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1 **Title: Plant biodiversity offsets negative effects of metals and metalloids soil multi-**  
2 **contamination on ecosystem multifunctionality**

3

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51 **Abstract**

52 Despite increasing metals and metalloids (MM) human-driven soil contamination, how it  
53 simultaneously alters biodiversity and ecosystem functioning remains unknown. We used a  
54 wide gradient of a 170-year-old MM soil multi-contamination in Mediterranean scrublands to  
55 assess the effects of soil multi-contamination on multiple plant biodiversity facets, microbial  
56 communities and ecosystem multifunctionality (EMF). We found an overall positive effect of  
57 plant biodiversity on EMF mediated by microbial communities, and allowing to offset the  
58 negative impacts of MM soil multi-contamination, especially on soil water holding capacity  
59 and nitrogen content. The diversity of distant plant lineages was the key facet promoting EMF  
60 by enhancing microbial communities, whereas the subordinate species richness altered EMF.  
61 By developing a holistic approach of these complex relationships between soil multi-  
62 contamination, plant biodiversity, microbial communities and ecosystem functioning, our  
63 results reveal the potential of plant biodiversity, and especially the diversity of evolutionary  
64 distant species, to offset the alteration of ecosystem functioning by MM soil multi-  
65 contamination. In this worldwide decade of ecosystems restoration, our study helps to identify  
66 relevant facets of plant biodiversity promoting contaminated ecosystem functioning, which is  
67 crucial to guide and optimize management efforts aiming to restore ecosystems and preserve  
68 human health.

69

70 **Key words**

71 Inorganic pollution, phylogenetic diversity, soil-plant-microorganism continuum, species  
72 dominance, soil functioning

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## 76        **1. Introduction**

77            Human-driven metals and metalloids (MM) soil contaminations are a major threat to  
78 soil functioning by representing 34.9% of soils contaminants in the European Union, especially  
79 in the Mediterranean region (Panagos et al., 2013; van Liedekerke et al., 2014; Stolte et al.,  
80 2016; Merino et al., 2019). MM soil contamination could alter biodiversity and ecosystem  
81 functioning by impacting all the components of the soil-plant-microorganisms continuum, i.e.,  
82 by affecting soil composition and functioning (Greger, 2004; Petruzzelli et al., 2020), microbial  
83 activities (Kızılkaya et al., 2004; Gülser & Erdoğan, 2008) and plant physiological processes  
84 (Cheng, 2003; Yadav, 2010). Since biodiversity and ecosystem functioning are tightly coupled  
85 (e.g., Gamfeldt et al., 2008; Isbell et al., 2011), their relationships may vary along a MM soil  
86 multi-contamination gradient (see Steudel et al., 2012 for biodiversity-ecosystem functioning  
87 relationships along environmental stress gradients). Yet, how MM soil multi-contamination  
88 gradients simultaneously impact on taxonomic (species richness and their abundances within a  
89 community), phylogenetic (the diversity of evolutionary lineages within a community) and  
90 functional (the diversity of forms and functions within a community) facets of plant  
91 biodiversity, and, ultimately, on the capacity of ecosystems to perform multiple functions (i.e.,  
92 ecosystem multifunctionality (EMF); Manning et al., 2018) remains unknown (Fig. 1).  
93 Deciphering the relationships between biodiversity and EMF under soil multi-contamination  
94 could not only expand our fundamental understanding of the biodiversity-EMF relationships  
95 but could also help to identify the relevant biodiversity facets to improve management and  
96 restoration of contaminated ecosystems (Balvanera et al., 2014; Brum et al., 2017).

97            According to the pervasive environmental filtering hypothesis (Keddy, 1992; Weiher et  
98 al., 1998), MM soil multi-contamination can act as a strong environmental filter simultaneously  
99 decreasing phylogenetic, taxonomic, and functional diversity, and ultimately resulting in the  
100 reduction or loss of ecosystem functions (Fig. 1). MM soil multi-contamination can exacerbate

101 the effects of water stress in plants (Poschenrieder & Barcelo, 1999; Rucińska-Sobkowiak,  
102 2016). This could result in the selection of few plant evolutionary lineages, forms and functions  
103 tolerant to MM-induced water stress, (e.g., synthesis of osmoprotective compounds (Sharmila  
104 & Pardha Saradhi, 2002), reduction of leaf size, thickness, and stomata density (Rucińska-  
105 Sobkowiak, 2016)). However, these predictions contrast with the often-high diversity that can  
106 be observed in stressful and degraded environments, resulting from the co-occurrence of stress-  
107 avoidant and stress-tolerant plant species (Poorter et al., 2009; Gross et al., 2013), positive  
108 interactions (e.g., Butterfield & Briggs, 2011; Butterfield & Munson, 2016), or spatial /  
109 temporal storage effects (Chesson, 2000; Chesson et al., 2004). This discrepancy between  
110 patterns and predictions reminds the “functional paradox” of water-limited environments  
111 (Maestre et al., 2021) which could exhibit a low taxonomic, but remarkable plant phenotypic  
112 diversity to cope with the environmental constraints of these areas (Chesson et al., 2004; Gross  
113 et al., 2013; Gross et al., 2017; Le Bagousse- Pinguet et al., 2017). Thus, testing the  
114 environmental filtering hypothesis and its discrepancies on the multiple facets of plant  
115 biodiversity in contaminated environments is crucial to extend our understanding of the  
116 relationships between biodiversity and EMF, one of the key challenges for the next generation  
117 of functional ecology research (Steudel et al., 2012).

118 Different assembly processes (*sensu* Keddy, 1992) can shape the diversity patterns of  
119 dominant plant species with high abundance within a plant community, and subordinate plant  
120 species with low abundance within a plant community (Arnillas & Cadotte, 2019; Arnillas et  
121 al., 2021). For instance, while environmental filtering can lead to functional similarity (i.e.,  
122 reduced range of functional traits values) among dominant species, niche differentiation (i.e.,  
123 differentiated use of resources in time and space) can lead to greater divergence (i.e., wide range  
124 of functional traits values) among subordinate species, allowing them to coexist with dominant  
125 ones (Maire et al., 2012). In addition, subordinate species can better tolerate water stress than

126 dominant ones (Mariotte et al., 2017) and enhance community resistance against drought  
127 (Mariotte et al., 2013a). Finally, dominant and subordinate species can differently influence  
128 ecosystem functioning (Gross et al., 2017; Le Bagousse-Pinguet et al., 2019; Le Bagousse-  
129 Pinguet et al., 2021; Lyons et al., 2005). Even if dominant species highly influence individual  
130 functions related to biomass production, carbon, and nutrient cycling (mass ratio hypothesis;  
131 Grime, 1998), subordinate species can have significant effects on EMF (richness effect; Gross  
132 et al., 2017; Le Bagousse-Pinguet et al., 2019; Le Bagousse-Pinguet et al., 2021). Thereby,  
133 deciphering the diversity patterns of dominant *versus* subordinate species is critical to our  
134 understanding of their respective implications on ecosystem functioning (Seabloom et al.,  
135 2013).

136         Here we evaluated whether and how the relationships between biodiversity and EMF  
137 change along a wide gradient of a 170-year-old MM soil multi-contamination in Mediterranean  
138 scrubland communities. We investigated (i) how MM soil multi-contamination impacts  
139 taxonomic, phylogenetic and functional facets of plant biodiversity and EMF, (ii) how these  
140 three plant biodiversity facets affect EMF, either directly or mediated by soil microbial  
141 communities and (iii) whether dominant (mass-ratio effect) *versus* subordinate (richness effect)  
142 plant species reveal different patterns of biodiversity-EMF relationships. We assessed these  
143 relationships using structural equation modeling approaches based on an index of MM soil  
144 multi-contamination combining five different MM, seven complementary indices reflecting  
145 taxonomic, phylogenetic and functional biodiversity facets, an index of microbial activities and  
146 biomass based on four microbial functions, and an index of EMF based on five soil functions.  
147 We tested the core hypothesis that the environmental filtering exerted by MM soil multi-  
148 contamination selects for tolerant lineages and species with similar trait values within  
149 communities (i.e., reducing plant biodiversity), resulting in decreased microbial activities and  
150 biomass, and organic matter, nutrients, and water contents (i.e., reducing EMF, Fig. 1).

