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HAL Authorization

1 **Current permissible levels of heavy metal pollutants harm**  
2 **terrestrial invertebrates**

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41 **Abstract**

42 The current decline of invertebrates worldwide is alarming. Several potential causes have  
43 been proposed but heavy metals, while being a widespread and major pollutants of air, soils  
44 and water, have so far been largely overlooked. Here, we ran a meta-analysis of 527 datasets  
45 on the effects of arsenic, cadmium, lead and mercury on terrestrial invertebrates. These four  
46 well-studied metals, for which international guidelines exist, significantly impact the  
47 physiology and behavior of invertebrates, even at levels below those recommended as ‘safe’  
48 for humans. Our results call for a revision of the regulatory thresholds to better protect  
49 terrestrial invertebrates, which appear to be more sensitive to metal pollution than vertebrates.  
50 More fundamental research is needed to improve international guidelines for metal pollutants,  
51 and to develop conservation plans to mitigate invertebrate declines and protect ecosystem  
52 services.

53

54

## 55 INTRODUCTION

56 Terrestrial invertebrate bioabundance and biodiversity are declining (Wagner 2020).  
57 Since invertebrates are basal to terrestrial food webs and provide key ecosystem services, the  
58 short-term ecological consequences of invertebrate decline could be very severe (Goulson  
59 2019; Sánchez-Bayo & Wyckhuys 2019). The rate of decline is especially alarming as it has  
60 been estimated that land-dwelling insects disappear at a rate of ca. 1% every year since a  
61 century (van Klink *et al.* 2020). Many factors have been proposed to explain this loss. These  
62 include climate change (Wilson *et al.* 2007), habitat reduction due to intensive agriculture and  
63 urbanization (Fattorini 2011; Dudley & Alexander 2019), introduced pathogens, predators and  
64 competitors (Goulson *et al.* 2015), as well as chronic exposure to agrochemicals (van  
65 Lexmond *et al.* 2015).

66 Here we argue that heavy metal pollution is a major, yet currently overlooked, stressor  
67 to insects and other terrestrial invertebrates that needs urgent attention from scientists and  
68 stakeholders. At trace levels, metals such as cobalt, copper, iron, manganese, selenium and  
69 zinc are essential micronutrients for animals and plants (Phipps 1981; WHO/FAO/IAEA  
70 1996). Others, such as arsenic, cadmium, chromium, mercury, lead and nickel have no useful  
71 biological function and exert toxic effects even at low concentrations (He *et al.* 2005;  
72 Tchounwou *et al.* 2012). While these metals are naturally present in the Earth's crust, mining  
73 and smelting operations, combustion of fossil fuels, industrial production, domestic and  
74 agricultural use of metals and metal-containing compounds (Bradl 2005), have considerably  
75 increased environmental concentrations of these metals above natural baselines (Zhou *et al.*  
76 2018), contaminating air (Suvarapu & Baek 2017), soils (Wuana & Okieimen 2011), water  
77 (Mance 1987) and plants (Krämer 2010).

78 Despite our knowledge of the detrimental impact of this heavy metal pollution on  
79 vertebrates, which include cellular damage, carcinogenesis and neurotoxicity (Tchounwou *et*

80 *al.* 2012; Chen *et al.* 2016), metal pollution is still increasing, calling for a more systematic  
81 assessment on the impact on biodiversity. For example, in 2019, the World Health  
82 Organization (WHO) stated that there was no safe level of lead for vertebrates (WHO 2019),  
83 yet the majority of industrial activities are consistently elevating the level of lead in the  
84 environment (Järup 2003; Li *et al.* 2014). The recent report that bees and flies in densely  
85 urbanized areas suffer from exposure to metallic air particles (Thimmegowda *et al.* 2020)  
86 suggests that the consequences of heavy metal pollution on terrestrial invertebrates could be  
87 extremely important and widespread (for a review on aquatic invertebrates see (Rainbow  
88 2002)).

89         Here, we assessed the impact of heavy metals on terrestrial invertebrates through a  
90 meta-analysis of the scientific literature on four well-studied metals over the past 45 years.  
91 We found that these metals have detrimental effects on a wide diversity of species at levels  
92 below those considered safe for humans. Based on these results, we discuss the need for more  
93 fundamental research into the impacts of heavy metal pollutants on insects to improve  
94 international guidelines for the regulation of metal pollutants, and better inform conservation  
95 plans.

