8
438 individuals) and the office park (27 individuals; Blair and Launer, 1997). Nevertheless, the
439 Shannon-Weaver diversity index (H') determined in our study (2.0) was higher than those
440 calculated in the residential neighborhood (1.6) and in the golf course (1.8) but lower than that
441 reported in the open-space recreational site (2.2; Blair and Launer, 1997). Furthermore, the
442 Simpson diversity (\tilde{D}) and the Pielou evenness (J) indices (0.84 and 0.79, respectively) tend to
443 show a great heterogeneity in the community structure of diurnal Lepidoptera. To conclude, the
444 biodiversity of day-active butterflies in the 1000-m² wasteland ecosystem of the glass industry
445 may be considered as correct regarding the diurnal Lepidoptera diversity of moderately
446 undisturbed areas to highly developed urban landscapes. However, it is worth noting that

447 Orthoptera and diurnal Lepidoptera inventories were conducted between July and August, thus
448 comparison with other full-year studies is more difficult as spring species were not recorded.

449 *4.2. Spatial biodiversity patterns based on vascular plant and diurnal Lepidoptera communities*

450 In the studied site, zone 3 recorded the highest plant diversity, with more wasteland and
451 edge species, and displayed the greatest level of endemism (Fig. 2). Richness is obviously
452 connected with endemism, but the number of plant species does not vary enough between areas
453 to fully explain the specificity reported in that zone. Zone 3 also shown the highest day-active
454 lepidopteran and the lowest orthopteran biodiversity levels (Fig. 3). Conversely, more grassland
455 plant species were identified in zone 1, the area with the lowest plant species richness. This was
456 concomitant with a butterfly community structure characterized by a very low degree of
457 endemism and the lowest abundances, similarly to that observed in zone 2 (zone 1 and 2
458 harbored the lowest day-active Lepidoptera biodiversity). Such relationship between the species
459 richness in both vascular plants and lepidopteran is consistent with Erhardt (1985) who found
460 that species richness of diurnal Lepidoptera was often positively influenced by plant species
461 diversity in cultivated and derelict grasslands. In urban environments, Giuliano et al. (2004)
462 observed that plant species richness was also positively correlated with butterfly species
463 richness and abundances in eight parks of New York. The mechanisms behind these patterns
464 must be related to the pollination function of adult Lepidoptera in ecosystems (Hammond and
465 Miller, 1998). Indeed, some studies have observed an increase in butterfly species richness with
466 the abundance (Kuussaari et al., 2007) and diversity of nectar plant species (Kitahara et al.,
467 2008). In addition, the richness of nectar flower species was closely connected with plant
468 species richness (Kitahara et al., 2008). Hence, we assumed that day-active lepidopteran
469 biodiversity was clearly related to the plant species richness found in the studied site as many
470 plants were at the flowering stage during the vascular flora inventory, in particular in zone 3
471 (Fig. S1, southern part of the plot).

472 *4.3. Influence of trace metal contamination in the topsoils on the biodiversity of wastelands of*
473 *the glass industry*

474 In the studied site, Pb was a major element (content exceeding 1 g kg⁻¹) as reported in
475 the blast furnace sludge from a derelict land located in North-Rhine Westphalia, Germany
476 (Mansfeldt and Dohrmann, 2004). Pseudo-total Pb concentrations in our study were comparable
477 to the levels recorded in several topsoils contaminated by Pb-Zn smelting activities (810-1695
478 mg Pb kg⁻¹ and 2920-7894 mg Pb kg⁻¹ at site 1 and site 4, respectively, in Rabitsch, 1995; 140-
479 2600 mg Pb kg⁻¹ measured in five sites near two Pb-Zn smelters, studied by Stone et al., 2001).
480 Regarding the chemical risk index, each zone was highly contaminated with trace metal
481 elements: all topsoils were characterized by high pseudo-total Pb and Cd contents. These two
482 elements are considered to be toxic agents for plants: (i) vascular flora can be affected by Pb
483 when a level of 500 mg kg⁻¹ is reached in the soils and, (ii) Cd is known to affect most biological
484 components even at low concentrations (Kabata-Pendias and Pendias, 2001). For instance,
485 Dazy et al. (2009) reported that Cd contamination in soils was one of the main factors
486 influencing the community structure of plants in a former coking plant site (Homécourt, France)
487 as significant negative correlations were found between the plant species richness (S) and high
488 levels of Cd contamination (4 to 28 mg Cd kg⁻¹ in the most polluted areas).