151

## 152 **2. Material and methods**

### 153 **2.1. Study area and sites**

154 We studied 30 scrubland communities located in the protected area of the Marseilleveyre  
155 massif (Calanques national park, Marseille, SE France), characterized by a semi-arid  
156 Mediterranean climate, calcareous, skeletal and oligotrophic soils. The studied communities  
157 correspond to young rosemary scrublands with at least one occurrence of *Rosmarinus officinalis*  
158 L., and one occurrence among four other characteristic species, i.e., *Cistus albidus* L., *Coronilla*  
159 *juncea* L., *Erica multiflora* L., and *Globularia alypum* L. (Pavon et al., 2018).

160 From the middle of the 19<sup>th</sup> century to the beginning of the 20<sup>th</sup> century, metallurgical  
161 and chemical industries generated massive amounts of MM-contaminated ashes in the  
162 Marseilleveyre massif (Daumalin & Raveux, 2016). MM dispersal by eolian and hydric erosion,  
163 influenced by topography, resulted in a diffuse, heterogenous and persistent MM soil multi-  
164 contamination (Laffont-Schwob et al., 2016). Soils are mainly contaminated by toxic elements  
165 as lead (Pb, from 110 to 29,412 mg.kg<sup>-1</sup>), arsenic (As, from 16 to 3,926 mg.kg<sup>-1</sup>), antimony (Sb,  
166 from 4 to 1,230 mg.kg<sup>-1</sup>), and essential elements for plant development as zinc (Zn, from 77 to  
167 6,420 mg.kg<sup>-1</sup>) and copper (Cu, from 15 to 52 mg.kg<sup>-1</sup>).

168 We chose to study scrubland communities, widely represented in the study area and  
169 more broadly in Mediterranean areas, and especially woody species because of their perennial  
170 and well-developed root systems subjected to MM soil multi-contamination. Thus, we first  
171 restricted the study area to areas potentially occupied by scrubland by selecting “323  
172 Sclerophyllous vegetation”, “333 Sparsely vegetated areas” and “332 Bare rock” land cover  
173 types from CORINE Land Cover 2012 database (available at:  
174 <https://www.data.gouv.fr/fr/datasets/corine-land-cover-occupation-des-sols-en-france/>).

175 Second, using a custom GIS procedure along with spatial databases, we selected areas with



176 scrubland vegetation characterized by homogeneous abiotic conditions influencing plant and  
177 microbial communities structure and functioning: distance to the sea, elevation, aspect, and  
178 slope (Heckenroth et al., 2016). The first step was to remove areas inferior to 250m to the sea  
179 to avoid the influence of sea dispersal spray on plant and microbial communities, using a 250m  
180 radius buffer area around the coastline provided by the Histolitt® database (available at:  
181 <https://www.data.gouv.fr/fr/datasets/trait-de-cote-histolitt-1/>). The second step was to calculate  
182 aspect and slope raster layers using a 5m resolution elevation raster layer provided by the  
183 RGEalti® database (available at: <https://geoservices.ign.fr/rgealti#telechargement5m>). Then,  
184 we checked for statistical associations between elevation, aspect and slope using principal  
185 components analysis and a correlation matrix. Since there was no relationship among these  
186 variables (Appendix A), we reduced our selection to the range between the 25<sup>th</sup> and 75<sup>th</sup>  
187 percentiles for each variable separately. As a result, our site selection encompasses sites from  
188 123 m to 283 m of elevation, from 23 % to 38 % of slope and south aspect. Then, we based our  
189 study sites selection on MM soil multi-contamination, using Marséco project gridded spatial  
190 data at a 250 m resolution (Laffont-Schwob et al., 2016). For each grid cell, corresponding to a  
191 measurement of Pb, As, Sb, Zn and Cu soil concentrations, a pollution load index (PLI) was  
192 calculated (Rashed, 2010; see below for calculation details) to evaluate the level of MM multi-  
193 contamination. Mapped MM contamination data relieved a spatially structured contamination  
194 gradient from West to East. These data are not used as value for the MM multi-contamination  
195 explanatory variable but only to support the choice of study sites localization by ensuring a  
196 representative sampling of the whole multi-contamination range. Then, we distinguished 4  
197 multi-contamination levels: very high ( $40 < \text{PLI} < 216$ ), high ( $10 < \text{PLI} < 30$ ), intermediate  
198 ( $5 < \text{PLI} < 10$ ) and low ( $1 < \text{PLI} < 5$ ) multi-contamination. We positioned 7 or 8 plots of 32 m<sup>2</sup> each  
199 within each multi-contamination level, in different patches of scrubland vegetation with a  
200 minimum distance of 70m between two plots to avoid pseudo replication (Fig. 2). The 32 m<sup>2</sup>

201 plot surface was estimated as the optimal surface to detect all woody species within a  
202 community, using the nested plot method.

203

## 204 **2.2. Soil analysis**

205 For each plot 5 soil subsamples of 200mL each, collected in the top 20cm-layer after litter  
206 removal and sieved at 2 mm, were pooled in a composite soil sample.

207

### 208 **2.2.1. MM soil multi-contamination**

209 Soils samples were dried at 40 °C, grounded less than 0.2 mm (RETSCH 200, agate mortar),  
210 digested in a microwave mineralizer (Milestone Start D) using aqua regia (1/3 HNO<sub>3</sub> + 2/3 HCl)  
211 and filtered through a 0.45 µm cellulose ester membrane filter (ISO 11466, 1995). Then, MM  
212 pseudo-total concentrations were determined by ICP-OES (Jobin Yvon Horiba, Spectra 2000).  
213 Quality controls and accuracy were checked using standard soil reference materials (CRM 049-  
214 050, from RTC-USA), with accuracies within 100 ± 10 %.

215 By using Pb, As, Sb, Zn and Cu pseudo-total concentrations, we calculated an index of MM  
216 soil multi-contamination, the PLI (Rashed, 2010; Table 1) for each plot:

$$217 \text{ PLI} = \sqrt[5]{\text{CF}_{\text{Pb}} \times \text{CF}_{\text{As}} \times \text{CF}_{\text{Sb}} \times \text{CF}_{\text{Zn}} \times \text{CF}_{\text{Cu}}} \quad \text{Eq. (1)}$$

218 where  $\text{CF}_{\text{MM}} = [\text{MM}]_{\text{pseudo-total}} / [\text{MM}]_{\text{local background value}}$  and  $[\text{MM}]_{\text{local background value}}$  are 42.9, 4.9,  
219 3.1, 66 and 7.5 mg.kg<sup>-1</sup> for Pb, As, Sb, Zn and Cu, respectively (Affholder et al., 2014).

220 Since calcareous Mediterranean soils are characterized by relatively high pH which can  
221 reduce MM bioavailability (Petruzelli et al., 2020; Fig. 1, H1), we measured soil pH  
222 (soil/osmosed water, 1/5, V/V; ISO 10390, 2005; Orion 2 Star Thermo Scientific SM30B), and  
223 the concentrations of potentially bioavailable MM using 0.05 M EDTA solution (pH = 7.0 ±  
224 0.1) as extractant with a 1/10 w/V ratio of soil/EDTA solution (Quevauviller, 1998; Appendix  
225 B). Concentrations of potentially bioavailable MM were strongly correlated with MM pseudo-

226 total concentrations and PLI (Appendix B). Therefore, we chose to keep MM pseudo-total  
227 concentrations because they allow us to calculate the index of MM soil multi-contamination  
228 (i.e., PLI) based on MM local background pseudo-total concentrations defined by Affholder et  
229 al. (2014). Because of their skewed distribution, PLI values used in analysis were log-  
230 transformed.

231

### 232 **2.2.2. Ecosystem multifunctionality**

233 We quantified EMF for each plot using five soil functions (Table 1): (i) total organic carbon  
234 (TOC), (ii) total Kjeldahl nitrogen (TKN), (iii) available phosphorus (P) contents, (iv) cation  
235 exchange capacity (CEC) and (v) water holding capacity (WHC).