96

## 97 **METHODS**

### 98 *Literature review and data extraction*

99         We focused on the four most hazardous heavy metals documented for humans  
100 (ATSDR 2019), for which international regulatory implementations exist (Table 1): arsenic  
101 (As), cadmium (Cd), mercury (Hg) and lead (Pb). We searched articles in the ISI Web of  
102 Knowledge database (search performed on 25/03/2020) using the keywords: heavy metals,  
103 metalloids, arsenic, cadmium, lead, mercury, insects, invertebrates. The search was restricted  
104 to articles published between 1975 and 2020 (maximum available year range on ISI Web of

105 Knowledge). Among the 460 hits, we selected those studies focusing on terrestrial  
106 invertebrates (i.e. protostomes) from the abstracts, and excluded review articles. This filtering  
107 yielded a subset of 154 articles from which we extracted 527 datasets of studies investigating  
108 effects of heavy metals on terrestrial invertebrates (see raw data in Table S1).

109 From each dataset, we extracted: (1) the name of targeted invertebrate species, (2) the  
110 heavy metal(s) used, (3) the experimental conditions (field, laboratory), (4) the mode of  
111 exposure to the metal (food, water, soil), (5) the type of exposure (acute: < 24h, chronic: >  
112 24h), (6) the range of metal concentrations tested (min- max in ppm), (7) the biological  
113 responses measured (e.g. survival, reproduction, behavior), and (8) the lowest metal  
114 concentration for which an effect was observed.

115 Briefly, the vast majority of the datasets focused on cadmium (46%) and lead (37%),  
116 while less information was available on arsenic (10%) and mercury (7%) (Figure 1A). 59% of  
117 the datasets were obtained in field surveys and 41% in laboratory experiments with controlled  
118 exposure. 79% of the studies were published in the last 13 years, indicating only recent  
119 attention to this issue. Since the effects can greatly vary depending on the duration of  
120 exposure and time of assessment, here we considered as acute exposure any case where  
121 individuals were exposed to a single dose and assessed within 24h. Despite the diversity of  
122 protocols, most studies used chronic exposure (95%), through the diet (49%) or the soil  
123 (43%).

124

#### 125 *Concentration ranges*

126 International permissible limits (i.e. recommended maximum concentrations) are  
127 based on values established for humans in food, water and soil (see Table S2). These limits  
128 vary across types of food, water (i.e. drinking, irrigation) and soils (i.e. allotment,  
129 commercial, residential, agricultural). For each of these matrices, we thus considered the

130 minimal and the maximal estimates of permissible limits. We defined three concentration  
131 ranges: below the minimal estimated limit, between the minimal and maximal estimated  
132 limits, and above the maximal estimated limit (Table 1). Note that for water, whenever  
133 possible, we considered the minimal value for drinking water and the maximal value for  
134 irrigation water.

135

## 136 **RESULTS**

### 137 *Few studies focus on functionally important species*

138         The 527 datasets covered 100 species (83% Arthropoda, 15% Annelida, 1.2%  
139 Rotifera, 0.4% Tardigrada, 0.2% Mollusca; Figure 1B). Studies were biased toward pest  
140 species with an economic impact (34% of datasets; e.g. the gypsy moth *Limantria dispar*, the  
141 grasshopper *Aiolopus thalassinus*, the beet armyworm *Spodoptera exigua*) and model species  
142 in biology (10%; e.g. fruit fly *Drosophila melanogaster*, large milkweed bug *Oncopeltus*  
143 *fasciatus*). Other groups were comparatively under-represented, including important  
144 bioindicators, such as decomposers (15%; e.g. *Lumbricus terrestris*, *Eisenia fetida* and *E.*  
145 *andre*), predators (10%; e.g. ants *Formica* spp., spiders *Araneus* spp. and *Pardosa* spp.) and  
146 pollinators (13%; e.g. the honey bee *Apis mellifera*). Some taxonomic orders that include  
147 large numbers of species with key functional roles for nutrient cycling (e.g. proturans,  
148 diplurans, earwigs), soil aeration (e.g. centipedes), or pollination (e.g. thrips) were not  
149 represented at all. Research is thus needed on these important invertebrate orders to get a  
150 more accurate picture of how metallic pollution disturb key ecological functions (Skaldina &  
151 Sorvari 2019).