489 Despite the high Pb and Cd pseudo-total contents reported in the topsoils, the wasteland
490 site of the glass industry was attractive for Orthoptera, and the biodiversity of diurnal
491 Lepidoptera was considered as attractive in the 1000-m² studied plot. Therefore, grasshoppers,
492 crickets, and day-active butterflies did not seem to be adversely affected by the high Pb and Cd
493 contamination in the topsoils. For the vascular flora, the diversity was considered to be rather
494 low on a site scale, suggesting an influence of Pb and Cd contamination in topsoils on the plant
495 species richness. However, regarding the distribution patterns of biodiversity in each zone, our
496 results were not consistent with this previous assumption. Firstly, zone 3 was the most

497 contaminated area with Pb and Cd although it recorded the greatest level of plant and diurnal
498 Lepidoptera biodiversity. Secondly, pseudo-total and available Pb concentrations in topsoils
499 and chemical risk indices were similar between zone 1 and zone 3, whereas the two zones
500 differed in terms of plant species richness, endemism and habitat preferences. Hence, trace
501 metal contamination in topsoils (Cd, Pb) does not appear to be a critical limiting factor for
502 vascular plant, Orthoptera and diurnal Lepidoptera biodiversity in this study.

503 Moreover, Pb often has a low bioavailability in soils, especially under alkaline
504 conditions (Kabata-Pendias and Pendias, 2001). In the superficial soils studied, DTPA-Pb
505 concentrations were high and negatively correlated with the total Ca content while the pH was
506 rather high (7.6-7.7). In the studied site, the high pH values are mainly reflected in negligible
507 CEC-exchangeable As, Cd, Cu, Pb and Zn contents compared to the major cations (Tab. 4).
508 Other authors have observed that a spontaneous forest can develop on former industrial sites
509 heavily contaminated with metals with a plant community comparable to that of similar natural
510 ecosystems (Lucisne et al., 2015) and a high diversity of meso- and macrofauna in the topsoil
511 layer (Huot et al., 2018). In comparison with this study, the Pb contents of the surface horizon
512 amounted to $> 3 \text{ g kg}^{-1}$ total Pb, 2060 mg kg^{-1} DTPA-exchangeable Pb and 0.1 mg kg^{-1} CEC-
513 exchangeable Pb (Huot 2013; Huot et al., 2018). It has been shown in urban soils that Pb was
514 mainly found in stable forms such as organic complexes, thus limiting its bioavailability (Ge et
515 al., 2000). Therefore, the high total and DTPA-exchangeable Pb contents of the studied surface
516 soils probably have little influence on the ecological recovery of glass industry wastelands.

517 *4.4. Influence of topsoil fertility on the biodiversity of wastelands of the glass industry*

518 In the studied site, the topsoils displayed high organic carbon contents regarding
519 grasslands (2.5%) and soils of urban, industrial, traffic, mining and military areas (SUITMA)
520 (3.5- 4.5%; Joimel et al., 2016; Mao et al., 2014). However, our results were of the same order
521 of magnitude as those measured in coking plant soils (7-10 %; Biache et al., 2008). All C:N