236 Soil physicochemical analyses were carried out on 40 °C dried soils, excepted for the water  
237 holding capacity (WHC). Total organic carbon (TOC) content was determined by the difference  
238 between (i) the total carbon content, measured by non-dispersive infrared (NDIR) spectroscopy  
239 after soil combustion at 900 °C in a furnace under oxygen flow, and (ii) the inorganic carbon  
240 content, measured by NDIR spectroscopy after soil acidification with phosphoric acid (40 %  
241 H<sub>3</sub>PO<sub>4</sub>) at 80 °C under oxygen flow (AnalytiK Jena N/C 2100 S, ISO 10694, 1995). Total  
242 Kjeldahl nitrogen (TKN) content was determined by Kjeldahl method (ISO 11261, 1995): (i)  
243 mineralization of the soil sample by concentrated sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) during 4h at 300°C  
244 (Büchi Speed Digester K-436), (ii) distillation of the obtained ammonium sulfate with a  
245 concentrated soda solution during 7min and neutralization with an excess of sulfuric acid  
246 solution (Büchi Distillation Unit 323), (iii) dosage of the excess of sulfuric acid by indirect  
247 titration with soda solution (Titroline Easy, Schott Instruments). Available phosphorus (P,  
248 Olsen method, ISO 11263, 1995) and cation exchange capacity (CEC, Metson method, NF X  
249 31-130, 1999) measurements were performed by the Laboratoire Développement Méditerranée  
250 (COFRAC accreditation n°1-5865, Alès, France). WHC was measured by soil immersion in

251 water during 48h, followed by soil re-wiping and drying during 48 h at 90 °C. Soils were  
252 weighed after soil re-wiping and after drying. WHC correspond to the difference between soil  
253 mass after re-wiping and dry soil mass, divided by the dry soil mass. WHC represents the  
254 maximum mass of water absorbed per 100 g of dry soil.

255 Then, we Z-scored each individual function (i.e, TOC, NTK, P, CEC and WHC):

$$256 \quad Z \text{ score} = \frac{F_{ij} - \text{Mean } F_i}{SD F_i} \quad \text{Eq. (2)}$$

257 where  $F_{ij}$  is the value of the function  $i$  in the community  $j$ , and  $\text{Mean } F_i$  and  $SD F_i$  are  
258 respectively the mean and the standard deviation of the function  $F_i$ , calculated for the 30  
259 scrubland plots. Finally, we calculated an index of EMF by averaging the Z-score of individual  
260 functions for each plot.

261

### 262 **2.2.3. Microbial biomass and activities**

263 We quantified microbial biomass and activities for each plot using four microbial functions  
264 (Table 1): (i) soil basal respiration (SBR), (ii) microbial biomass (MB), (iii) average well color  
265 development (AWCD) and (iv) catabolic diversity ( $H_{\text{cat}}$ ).

266 SBR and substrate-induced respiration (SIR) were measured to assess global microbial  
267 activity and biomass (Anderson, 2003; Anderson & Domsch, 1978). 10 g dry weight equivalent  
268 of fresh soil at 30 % of the WHC were placed in 117 mL slightly open ajar glass jars and  
269 incubated for 48 h at 23 °C. For SBR measurement, each sample was oxygenated for 4 min and  
270 incubated for 4 h at 23 °C in tightly closed glass jars. Then, 1 mL of their atmosphere was  
271 sampled in the headspace with a syringe and injected into a gas chromatograph (Chrompack  
272 CHROM 3 – CP 9001) to analyse CO<sub>2</sub> production by microbial oxidation of the soil organic  
273 matter. SIR was estimated using a procedure from Anderson (2003). Soil samples were  
274 amended with 450 mg of talc and 50 mg of powdered anhydrous glucose to maximize the  
275 respiration rate and incubated for 2 h at 23 °C in slightly open ajar glass jars. Then, each sample

276 was oxygenated for 4 min and then incubated for 2 h at 23 °C in tightly closed glass jars. 1 mL  
277 of their atmosphere was sampled in the head space with a syringe and injected into a gas  
278 chromatograph to analyse CO<sub>2</sub> production by microbial oxidation of glucose. The gas  
279 chromatograph was equipped with a thermal conductivity detector and a packed column  
280 (Porapack). The carrier gas helium flow was regulated at 60 mL.h<sup>-1</sup>. Ambient CO<sub>2</sub>  
281 concentrations were subtracted from sampled CO<sub>2</sub> concentrations and resulting values were  
282 adjusted at 22 °C according to Ideal Gas Laws using a Q10 = 2. SIR was converted into  
283 microbial biomass (MB) using the relation established by Beare et al. (1990).

284 Community level physiological profiling (CLPP) of cultivable bacterial communities, i.e.,  
285 their potential to oxidize different C-substrates (Appendix C), was carried out using the  
286 Ecoplate microplates (Biolog, Hayward, CA, USA) according to a procedure adapted from  
287 Garland & Mills (1991). For each soil sample, 100 mL of 0.1 % sodium pyrophosphate sterile  
288 solution was added to 5 g dry weight equivalent of fresh soil in a 250 mL flask and the whole  
289 was shaken for 2 h at 80 rpm. The obtained soil solution was diluted and standardized at optical  
290 density (OD) 595 nm=0.03 with a sterile physiological solution (NaCl 0.85 %) and 125 µL were  
291 used to inoculate each well of the plates. After an incubation period of 5 days at 25 °C, microbial  
292 response was assessed by reading the absorbance at 590 nm using a TECAN®  
293 spectrophotometer (Tecan Trading AG, Switzerland). For each well, the absorbance value was  
294 blanked against the control well and negative absorbance values were set to zero. The minimum  
295 OD for a positive well was fixed at 0.1. The intensity of microbial C-use was assessed using  
296 the AWCD index:

$$297 \text{ AWCD} = \frac{\sum \text{OD}_i}{N} \quad \text{Eq. (3)}$$

298 where  $\text{OD}_i$  is the OD of the  $i^{\text{th}}$  well and  $N$  the number of wells in the plate.

299  $H_{\text{cat}}$  was assessed using Shannon's index:

$$300 H_{\text{cat}} = \sum_{i=1}^N (p_i \times \ln(p_i)) \quad \text{Eq. (4)}$$

301 where  $p_i$  is the ratio between the absorbance of the  $i^{th}$  well and the sum of the absorbances of  
302 all the wells of the plate.

303 Then, we Z-scored each individual function. Finally, we calculated an index of microbial  
304 biomass and activities by averaging the Z-score of individual functions for each plot.

305

306

## 307 **2.3. Plant biodiversity facets**

### 308 **2.3.1. Phylogenetic and taxonomic diversity**

309 In each plot, the presence of each species was recorded using a 2m<sup>2</sup> quadrat which was  
310 moved 16 times over the whole 32m<sup>2</sup> plot surface. 32 woody species, all native to the study  
311 area, have been recorded in total (Appendix D). According to the PNCal's regulations, two  
312 species were not included in our study because of their conservation status which prevent  
313 sampling: *Teucrium polium* L. and *Helianthemum syriacum* (Jacq.) Dum.Cours. The relative  
314 abundance of each species in each plot was calculated by dividing the number of quadrats in  
315 which it occurs by the sum of all species occurrences by quadrat.

316 We computed four phylogenetic diversity indices (*picante* R package, *mpd* and *mntd*  
317 functions; Kembel et al., 2010) for each plot using a phylogenetic tree (*V.PhyloMaker* R  
318 package, *phylo.maker* function, "S3" method; Jin & Qian, 2019) based on woody species  
319 occurrences: (i) weighted and (ii) non-weighted mean phylogenetic distance (respectively  
320 wMPD and MPD), which account for both closely related and distant evolutionary lineages,  
321 (iii) weighted and (iv) non-weighted mean nearest taxon distance (respectively wMNTD and  
322 MNTD), which represent the diversity of evolutionary lineages at the tips, i.e., focusing on  
323 closely related lineages. We calculated two taxonomic diversity indices (*vegan* R package;  
324 Oksanen et al., 2019) for each plot: (i) species richness and (ii) Pielou's evenness.

325

### 326        **2.3.2. Functional diversity**

327        We focused on three plant functional traits describing one of the major axes of plant form  
328        and function globally (Díaz et al., 2016), and that are key determinants of functional diversity  
329        and ecosystem functioning in Mediterranean scrublands (Gross et al., 2013; Valencia et al.,  
330        2015; Le Bagousse-Pinguet et al., 2015): Leaf Area (LA), Specific Leaf Area (SLA) and Leaf  
331        Dry Matter Content (LDMC). Traits such as LA are related to light interception and water stress  
332        tolerance (Westoby et al., 2002). SLA and LDMC are related to the leaf economic spectrum,  
333        reflects the relative growth rate of plants, and are associated with plant strategies to acquire,  
334        use and/or conserve resources such as light, nutrients and water (Wright et al., 2004).