152

### 153 *Heavy metals have detrimental effects below permissible limits*

154 Deleterious effects were reported in 70% of the datasets (Figure 2A), be them  
155 collected in the laboratory (84%, N=263) or in the field (49%, N=104), following chronic  
156 (79%, N=348) or acute exposure (69%, N=19). When considering only the datasets reporting  
157 deleterious effects, 73% of these effects (N=269) were measured at concentrations above the  
158 maximal estimated permissible limit (see Table 1). Yet, 12% (N=45) were measured in  
159 between the regulatory thresholds and 15% (N=53) below the minimal estimated limit (Figure  
160 2A). In addition, a majority (57%, N=53) of the datasets using at least one concentration  
161 below the minimal estimated permissible limit found a negative effect, irrespective of the  
162 metal.

163 When considering only the laboratory studies, in which exposure concentrations were  
164 controlled (Figure 2B-C), 67% of the studies that examined levels below the permissible  
165 limits (N=29) reported deleterious effects on invertebrates. Of the laboratory studies  
166 investigating acute exposure below the permissible limits (N=10), seven found deleterious  
167 effects (Figure 2B). Hence, acute exposure, while presumably rare in nature, can have  
168 deleterious effects on invertebrates below current permissible exposure levels. This suggests  
169 that the permissible limits designed for humans are not appropriate for terrestrial  
170 invertebrates, who seem to be more sensitive to metal pollutants.

171

### 172 *Few studies address the behavioral effects of heavy metals*

173 Research on the impact of heavy metals on terrestrial invertebrates has developed  
174 quite recently. This is illustrated by the fact that 79% of the studies sampled for our analysis  
175 were published after 2007 (Figure 3A). About half of the experiments focused on physiology  
176 (52%), followed by studies on development (17%), survival (13%), population dynamics  
177 (6%), reproduction (6%) and behavior (6%) (Figure 3B). It has become increasingly clear that  
178 understanding the sublethal behavioral effects of a stressor (e.g. mobility, navigation, feeding

179 behavior, learning, memory) is crucial to assess the long-term impact of that stressor on  
180 invertebrate populations (Mogren & Trumble 2010). This has become evident for bees, for  
181 instance, for which any impairment of the cognitive functions involved in foraging is a direct  
182 threat for the larvae or the whole colony that cannot appropriately access nutrients (Klein *et*  
183 *al.* 2017). In our meta-analysis, 33 experiments reported behavioral effects (Figure 3B), but  
184 only two explored cognitive effects (Philips *et al.* 2017; Piccoli *et al.* 2020). This is a very  
185 low number considering the well-known neurotoxic effects of the four metals on humans  
186 (Wright & Baccarelli 2007; Chen *et al.* 2016) and other animals, including aquatic  
187 invertebrates (Salanki 2000).

188

189 *Few studies investigated co-occurrences despite clear synergistic effects*

190 In nature, pollutants rarely occur alone. Heavy metals are no exception since they  
191 share common emission sources (Vareda *et al.* 2019). For instance, cadmium, copper, zinc  
192 and lead frequently co-occur due to the output from smelters, or the application of sewage  
193 sludge as fertilizer (Bradl 2005). High positive correlations between chromium, cadmium and  
194 arsenic amounts have been found in soil samples (Chen *et al.* 1999; Navas & Machín 2002),  
195 and many studies have shown the co-accumulation of several trace metals in insects (Wilczek  
196 & Babczyk 2000; Nummelin *et al.* 2007; Goretti *et al.* 2020). As such, co-occurring metals  
197 could have additive, antagonistic or synergistic effects (Jensen & Trumble 2003). These  
198 interactive effects may also be influenced by the presence of other environmental stressors,  
199 such as pesticides or parasites (Alaux *et al.* 2010).