522 ratios were elevated compared to C:N values from typical meadows (around 10; Ponge et al.,
523 2013; Stauffer et al., 2014), thus corresponding to moder-like non-biologically active types of
524 humus that are commonly found in forests on non-calcareous soils. High C:N ratios can be
525 related to low N contents. In the studied topsoils, our results were similar to the values recorded
526 for coking plant soils (Biache et al., 2008) and typical of the high ratios found in industrial soil
527 substances, suggesting a technogenic origin for organic matter (Morel et al., 2005). Soil
528 electrical conductivity was rather low, suggesting that topsoils were not affected by salinity, a
529 problem often encountered on brownfield sites. Indeed, the topsoils were strongly depleted in
530 pseudo-total Na and K, when compared to the average compositions of surface soils (Sposito,
531 1989). CEC was almost in the range of values found in some urban park or garden soils (32
532 $\text{cmol}^+ \text{kg}^{-1}$; Santorufo et al., 2014) or even the same as reported in meadows ($35 \text{ cmol}^+ \text{kg}^{-1}$;
533 Stauffer et al., 2014). With a slightly alkaline pH and a high CEC saturated by calcium, the
534 studied site shared common features with natural carbonated soils as reported by Huot et al.
535 (2013) for the topsoils of a settling pond developing on iron industry by-products.
536 Alkalinization might be induced by the presence of gravel and concrete in the studied materials
537 (Morel et al., 2005; Rokia et al., 2014). From the point of view of agronomic soil fertility, the
538 studied soils presented high CEC-exchangeable K and Mg contents (Schvartz et al., 2005).
539 However, Olsen P concentrations were very low according to SUTMA's (45 mg kg^{-1}) and
540 grasslands soils (31 mg kg^{-1} ; Joimel et al., 2016). To conclude, as measured in this study, the
541 topsoils showed a medium fertility in comparison with other soils.

542 Regarding the distribution patterns of biodiversity in each zone, great differences were
543 noted in the structure of Orthoptera and diurnal Lepidoptera communities and in plant
544 endemism whereas topsoil fertility was similar between the three zones. However, in detail,
545 zone 3 displayed the lowest chemical fertility in the topsoils although it recorded the greatest
546 level of plant and diurnal Lepidoptera biodiversity. In fact, plant species richness was probably

547 related to the low amounts of N and available P reported in the topsoils of zone 3, limiting the
548 prevalence of some species (Heinze et al., 2015). We also noted a possible influence of the
549 calcareous status of topsoils on the plant species composition in the studied site. Our results in
550 zone 2 are consistent with this previous assumption as the area recorded the largest number of
551 calcareous plant species while pseudo-total Ca contents were more elevated in that zone. The
552 calcareous status and nutrient contents (total N and P Olsen) in topsoils have probably an impact
553 on plant assemblages (species richness and habitat preferences), and thus an indirect influence
554 on the diurnal Lepidoptera biodiversity. Thus, biodiversity could be more influenced by soil
555 fertility parameters than by trace metal contamination in the studied topsoils, as reported by
556 Vincent et al. (2018) for several derelict soils.

557 Nonetheless, as measured in this study, topsoil fertility proxies are probably not the most
558 impacting variables for plant, Orthoptera and diurnal Lepidoptera biodiversity. Other
559 environmental factors could have an influence on wasteland biodiversity of the glass industry.
560 On the one hand, the plot was surrounded by well-forested borders that may be involved in the
561 spontaneous colonization of the site by vascular flora and insect communities. These wooded
562 areas could also protect diurnal Lepidoptera from the wind, leading to the rather high diversity
563 reported in the investigation zone. On the other hand, biodiversity levels could be explained by
564 the poor land management practices carried out on the plot. For instance, Zerbe et al. (2003)
565 also reported a higher plant species richness in less disturbed areas and Erhardt (1985) indicated
566 that day-active Lepidoptera diversity was greater in undisturbed meadows than in intensively
567 managed grasslands.