335        In each plot, we sampled three mature individuals of each species at most to account for  
336        intraspecific functional trait variability (Albert et al., 2011), a key biotic component involved  
337        in species interactions (Kraft et al., 2014), community assembly and dynamics (Jung et al.,  
338        2010; Bolnick et al., 2011; Le Bagousse-Pinguet et al., 2014, 2015), and ecosystem processes  
339        (Fridley & Grime 2010). A total of 25089 leaves were sampled from 983 individuals. On each  
340        individual, a branch section with mature leaves was collected. We avoided sampling of leaves  
341        with herbivory or pathogen attack symptoms. Then, the samples were wrap in moist paper, put  
342        in closed plastic bags and stored at 4 °C before processing in the next 24 h (Pérez-Harguindeguy  
343        et al., 2016). The number of analyzed leaves per individual depended on the species considered.  
344        Preliminary tests allowed us to determine the minimum number of leaves to be analyzed so that  
345        the minimum LA per individual is 1 cm<sup>2</sup> (Appendix D). First, leaves were cut from the branch.  
346        We included or excluded the petiole depending on the species considered, based on the  
347        functional definition of a leaf, i.e., all leaf parts which are photosynthetic and then pigmented  
348        (Appendix D). Then, all the fresh leaves from a same individual were pooled, scanned, weighted  
349        (fresh mass), dried at 40°C until constant mass and reweighted (dry mass). The LA of each leaf  
350        was measured with Fiji (Schindelin et al., 2012). SLA was calculated per individual as the ratio

351 between the LA and the dry mass of all the leaves from a same individual. LDMC was  
352 calculated as the ratio between the fresh mass and the dry mass of all the leaves from a same  
353 individual.

354 We computed six functional indices (*FD* R package; Laliberté et al., 2014): the community-  
355 weighted mean of (i) LA (CWM LA), (ii) SLA (CWM SLA) and (iii) LDMC (CWM LDMC),  
356 and three multi-trait indices, i.e., (iv) functional richness (FRic), and (v) weighted and (iv) non-  
357 weighted functional dispersion (respectively wFDis and FDis). Because of their skewed  
358 distributions, functional trait values used in indices calculation were log-transformed.

359

## 360 **2.4. Data analyses**

### 361 **2.4.1. Conceptual model**

362 We developed an *a priori* conceptual model of the relationships among MM soil multi-  
363 contamination, biodiversity facets (phylogenetic, taxonomic, and functional diversity),  
364 microbial biomass and activities and EMF ecologically meaningful and framed from current  
365 knowledge (Fig. 1).

366

### 367 **2.4.2. Plant explanatory variables selection**

368 From this conceptual model, we avoided redundancy between plant explanatory variables  
369 by performing Pearson's correlation. If two variables were strongly correlated (Pearson  
370 coefficient  $> 0.70$  or  $< -0.70$ ), then only one of the 2 variables was retained (Appendix E). Based  
371 on this selection criterion, several associations of plant explanatory variables could have been  
372 retained for each biodiversity facet: (i) wMNTD and MPD or MNTD and wMPD for  
373 phylogenetic diversity, (ii) richness and evenness for taxonomic diversity, (iii) CWM LA,  
374 CWM SLA and FDis (including LA and SLA) or CWM LA, CWM SLA and FRic (including



375 LA and SLA), or CWM LA, CWM SLA, CWM LDMC, FRic and FDis (including LA, SLA  
376 and LDMC) for functional diversity.

377

### 378 **2.4.3. Best adequate initial model**

379 We tested all possible combinations of plant explanatory variables in 6 path analysis (2  
380 combinations for phylogenetic diversity x 1 combination for taxonomic diversity x 3  
381 combinations for functional diversity; *lavaan* R package; Rosseel, 2012), and kept the  
382 combination of variables that provided the best adequate initial model according to Grace  
383 (2006) and Rosseel (2012) criteria: Chi-squared test  $> 0.05$ , Comparative Fit Index (CFI)  $>$   
384  $0.90$ , Root Mean Squared Error (RMSEA)  $< 0.10$  and Standardized Root Mean Square Residual  
385 (SRMR)  $< 0.08$ . Our best adequate initial model retained included the following plant  
386 explanatory variables: (i) MNTD and wMPD for phylogenetic diversity, (ii) richness and  
387 evenness for taxonomic diversity, and (iii) CWM LA, CWM SLA and FDis for functional  
388 diversity (Fig. 3 and Table 1).

389

### 390 **2.4.4. Minimal parsimonious model**

391 From the best adequate initial model, we performed path analyses to assess: (i) the direct  
392 and indirect effects of MM soil multi-contamination on the plant biodiversity facets, on  
393 microbial biomass and activities and on EMF, (ii) the relationships between biodiversity facets,  
394 each of the soil individual functions (i.e., TOC, TKN, P, CEC, WHC), and ultimately EMF,  
395 and (iii) whether and how the relationships between plant biodiversity and EMF differ once  
396 considering either dominant or subordinate species within each community. We defined  
397 dominant and subordinate species within each plant community using a relative abundance  
398 threshold of  $0.06$ , based on the median of species relative abundances in all the 30 communities  
399 ( $0.06306$ ) and the median of median species abundances among the 30 plant communities

400 (0.06660). As a result, dominant species have a relative abundance between or equal to 0.06  
401 and 0.28 and subordinate species have a relative abundance between 0.007 and 0.06. The initial  
402 model was simplified by stepwise exclusion of non-significant variables, as estimated by AIC  
403 (Akaike Information Criterion), until that a minimal parsimonious model was reached. Finally,  
404 the adequacy of the minimal parsimonious model was evaluated by non-significant differences  
405 between predicted and observed covariance matrices ( $p$  of Chi-squared test  $> 0.05$ ), a high CFI  
406 ( $> 0.90$ ), a low RMSEA ( $< 0.10$ ) and a low SRMSR ( $< 0.08$ ) (Grace, 2006; Rosseel, 2012;  
407 Appendix F).

408

### 409 **3. Results**

#### 410 **3.1. Effects of MM soil multi-contamination on plant biodiversity facets and EMF**

411 The effects of MM soil multi-contamination on plant biodiversity facets and EMF were not  
412 influenced by the reduced range of soil pH values (7.88-8.38). MM soil multi-contamination  
413 did not affect TOC (Fig. 5a), P (Fig. 5b) and CEC (Fig. 5c), but it decreased WHC ( $r = -0.383$ ,  
414 Fig. 5d) and TKN ( $r = -0.319$ , Fig. 5e), resulting in a direct negative effect on EMF ( $r = -0.301$ ,  
415 Fig. 4a). In contrast, we did not observe any effect of MM soil multi-contamination on plant  
416 biodiversity (neither at the whole community level, nor when segregating between dominant  
417 and subordinate species) and on microbial biomass and activities.

418

#### 419 **3.2. Effects of plant biodiversity facets on EMF**

420 Plant biodiversity had an overall indirect positive effect on EMF ( $r = 0.253$ , Fig. 4a), TOC  
421 ( $r = 0.242$ , Fig. 5a), P ( $r = 0.278$ , Fig. 5b), CEC ( $r = 0.231$ , Fig. 5c) and WHC ( $r = 0.238$ , Fig.  
422 5d). Of the seven plant biodiversity indices considered in the initial model (Fig. 3), three  
423 significantly influenced EMF and soil individual functions, i.e., wMPD, CWM LA and species  
424 richness. Except for TKN, the significant effects of biodiversity facets on EMF and soil

425 individual functions were always mediated through the positive effect of microbial biomass and  
426 activities on EMF ( $r = 0.462$ , Fig. 4a), TOC ( $r = 0.443$ , Fig. 5a), P ( $r = 0.508$ , Fig. 5b), CEC ( $r$   
427  $= 0.423$ , Fig. 5c) and WHC ( $r = 0.435$ , Fig. 5d). wMPD and CWM LA had indirect positive  
428 effects on EMF ( $r = 0.291$  and  $r = 0.165$  respectively; Fig. 4a), TOC ( $r = 0.280$  and  $r = 0.159$   
429 respectively; Fig. 5a), P ( $r = 0.321$  and  $r = 0.182$  respectively; Fig. 5b), CEC ( $r = 0.267$  and  $r =$   
430  $0.151$  respectively; Fig. 5c) and WHC ( $r = 0.275$  and  $r = 0.156$  respectively; Fig. 5d), and  
431 wMPD always showed the strongest effect. In contrast, species richness had an indirect negative  
432 effect on EMF ( $r = -0.204$ , Fig. 4a), TOC ( $r = -0.196$ , Fig. 5a), P ( $r = -0.224$ , Fig. 5b), CEC ( $r$   
433  $= -0.187$ , Fig. 5c) and WHC ( $r = -0.192$ , Fig. 5d). The negative effects of species richness on  
434 EMF, TOC, P, CEC and WHC were compensated by the positive effects of wMPD and CWM  
435 LA. The overall indirect positive effect of plant biodiversity on EMF and WHC and the direct  
436 positive effect of CWM LA on TKN ( $r = 0.337$ , Fig. 5e) partially and fully offset the direct  
437 negative effects of MM soil multi-contamination, respectively.