200 Only 7 out of the 154 studies addressed the question of combined effects in laboratory  
201 conditions (Figure 1A). Nonetheless the effects are clear: 55% of the experiments (N=10)  
202 reported synergistic detrimental consequences. For instance, ants (*Formica aquilonia*)  
203 chronically exposed to both cadmium and mercury failed to develop compensatory

204 mechanisms to maintain energetic balance, causing colony collapse, while being able to cope  
205 when exposed to each metal alone (Migula *et al.* 1997). Similarly, the lethal effects of  
206 cadmium and zinc on aphids (*Myzus persicae*) were potentiated when the two metals were  
207 combined, which led to accelerated extinction of the treated population (Stolpe & Müller  
208 2016). These two metals were reported to be either synergistic or antagonist on earthworms  
209 (*E. fetida*) depending on their concentrations (Wu *et al.* 2012). Finally, the joint exposure of  
210 honey bees (*A. mellifera*) to cadmium and copper induced increased development duration  
211 and mortality, and decreased food intake and sucrose response (Di *et al.* 2020). Thus, the  
212 effects of metal co-exposure are complex and variable. The paucity of studies may be because  
213 they require more sophisticated experimental designs, larger sample sizes (factorial designs)  
214 and may yield results that are more difficult to interpret. Yet, these studies are crucial if we  
215 are to revise the current regulations which presently only consider permissible limits for  
216 metals in isolation (Tables 1 and S2).

217

## 218 **DISCUSSION**

219 Our meta-analysis of the literature on lead, arsenic, cadmium and mercury shows  
220 many negative effects of these heavy metal pollutants on terrestrial invertebrates. Particularly  
221 worrisome are the reports of negative effects observed at doses below permissible limits in  
222 most of the studied taxa. There are reported lethal effects on grasshoppers (Schmidt *et al.*  
223 1991), moths (Andrahennadi & Pickering 2008), flies (Massadeh *et al.* 2008) and other  
224 groups (Osman *et al.* 2015; Polykretis *et al.* 2016; Stolpe *et al.* 2017). Metal exposure induces  
225 a number of sublethal effects, sometimes difficult to assess, such as impaired fertility  
226 (grasshoppers: Schmidt *et al.* 1991; springtail: Crouau & Pinelli 2008; earthworm: Konečný  
227 *et al.* 2014), developmental defects (blowfly: Nascarella *et al.* 2003; moth: van Ooik *et al.*  
228 2007; ant: Skaldina *et al.* 2018), resistance to pathogens (ant: Sorvari *et al.* 2007; honey bee:

229 Polykretis *et al.* 2016) but also altered feeding behavior (aphid: Stolpe *et al.* 2017; honey bee:  
230 Burden *et al.* 2019).

231

232 *The impact of heavy metal pollutants is poorly understood*

233         At present, it is likely that the severity of these effects is underestimated. Many  
234 laboratory experiments gave animals rather limited exposure times, rarely reaching the  
235 duration of a complete life cycle. Besides, most studies overlooked any consequences of  
236 exposure to multiple metal contaminants, which would be a common occurrence in nature.  
237 There is now clearly growing interest in assessing the sublethal impacts of metals. This trend  
238 echoes the recent shift seen in pesticide research on beneficial insects, especially pollinators,  
239 which has moved from decades of standard survival assays to experimental designs aiming at  
240 characterizing the effects on behavior and cognition (Desneux *et al.* 2007; Klein *et al.* 2017).  
241 Just like pesticides, heavy metal pollutants have subtle, but potentially serious, effects on  
242 pollinators' behavior by disturbing foraging activity (Sivakoff & Gardiner 2017; Xun *et al.*  
243 2018), food perception (Burden *et al.* 2019) as well as the learning and memory abilities  
244 required for efficient foraging (Burden *et al.* 2016; Monchanin *et al.* 2020). Through all of  
245 these mechanisms, heavy metal exposure can compromise food supply to the offspring, and  
246 hence the viability of a colony or population.

247         There are potentially complex interactions between behavior and pollutant exposure.  
248 Since an animal's behavior can influence how much metal pollution it is exposed to (Mogren  
249 & Trumble 2010; Gall *et al.* 2015), behavioral disturbances may affect exposure and  
250 sensitivity to metals. For example, impaired locomotion may reduce the capacity of  
251 individuals to avoid contaminated sites (Hirsch *et al.* 2003) and indiscriminate oviposition  
252 may jeopardize the survival of offspring if they are deposited on an unfavorable food plant  
253 (Cervera *et al.* 2004; Tollett *et al.* 2009). It is thus likely that we are currently underestimating

254 the impact of metal pollution on invertebrates, due to a lack of understanding of their  
255 sublethal effects on most species.