568 **5. Conclusion**

569 In our study, wastelands of the glass industry displayed a higher biological quality than
570 expected regarding the high Pb and Cd contamination reported in the topsoils. Indeed, the 1000-

571 m² studied plot was attractive for Orthoptera and the biodiversity of diurnal Lepidoptera was
572 correct. Thus, several ecosystem services were already provided by wastelands of the glass
573 industry, giving habitats for some terrestrial arthropods communities that harbor a great
574 Orthoptera diversity. According to the distribution patterns of biodiversity and topsoil
575 contamination in each zone, the results highlighted that the most contaminated area recorded
576 the greatest level of plant and diurnal Lepidoptera biodiversity. This can be accounted for by
577 the low bioavailability of Pb under sub-alkaline conditions and the positive influence of plant
578 species richness on diurnal Lepidoptera biodiversity. Hence, trace metal contamination in the
579 topsoils does not appear to be a main limiting factor for vascular plant, Orthoptera and diurnal
580 Lepidoptera communities. Our results showed that wastelands of the glass industry had a
581 medium chemical fertility, in accordance with the biodiversity levels recorded in this study.
582 Moreover, the least fertile area was characterized by the highest plant and day-active
583 Lepidoptera diversity, suggesting a positive effect of soil nutrient depletion (N, P) on plant
584 species richness. Therefore, we assumed that biodiversity in wastelands of the glass industry
585 was more influenced by soil fertility parameters than by trace metal contamination in the
586 topsoils. To conclude, these sites are able to provide biodiversity ecosystem services, acting as
587 wildlife sanctuaries for Orthoptera and diurnal Lepidoptera, and strategic metals by
588 phytoextraction in a circular economy model.

589 **6. Acknowledgements**

590 This work was carried out within the framework of the SOLAMET project, supported
591 by the ANRT convention Cifre n° 2019/1926 and ADEME convention n° 1972C0020. The
592 vascular plant inventory was conducted with the help of Johanna Bonassi, a botanical expert of
593 the Pôle Lorrain du Futur Conservatoire Botanique National Nord-Est (CBNNE). The data
594 collected were then integrated into the database of the botanical conservatory. The Orthoptera
595 and diurnal Lepidoptera inventory was performed with the help of Anne Vallet, an entomologist

596 expert collaborating with the Société Lorraine d'Entomologie (SLE). The authors wish to thank
597 Econick and the technical staff of the LSE.

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806 **Tables**

807 **Tab. 1.** Spatial distribution of vascular plant species regarding their habitat preferences.
 808 Habitats represented by only one species were not mentioned. The numbers were computed
 809 from the data listed in Tab. S1.

HABITAT PREFERENCES OF SPECIES	ZONE 1	ZONE 2	ZONE 3	SITE
Mesophilic grasslands	9	7	7	11
Wastelands and ruderal vegetation	8	6	10	11
Edges - calcareous edges and nitrophilic hems	5	6	7	10
Calcareous edges and calcareous lawns	4	6	4	7
Thickets	1	3	3	5
Crops	1	2	3	4

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811 **Tab. 2.** Shannon-Weaver diversity (H'), Simpson diversity (\tilde{D}) and Pielou evenness (J) indices
 812 determined in each zone for Orthoptera and diurnal Lepidoptera communities, from Tab. S2.

ECOLOGICAL INDICES	ZONE 1	ZONE 2	ZONE 3	SITE
ORTHOPTERA				
Shannon-Weaver index (H')	1.22	1.62	0.81	1.70
Simpson diversity index (\tilde{D})	0.65	0.81	0.52	0.79
Pielou evenness index (J)	0.68	0.91	0.74	0.82
LEPIDOPTERA				
Shannon-Weaver index (H')	1.18	0.83	1.95	2.01
Simpson diversity index (\tilde{D})	0.59	0.55	0.83	0.84
Pielou evenness index (J)	0.66	0.75	0.79	0.79

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821 **Tab. 3.** Pseudo-total concentrations for major and trace elements in the topsoils of the three
 822 zones. Elements were presented in descending order according to their average concentrations
 823 (\pm standard deviation, n=5). Five soil samples were analyzed for each zone. Anova results are
 824 presented as F value with the level of significance (with *, ** and *** symbols representing P
 825 ≤ 0.05 , 0.01 and 0.001, respectively).