438

### 439 **3.3. Effect of plant species relative abundance on biodiversity-EMF relationships**

440 Considering dominant and subordinate species separately highlighted different patterns of  
441 biodiversity-EMF relationships. MM soil multi-contamination had a direct and negative effect  
442 on EMF when focusing on dominant species ( $r = -0.303$ , Fig. 4b), while no effect occurred  
443 when considering subordinate species. wMPD is the only biodiversity index which had a  
444 significant positive effect on EMF mediated through microbial biomass and activities in both  
445 dominant ( $r = 0.261$ , Fig. 4b) and subordinate ( $r = 0.346$ , Fig. 4c) species. In dominant species,  
446 the positive effect of wMPD on EMF partially offset the negative effect of MM soil multi-  
447 contamination on EMF ( $r = -0.303$ , Fig. 4b). In addition, species richness had a significant  
448 negative effect on EMF mediated through microbial biomass and activities in subordinate  
449 species ( $r = -0.188$ , Fig. 4c), but no significant effect in dominant species. In subordinate

450 species, the indirect negative effect of species richness on EMF was compensated by the  
451 indirect positive effect of wMPD on EMF. Thus, plant biodiversity had an overall indirect  
452 positive effect on EMF in subordinate species ( $r = 0.158$ , Fig. 4c).

453

#### 454 **4. Discussion**

455 We investigated how MM soil multi-contamination influences the relationships between  
456 multiple plant biodiversity facets and EMF in Mediterranean scrublands. Our results highlight  
457 that plant biodiversity offset the negative impacts of MM soil multi-contamination on EMF  
458 (Fig. 4a), underlining its potential for ecological restoration of contaminated ecosystems. We  
459 also found antagonism between the positive effects of phylogenetic and functional facets and  
460 the negative effects of taxonomic facet on ecosystem functioning, warning for the need to  
461 consider multiple biodiversity facets for the restoration of contaminated ecosystems.

462 MM soil multi-contamination impacted differently the components of the soil-plant-  
463 microorganisms continuum. On the one hand, MM soil multi-contamination altered EMF (Fig.  
464 4a), and mostly soil water holding capacity (Fig. 5c) and total nitrogen content (Fig. 5d). The  
465 observed soil alteration likely results from competition for adsorption onto soil particles  
466 between toxic MM (e.g., lead and arsenic) and nutrients, resulting in changes in soil  
467 composition and functioning (Greger, 2004; Petruzzelli et al., 2020). On the other hand, MM  
468 soil multi-contamination did not impact plant biodiversity and microbial biomass and activities.  
469 Since MM and water stresses lead to similar physiological responses of plants (e.g., synthesis  
470 of osmoprotective compounds (Sharmila & Pardha Saradhi, 2002), reduction of leaves sizes,  
471 thickness, and stomata density (Rucińska-Sobkowiak, 2016)), our results support the  
472 environmental filtering hypothesis (Keddy, 1992; Weiher et al., 1998), i.e., the presence of  
473 tolerant species to water stress preadapted to MM contaminated soils (Affholder et al., 2014;  
474 Heckenroth et al., 2022; Salducci et al., 2019). Altogether our study highlights the complex

475 responses of degraded ecosystems to soil contaminations, and thus calls for the need of holistic  
476 approaches simultaneously considering the effects of contamination on above- and  
477 belowground biodiversity and soil functions to target the relevant components of the soil-plant-  
478 microorganisms continuum that restoration and management efforts should focus on.

479         Despite having both additive and antagonistic effects, depending on the facet  
480 considered, the overall effect of plant biodiversity on ecosystem functioning mediated through  
481 microbial communities was positive, allowing to partially offset the negative impacts of MM  
482 soil multi-contamination. Of the seven plant biodiversity indices under consideration (Fig. 3),  
483 three indices from phylogenetic, taxonomic and functional diversity influenced EMF (Fig. 4a)  
484 and soil individual functions (Fig. 5). wMPD and CWM LA had additive positive effects on  
485 ecosystem functioning whereas plant richness altered it. Increasing CWM LA enhance litter  
486 inputs, i.e., substrates supporting microbial activities (Lange et al., 2015; Chen et al., 2019),  
487 which can improve soil functioning (Le Bagousse-Pinguet et al., 2021). Numerous studies have  
488 reported that the coexistence of many species within plant communities (i.e., high plant  
489 richness) allows to support several functions simultaneously (e.g., Gamfeldt et al., 2008; Isbell  
490 et al., 2011; Maestre et al., 2012). However, our results, as other studies (e.g., Creed et al.,  
491 2009), reconsider this prevailing assumption of a positive relationship between plant richness  
492 and EMF because increasing number of plant species can also increase the probability of  
493 recruiting species which have negative effects on ecosystem functioning (selection effect;  
494 Tilman et al., 1997). Our results show that considering several facets of plant biodiversity is  
495 crucial to understand the complex overall effect of biodiversity on ecosystem functioning, and  
496 ultimately guide restoration efforts toward the relevant plant biodiversity facets to enhance  
497 ecosystem functioning.

498         Phylogenetic diversity was the key facet promoting EMF at all biological levels (i.e.,  
499 whole community, dominant and subordinate species; Fig. 4) and individual functions (i.e.,

500 total organic carbon, available phosphorus, cation exchange capacity, and water holding  
501 capacity; respectively Fig. 5a, b, c and d) through microbial communities. Phylogenetic  
502 diversity can effectively encompass hidden biological traits (Flynn et al., 2011), such as those  
503 involved in root associations with rhizobia (Werner et al., 2015) and mycorrhizal fungi  
504 (Brundrett, 2002; Brundrett & Tedersoo, 2017) which highly influence soil functioning  
505 (Bardgett et al., 2014) and enhance MM plant tolerance (Gamalero et al., 2009) For example,  
506 *C. juncea* is a leguminous species (Fabaceae) in association with nitrogen-fixing bacteria  
507 (Robles et al., 2002; Heckenroth et al., 2022), and mycorrhizal fungi (Carrasco et al., 2011).  
508 These root associations can improve soil total nitrogen and bioavailable phosphorus contents,  
509 enhancing plants mineral nutrition and growth on MM contaminated soils. Since wMPD  
510 (accounting for both closely related and distant lineages) had a strong effect on ecosystem  
511 functioning, contrary to MNTD (biased towards closely related lineages), our result indicates  
512 that the effect of very distinct plant lineages diversity was particularly important, as reported  
513 for EMF in global drylands (Le Bagousse-Pinguet et al., 2019). This could be explained by the  
514 very ancient evolutionary history of plant-microorganisms associations (Brundrett, 2002;  
515 Brundrett & Tedersoo, 2017; Sprent, 2007). Thus, the coexistence of evolutionary distinct  
516 lineages within plant communities could ensure a high functional diversity, allowing for  
517 differentiated resources use in time and space (i.e., niche differentiation), and thus the  
518 maintenance of several ecosystem functions simultaneously (Gamfeldt et al., 2008; Isbell et al.,  
519 2011). Our results highlight the importance of considering plant phylogenetic diversity, in  
520 particular distant lineages, in restoration of contaminated ecosystems, not only to better  
521 understand MM tolerance mechanisms but also because the co-occurrence of distant lineages  
522 provides an important source of biodiversity to offset the negative effects of contamination on  
523 ecosystem functioning.