256

257 *Multiple possible causes of invertebrates' high sensitivity to metal pollution*

258         Our survey of the literature suggests that invertebrates may be more sensitive to the  
259 damaging effects of heavy metals than the mammals (e.g. humans, rodents) typically used to  
260 determine “safe” environmental levels. Sensitivity varies between species and depends on the  
261 considered metals (Malaj *et al.* 2016). Some can discriminate metal contaminated food from  
262 uncontaminated food (Mogren & Trumble 2010), but other species seem unable to (Stolpe *et*  
263 *al.* 2017; Burden *et al.* 2019). This is particularly critical for animals feeding on resources that  
264 can accumulate metals, such as leaves (Krämer 2010) or nectar (Gutiérrez *et al.* 2015).

265         Perhaps more importantly, there is also emerging evidence that invertebrates may have  
266 higher levels of exposure to heavy metals in the field than large mammals. Surveys of  
267 terrestrial biotopes show that non-essential metals tend to be more highly accumulated in  
268 invertebrates than in vertebrates (Hsu *et al.* 2006), which seems also the case for aquatic taxa  
269 (Xin *et al.* 2015). Due to their small size, their relatively high surface area/volume ratio and  
270 the niches they occupy, invertebrates are frequently in intimate contact with soils and  
271 vegetation and can get contaminated by specific feeding modes such as filter-feeding or  
272 deposit-feeding (De Lange *et al.* 2009). Their limited dispersive capacities may greatly reduce  
273 their ability to move away from polluted areas, even if they can detect harmful levels of trace  
274 elements. As a result, metals accumulate in the bodies of individuals (Nannoni *et al.* 2011;  
275 Mukhtorova *et al.* 2019; Schrögel & Wätjen 2019; Goretti *et al.* 2020) and in the nest in the  
276 case of social species (Skaldina *et al.* 2018; Veleminsky *et al.* 1990). Some terrestrial  
277 invertebrates (e.g. ants, earthworms, bees, Isopoda) could therefore be relevant and sensitive  
278 bioindicators due to their particular vulnerability to metal contamination.

279 Invertebrates do have mechanisms to process heavy metals. Excessive metals can be  
280 eliminated through feces (Przybyłowicz *et al.* 2003), accumulated in insect exoskeleton before  
281 molting (Borowska *et al.* 2004), or stored in specific organs (Nica *et al.* 2012). They can also  
282 induce expression of proteins involved in excretion and/or detoxification (for reviews, see  
283 Janssens *et al.* 2009; Merritt *et al.* 2017). Yet, while these detoxification mechanisms may  
284 protect species to a point, they are unlikely to spare them from the sublethal effects resulting  
285 from impaired brain or organ function. We clearly need a better characterization of the  
286 physiological and molecular mechanisms underlying metal transfer, toxicity and tolerance in  
287 invertebrates in order to better understand their sensitivity to heavy metals.

288

289 *A need to revise guidelines of safe environmental levels of heavy metals*

290 Since heavy metals are such widespread and persistent pollutants in the environment,  
291 it is a priority to develop a better assessment of their impacts on invertebrates. Our most  
292 concerning finding is the evidence that terrestrial invertebrates are highly sensitive to heavy  
293 metals, but our meta-analysis also highlights important gaps in our knowledge. We need to  
294 study a larger diversity of species, consider potential cocktail effects, and extend studies  
295 beyond the four metals addressed here. Other heavy metals have been reported to negatively  
296 impact terrestrial invertebrate populations at low doses, such as selenium (deBruyn &  
297 Chapman 2007), zinc (e.g. Cheruiyot *et al.* 2013), copper (e.g. Di *et al.* 2016), cobalt (e.g.  
298 Cheruiyot *et al.* 2013), manganese (Ben-Shahar 2018) and chromium (e.g. Sgolastra *et al.*  
299 2018). Characterizing the impacts of heavy metals on insect fitness is going to demand an  
300 integrative and interdisciplinary research agenda, just like what has been established to assess  
301 pesticide impacts on beneficial insects. In particular, focusing on the sublethal effects of  
302 neonicotinoids on pollinators (Henry *et al.* 2012; Crall *et al.* 2018), triggered a revision of the  
303 risk assessments scheme and their ban in the European Union in 2018.