Pseudo-total elements	ZONE 1	ZONE 2	ZONE 3	ANOVA
<i>Major elements (content exceeding 1 g kg⁻¹) (in g kg⁻¹)</i>				
Ca	109 \pm 16.2	159 \pm 16.3	117 \pm 25.2	9.14 **
Fe	16.9 \pm 1.2	13.8 \pm 1.6	16.2 \pm 1.5	6.39 *
Al	15.3 \pm 1.2	14.8 \pm 1.4	15.1 \pm 1.1	0.15
K	4.0 \pm 0.3	3.7 \pm 0.7	4.0 \pm 0.5	0.44
Mg	3.8 \pm 0.1	4.0 \pm 0.2	3.8 \pm 0.5	0.60
Pb	2.6 \pm 1.1	1.8 \pm 0.4	3.1 \pm 1.4	2.33
<i>Minor elements (content ranging from 1 to 1000 mg kg⁻¹) (in mg kg⁻¹)</i>				
P	760 \pm 97.6	797 \pm 121	735 \pm 139	0.33
S	705 \pm 60.1	847 \pm 77.1	701 \pm 173	2.63
Mn	369 \pm 63.4	523 \pm 98.6	455 \pm 276	0.99
Na	335 \pm 49.1	389 \pm 95.1	335 \pm 97.5	0.69
Zn	233 \pm 75.8	276 \pm 137	223 \pm 54.8	0.43
As	81.5 \pm 20.8	53.4 \pm 12.4	95.1 \pm 33.2	4.00 *
Cu	86.7 \pm 47.0	55.2 \pm 28.6	44.4 \pm 17.3	2.17
Cr	26.8 \pm 2.9	24.6 \pm 3.1	26.8 \pm 2.9	0.92
Ni	28.1 \pm 4.9	23.1 \pm 5.1	25.1 \pm 7.7	0.90
B	22.7 \pm 4.2	21.9 \pm 3.6	21.6 \pm 3.0	0.11
Co	7.8 \pm 0.6	6.1 \pm 0.5	7.0 \pm 0.9	8.11 **
Cd	4.1 \pm 2.0	3.8 \pm 0.3	5.7 \pm 4.7	0.59