524 The relationships between MM soil multi-contamination, biodiversity and EMF  
525 strongly differed between dominant and subordinate species within a given community. We  
526 found that EMF was impacted by MM soil multi-contamination but not by plant richness in  
527 dominant species (Fig. 4b) and *vice versa* in subordinate species (Fig. 4c). Several studies have  
528 reported that subordinate species can enhance microbial communities and soil functions  
529 (Mariotte et al., 2013b; Peltzer et al., 2009). However, we found that subordinate species  
530 richness altered EMF, potentially due to allelopathic effects (i.e., plant-plant or plant-  
531 microorganisms biochemical interactions) of a pool of subordinate species: *Ruta angustifolia*  
532 Pers., *Helichrysum stoechas* (L.) Moench, *Lavandula latifolia* Medik. and *Cephalaria*  
533 *leucantha* (L.) Schrad. Ex Roem. & Schult. (known allelopathic effects of closely related  
534 species, respectively: *R. graveolens* (Hale et al., 2004), *H. italicum* (Mastelic et al., 2005), *L.*  
535 *officinalis* (Akbarzadeh et al., 2013) and *C. syriaca* (Ali et al., 2012)). In addition, comparison  
536 of plant diversity between communities in dominant *versus* subordinate species (i.e., beta  
537 diversity; Appendix G) indicate that (i) communities shared a common dominant species pool  
538 and (ii) differences in species composition among communities was mainly driven by  
539 subordinate species. As a result, the effect of plant biodiversity on EMF was mainly driven by  
540 the diversity of subordinate species. Our results highlight the importance of considering  
541 subordinate species in restoration programs because they can strongly influence EMF (richness  
542 effect; Gross et al., 2017; Le Bagousse-Pinguet et al., 2019; Le Bagousse-Pinguet et al., 2021),  
543 including by enhancing community resistance to drought (Mariotte et al., 2013a). This is  
544 particularly important in Mediterranean ecosystems where water stress is accentuated by MM  
545 (Poschenrieder & Barcelo, 1999; Rucińska-Sobkowiak, 2016) and droughts will become more  
546 and more intense and frequent in the context of global warming (Cramer et al., 2018).

547

548

549 **5. Conclusion**

550 Our results reveal the potential of plant biodiversity, and especially the diversity of  
551 evolutionary distant species, to offset the alteration of ecosystem functioning by MM soil multi-  
552 contamination. These findings require the use of holistic approaches considering soil-plant-  
553 microorganisms continuum, multiple plant biodiversity facets, and plant species relative  
554 abundance. In this worldwide decade of ecosystems restoration (FAO, IUCN CEM, SER,  
555 2021), our study helps to identify relevant facets of plant biodiversity promoting contaminated  
556 ecosystem functioning, which is crucial to guide and optimize management efforts aiming to  
557 restore ecosystems without removing MM contaminated soils and preserve living soils and  
558 human health.

559

560 **Data availability**

561 Dataset and R codes are available from Figshare  
562 (<https://figshare.com/s/fd5a224d980758f2d049>) and will be made public if the manuscript is  
563 accepted.

564

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573



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921 **Figure legend**

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923 **Figure 1.** Conceptual model and associated hypotheses based on scientific literature and  
924 assessing the relationships between metals and metalloids soil multi-contamination,  
925 biodiversity facets (phylogenetic, taxonomic, and functional diversity), microbial biomass and  
926 activities and ecosystem multifunctionality in the 30 Mediterranean scrubland communities.  
927 The dashed arrows represent the theoretical relationships.

928

929 **Figure 2.** Location of the 30 scrubland plots (represented by circles) along a wide gradient of  
930 a 170-year-old MM soil multi-contamination (evaluated by the pollution load index including  
931 lead, arsenic, antimony, zinc and copper) due to former industrial activities (represented by  
932 triangles) in the currently protected area of the Marseillevyre massif (Calanques national park,  
933 Marseille, SE, France).

934

935 **Figure 3.** The best adequate initial model resulting from variables selection and assessing the  
936 relationships between metals and metalloids soil multi-contamination, biodiversity facets  
937 (phylogenetic, taxonomic, and functional diversity), microbial biomass and activities and  
938 ecosystem multifunctionality in the 30 Mediterranean scrubland communities. The dashed  
939 arrows represent the potential relationships.

940

941 **Figure 4.** Path analysis of the relationships between metals and metalloids soil multi-  
942 contamination, biodiversity facets (phylogenetic, taxonomic, and functional diversity),  
943 microbial biomass and activities and soil multifunctionality in (a) scrubland whole  
944 communities, (b) dominant species and (c) subordinate species. Effect sizes and correlation  
945 significance levels (\*:  $p \leq 0.05$ ; \*\*:  $p \leq 0.01$ ; \*\*\*:  $p \leq 0.001$ ) are indicated on each arrow. Bar

946 charts show the effect sizes of the direct impact of metals and metalloids soil multi-  
947 contamination and of the indirect effects of biodiversity facets on ecosystem multifunctionality  
948 mediated through microbial biomass and activities. Indirect effects were calculated by  
949 multiplying the direct effect of biodiversity facets on biomass and microbial activities, and the  
950 direct effect of biomass and microbial activities on ecosystem multifunctionality.

951

952 **Figure 5.** Path analysis of the relationships between metals and metalloids soil multi-  
953 contamination, biodiversity facets (phylogenetic, taxonomic, and functional diversity),  
954 microbial biomass and activities and individual soil functions ((a) total organic carbon, (b)  
955 available phosphorus, (c) cation exchange capacity, (d) water holding capacity and (e) total  
956 Kjeldahl nitrogen) in the 30 Mediterranean scrubland communities. Effect sizes and correlation  
957 significance levels (\*:  $p \leq 0.05$ ; \*\*:  $p \leq 0.01$ ; \*\*\*:  $p \leq 0.001$ ) are indicated on each arrow. Bar  
958 charts show the effect sizes of the direct impact of metals and metalloids soil multi-  
959 contamination and of the indirect effects of biodiversity facets on individual soil functions  
960 mediated through microbial biomass and activities. Indirect effects were calculated by  
961 multiplying the direct effect of biodiversity facets on biomass and microbial activities, and the  
962 direct effect of biomass and microbial activities on individual soil functions.

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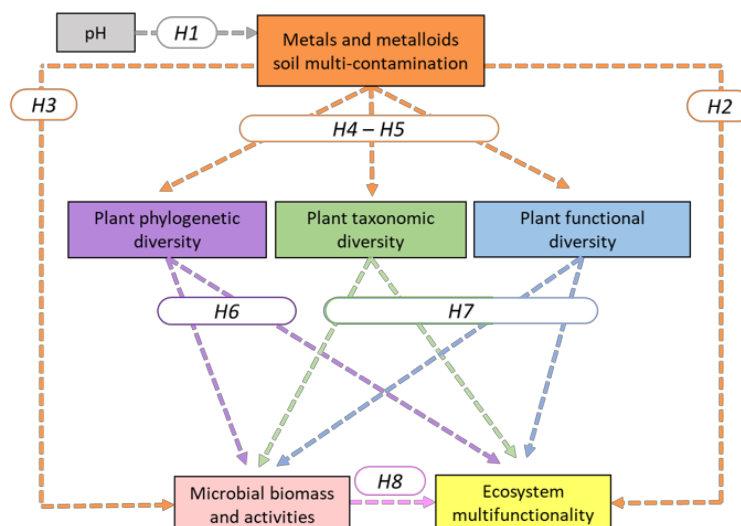