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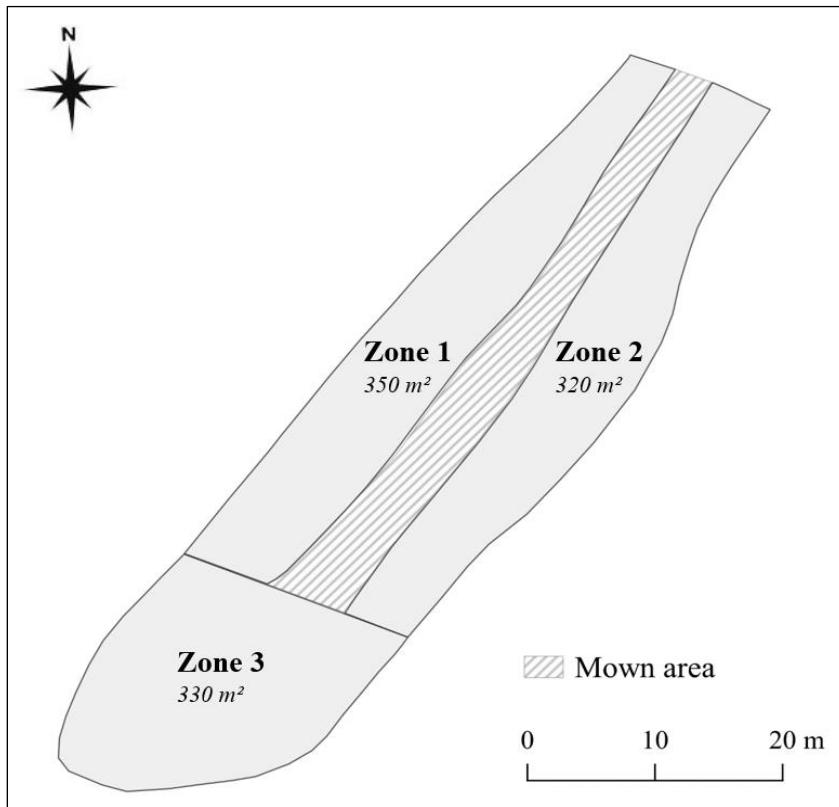
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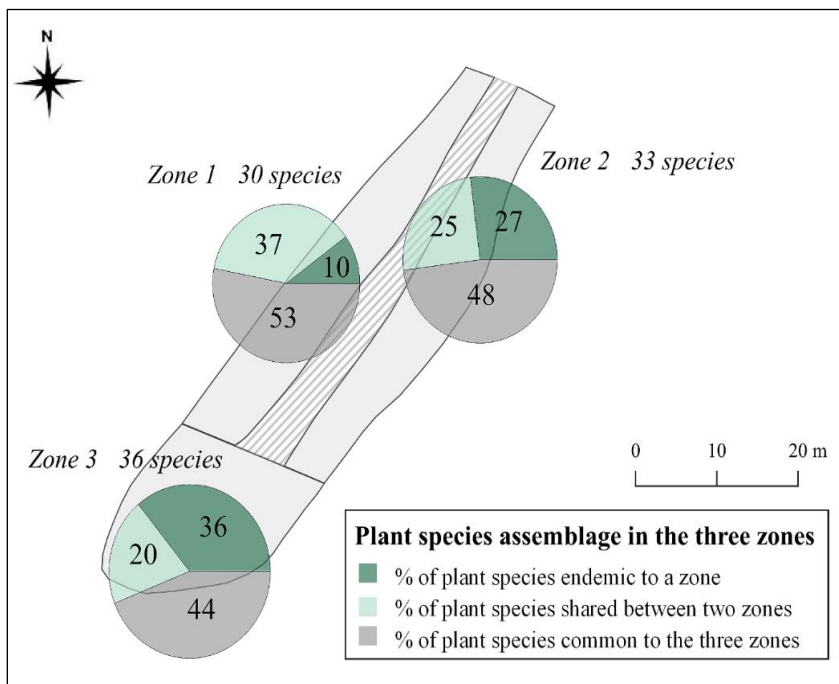
833 **Tab. 4.** Distribution of soil fertility parameters and bioavailability of trace metal elements in
 834 the topsoils of the three zones. EC = electric conductivity. S = total amount of exchangeable
 835 cations. LQ = quantification limit. Values are given in means (\pm standard deviation). Five
 836 samples were analyzed for each zone. Anova results are presented as F value with the level of
 837 significance (with *, ** and *** symbols representing $P \leq 0.05$, 0.01 and 0.001, respectively).

Topsoil parameters	ZONE 1	ZONE 2	ZONE 3	ANOVA
Org C (%)	8.0 \pm 1.9	7.1 \pm 1.1	6.6 \pm 1.4	1.18
Total N (%)	0.41 \pm 0.06	0.40 \pm 0.09	0.33 \pm 0.05	1.00
C/N	24.5 \pm 5.2	25.6 \pm 4.3	27.5 \pm 5.8	0.36
pH water	7.69 \pm 0.07	7.64 \pm 0.14	7.65 \pm 0.13	0.31
EC (in dS m ⁻¹)	0.147 \pm 0.023	0.155 \pm 0.011	0.156 \pm 0.009	0.50
P _{olsen} (mg kg ⁻¹)	17.5 \pm 5.1	18.7 \pm 7.1	17.1 \pm 3.1	0.12
<i>Cation exchange capacity (CEC) and exchangeable cations (in cmol⁺ kg⁻¹)</i>				
CEC	27.1 \pm 3.7	25.5 \pm 2.2	24.2 \pm 1.0	0.51
Ca _{exch}	23.2 \pm 2.9	22.9 \pm 2.9	20.1 \pm 1.0	2.47
Mg _{exch}	0.95 \pm 0.28	0.84 \pm 0.17	0.93 \pm 0.17	0.40
K _{exch}	0.88 \pm 0.27	0.78 \pm 0.13	1.06 \pm 0.18	2.35
Na _{exch}	<LQ	<LQ	<LQ	
S/CEC	0.92 \pm 0.01	0.96 \pm 0.05	0.91 \pm 0.03	0.34
<i>Other exchangeable elements (Al, Fe, Mn, As, Cd, Cr, Cu, Ni, Pb, Zn)</i>				
	<LQ	<LQ	<LQ	
<i>DTPA-extractable element (mg kg⁻¹)</i>				
Pb	1242 \pm 509	797 \pm 216	1335 \pm 527	2.12
Zn	20 \pm 6.6	30 \pm 15.4	18 \pm 5.1	1.98
Fe	18.5 \pm 6.0	23.5 \pm 4.2	14.1 \pm 4.3	4.58 *
Cu	10.4 \pm 4.3	6.5 \pm 1.0	7.6 \pm 4.4	1.51
Mn	8.0 \pm 2.0	9.7 \pm 2.4	7.1 \pm 2.8	1.47
Cd	1.3 \pm 0.7	1.0 \pm 0.2	1.4 \pm 1.2	0.45
Ni	0.68 \pm 0.13	0.56 \pm 0.09	0.50 \pm 0.12	2.90
Co	<LQ	<LQ	<LQ	
Cr	<LQ	<LQ	<LQ	



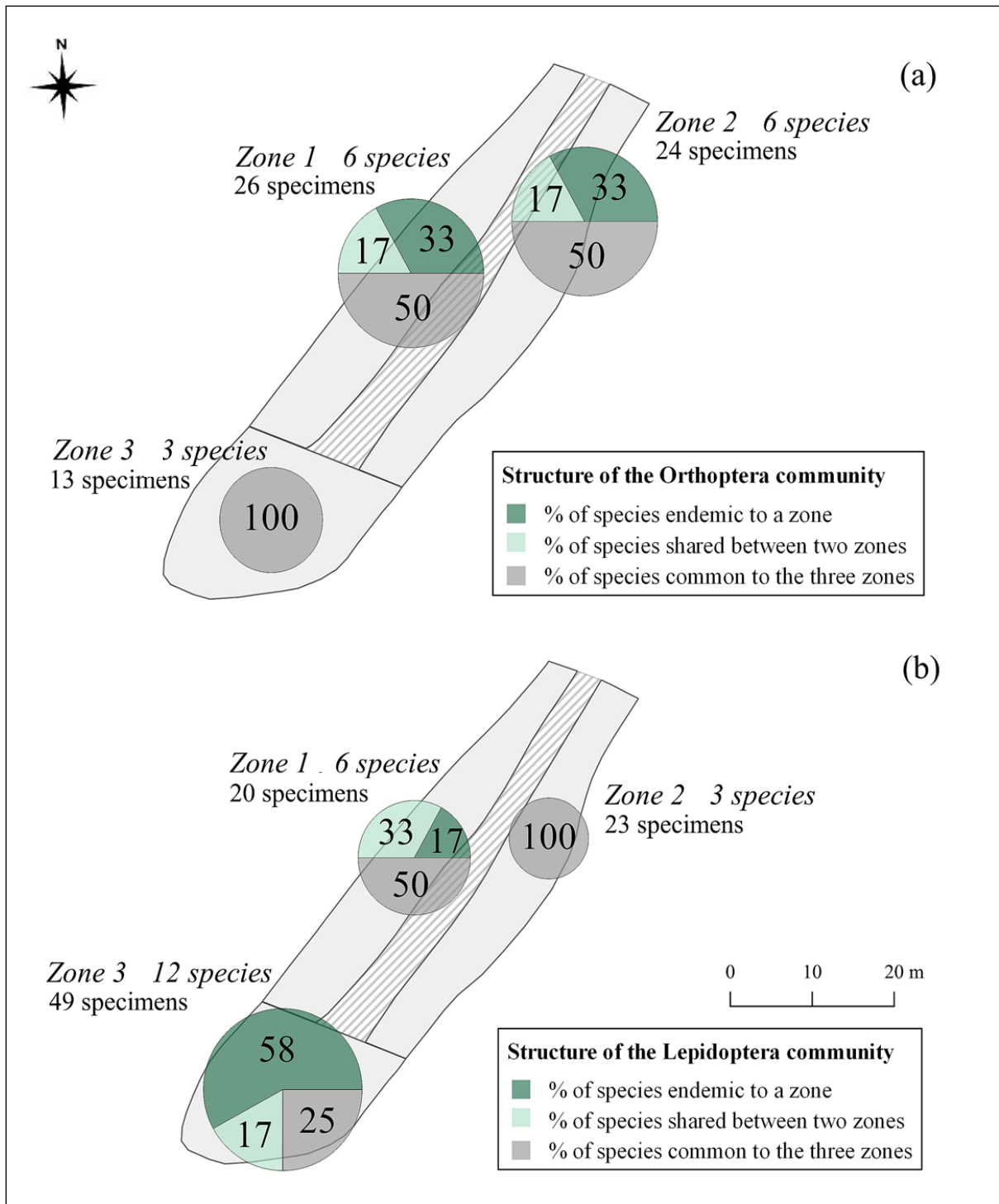
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841 **Fig. 1.** Subdivision of the 1000-m² studied area into three zones.