971 **Table legend**

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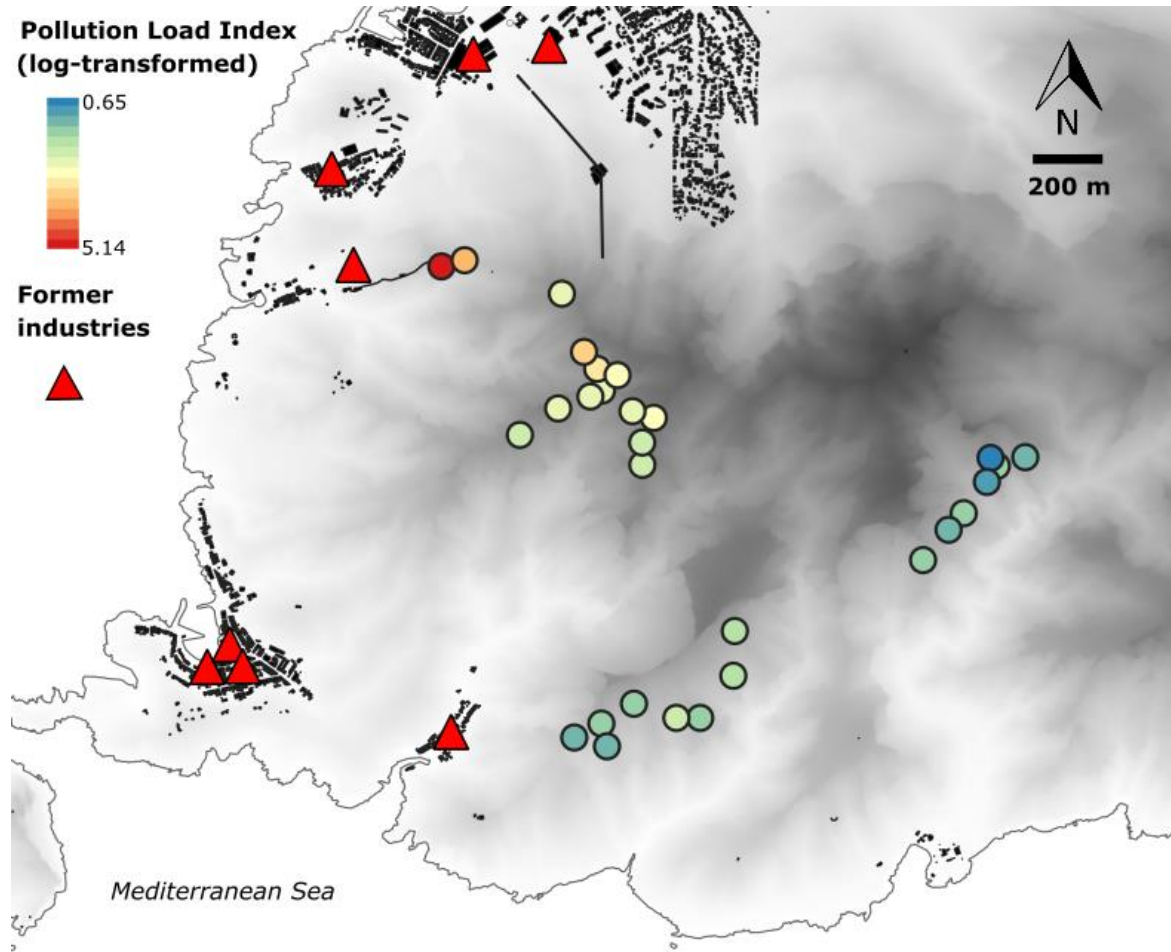
973 **Table 1.** Minimum, first quartile, median, third quartile and maximum of soil (pH, TOC: Total  
974 Organic Carbon, TKN: Total Kjeldahl Nitrogen, WHC: Water Holding Capacity, P: available  
975 phosphorus, CEC: Cation Exchange Capacity, PLI: Pollution Load Index), microbial  
976 community (SBR: Soil Basal Respiration, MB: Microbial Biomass, AWCD: Average Well  
977 Color Development, Hcat: catabolic diversity) and plant community (species richness,  
978 evenness, CWM LA: Community Weighted Mean of Leaf Area, CWM SLA: Community  
979 Weighted Mean of Specific Leaf Area, FDis: Functional Dispersion; wMPD: weighted Mean  
980 Phylogenetic Distance) variables used in the path analysis.

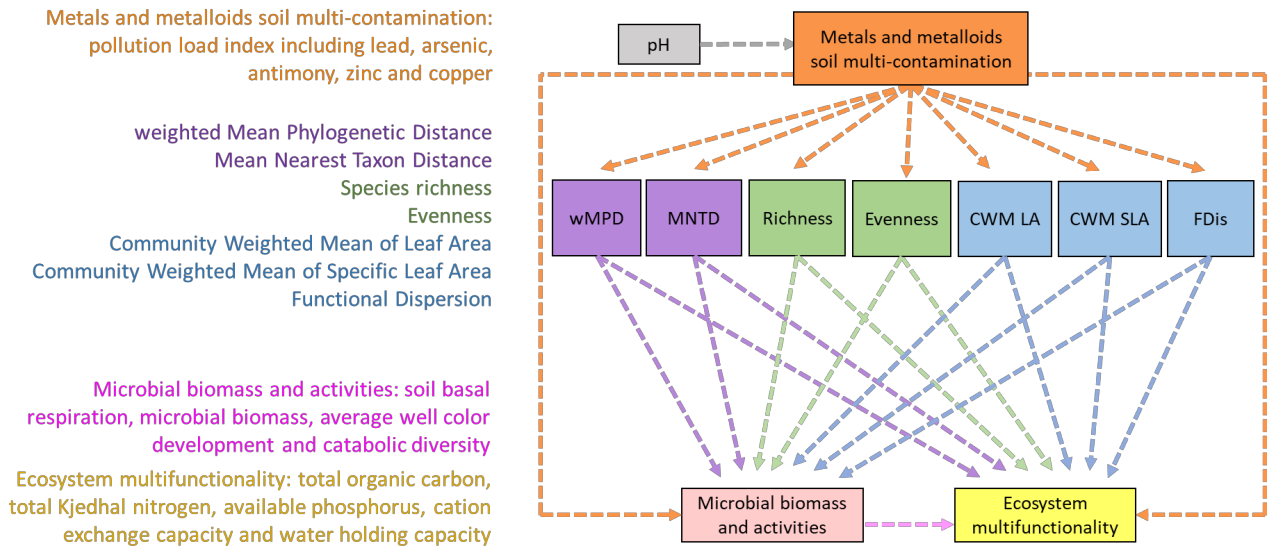
Variables	Minimum	First quartile	Median	Third quartile	Maximum
pH	7.88	8.08	8.19	8.28	8.38
TOC (g.kg <sup>-1</sup> of dry soil)	35.98	51.96	67.61	88.40	198.31
TKN (g.kg <sup>-1</sup> of dry soil)	1.89	4.13	4.80	6.03	11.58
Soil WHC (g.100 g <sup>-1</sup> of dry soil)	59.00	78.75	91.50	105.75	152.00
P (g.kg <sup>-1</sup> of dry soil)	13.00	20.50	29.00	35.00	68.00
CEC (m <sup>+</sup> eq.100 g <sup>-1</sup> of dry soil)	15.40	24.43	31.05	35.93	64.60
PLI	1.91	4.85	7.81	13.58	171.41
SBR (μg CO <sub>2</sub> -C.g <sup>-1</sup> .h <sup>-1</sup> )	0.02	0.02	0.03	0.04	0.08
Microbial MB (μg C <sub>mic</sub> .g <sup>-1</sup> )	0.01	0.02	0.02	0.03	0.06
Microbial community AWCD	0.35	0.72	0.89	0.97	1.44
H <sub>cat</sub>	2.72	2.95	3.07	3.14	3.24
Species richness	6.00	11.00	13.00	15.00	19.00
Evenness	0.80	0.87	0.88	0.91	0.97
Plant community CWM LA (mm <sup>2</sup> )	22.77	47.64	60.78	94.52	142.46
CWM SLA (mm <sup>2</sup> .mg <sup>-1</sup> )	15.50	42.51	49.05	57.59	90.83
FDIS	0.17	0.58	0.70	0.99	2.04
WMPPD	166.80	185.11	200.19	228.08	254.64