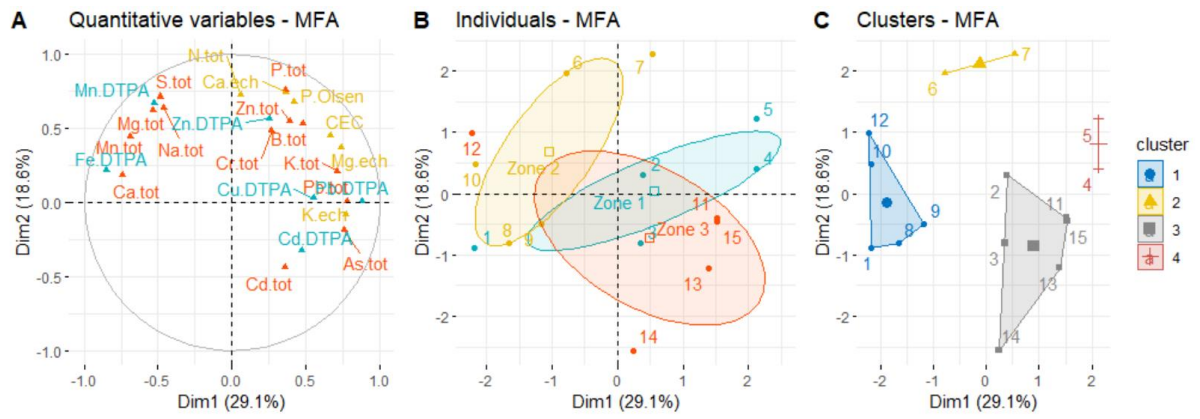
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843 **Fig. 2.** Plant species assemblage in the three zones. Species richness (S) and the ratios were
 844 computed from the data listed in Tab. S1.



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846 **Fig. 3.** Differences in the community structure of Orthoptera (a) and diurnal Lepidoptera (b)
 847 reported between the three zones. Species richness (S), abundances (A) and the ratios were
 848 computed from the data listed in Tab. S2.



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850 **Fig. 4.** Multiple-factor analysis (MFA) performed on abiotic parameters in the topsoils. A.
 851 Correlation circle of variables. B and C. Representation of individuals on the factorial map
 852 according to their zone or cluster of origin.