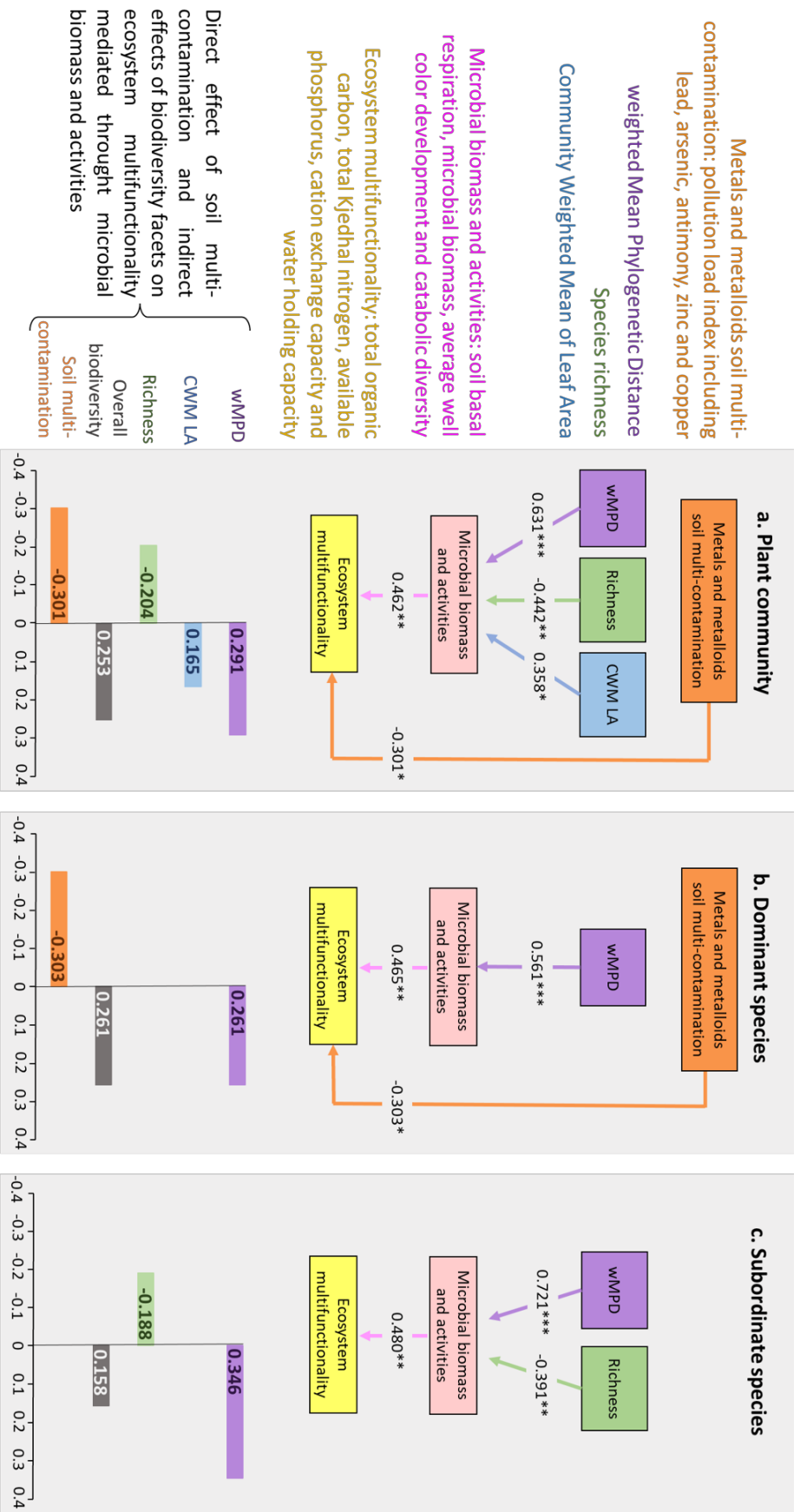


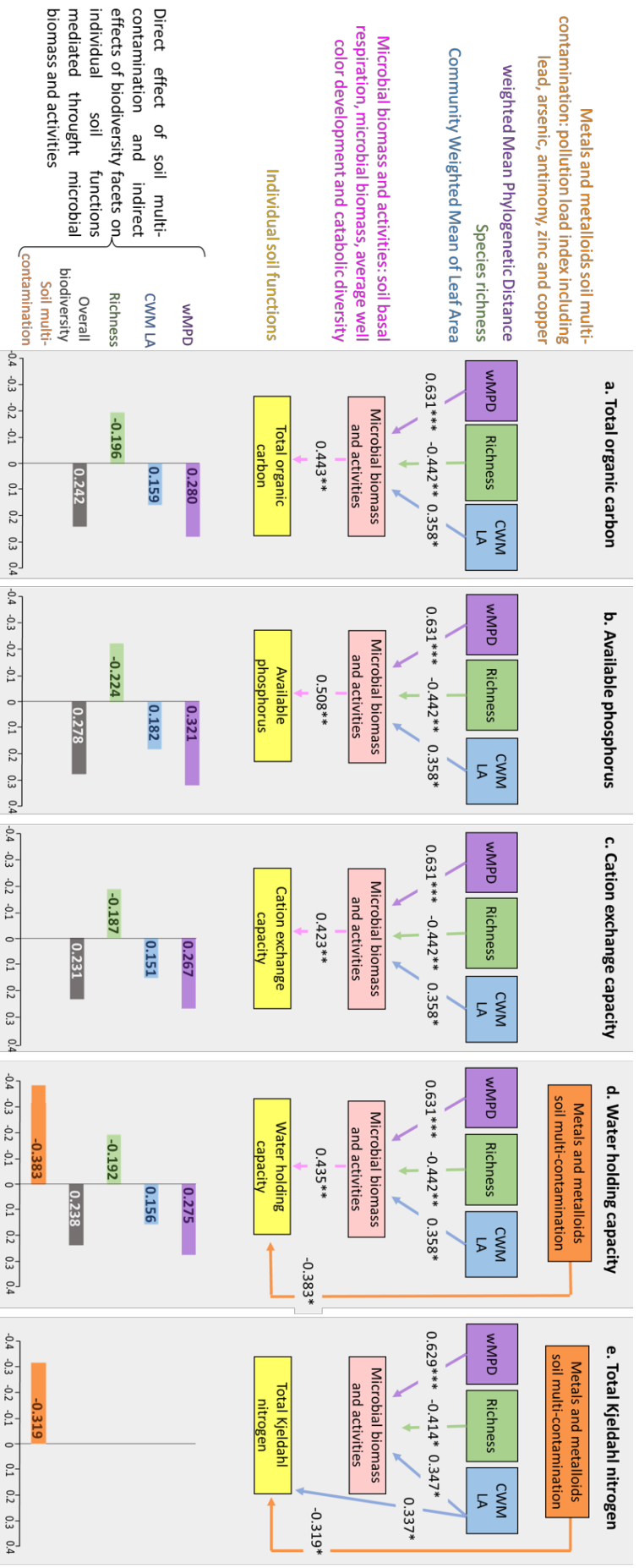
#### Hypotheses supporting the conceptual model


- H1** Relatively high pH in calcareous soils can limit MM mobility and bioavailability (Greger, 2004; Petruzzelli et al., 2020).
- MM soil multi-contamination generate physicochemical constraints on soil by adsorption of toxic MM (i.e., lead, arsenic, antimony) onto soil particles instead of nutrients, resulting in alteration of soil composition and functioning
- H2** (Greger, 2004; Petruzzelli et al., 2020).
- H3** MM soil multi-contamination generates stress on microbial communities resulting in decreased microbial biomass and activities (Kızılkaya et al., 2004 ; Gülser & Erdoğan, 2008).
- MM soil multi-contamination can act as a strong environmental filter (Keddy, 1992; Weiher et al., 1998) inducing plant physiological dysfunctions, especially water stress (Poschenrieder & Barcelo, 1999; Rucińska-Sobkowiak, 2016). As a result, MM soil multi-contamination can select MM-tolerant plant evolutionary lineages, forms and functions, resulting in reduction of phylogenetic, taxonomic, and functional diversity.
- Since dominant and subordinate species are not subjected to similar assembly processes (Arnillas & Cadotte, 2019; Arnillas et al., 2021), MM soil multi-contamination can differently impact on biodiversity of dominant and subordinate species, resulting in different patterns of biodiversity-ecosystem functioning relationships.
- H4**
- H5**
- Phylogenetic diversity can integrate plant-microorganisms coevolutionary processes and unmeasured biological traits (Flynn et al., 2011) as root associations with beneficial microorganisms (Brundrett, 2002; Werner et al., 2015) which highly influence soil functioning (Bardgett et al., 2014). As a result, decrease in phylogenetic diversity can lead to reduction of soil functioning.
- H6**
- H7**
- H8**
- H7** Taxonomic and functional diversities influence the quantity and the diversity of litter inputs, i.e., microbial substrates (Lange et al., 2015; Chen et al., 2019). As a result, decrease in taxonomic and functional diversity can lead to reduction of soil functioning.
- H8**



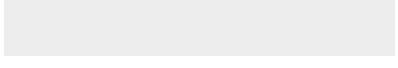









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## Declaration of Interest Statement

### **Declaration of interests**

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

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

**CRedit authorship contribution statement**

**Lorène Tosini:** Conceptualization, Methodology, Investigation, Software, Formal analysis, Writing – Original Draft, Writing – Review & Editing, Project administration, Funding acquisition. **Manuel Cartereau:** Conceptualization, Methodology, Investigation, Formal analysis, Writing – Review & Editing. **Yoann Le Bagousse-Pinguet:** Conceptualization, Methodology, Formal analysis, Writing – Review & Editing. **Isabelle Laffont-Schwob:** Conceptualization, Methodology, Investigation, Writing – Review & Editing, Project administration, Funding acquisition. **Pascale Prudent:** Conceptualization, Methodology, Investigation, Writing – Review & Editing. **Anne-Marie Farnet Da Silva:** Investigation, Writing – Review & Editing. **Nicolas Montès:** Methodology, Investigation, Software, Writing – Review & Editing. **Yoan Labrousse:** Investigation. **Laurent Vassalo:** Investigation. **Hélène Folzer:** Conceptualization, Methodology, Investigation, Writing – Review & Editing, Project administration, Funding acquisition.