304

## 305 **CONCLUDING REMARKS**

306           This survey of the existing literature clearly indicates that terrestrial invertebrates  
307 appear particularly vulnerable to heavy metal pollutants, and that most existing standards are  
308 not suited to protect them. We now need more integrative toxicological studies, on a broader  
309 range of invertebrate species, to better assess the impact of metal pollutants on fitness, and to  
310 update the current environmental regulation of metals. Only by addressing these important  
311 challenges will we be able to mitigate consequences on ecosystems and food safety, in a  
312 context of rapid and widespread decline of invertebrate biodiversity.

313

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321

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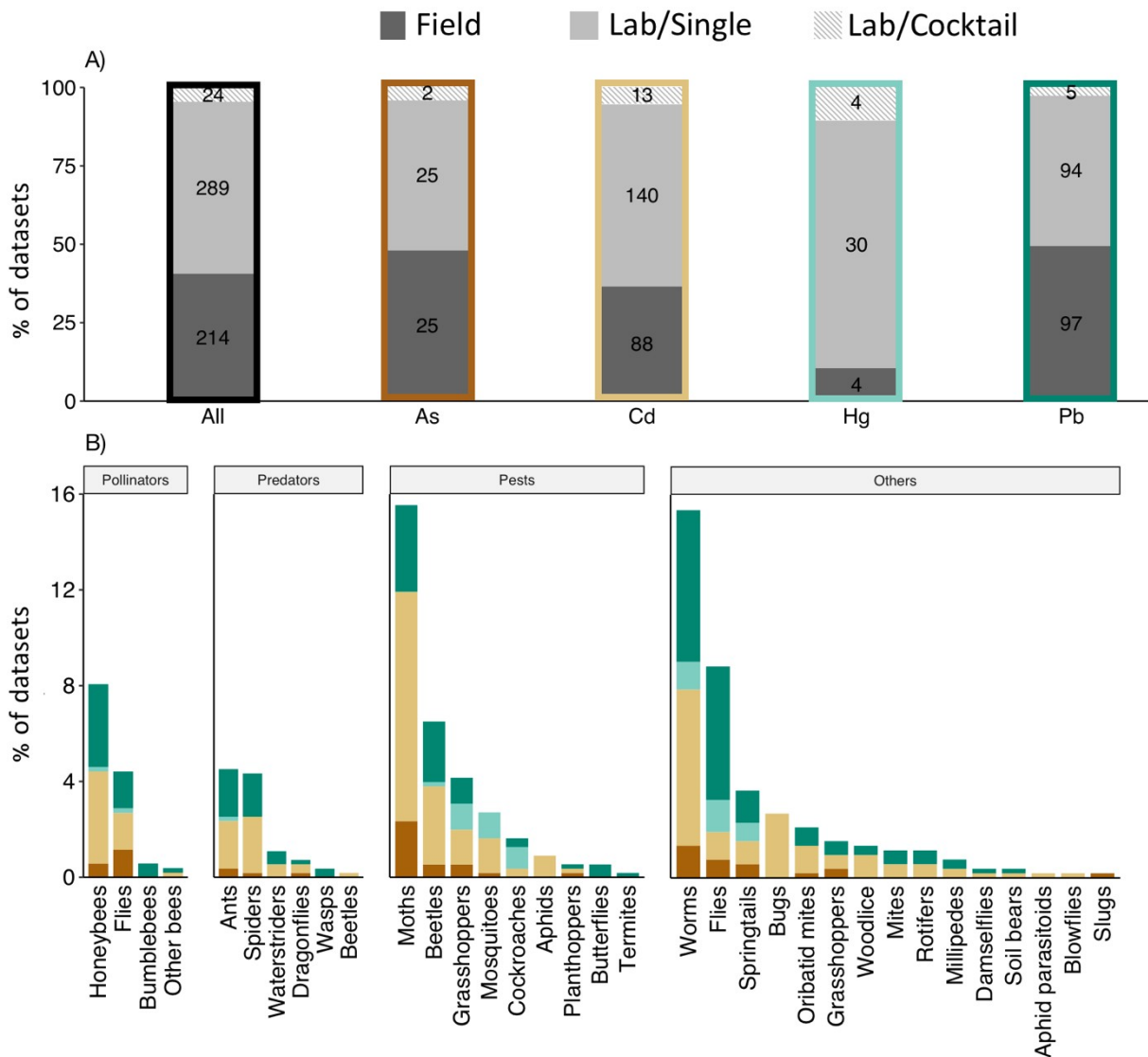
574 **TABLES**

575 **Table 1: Permissible limits (ppm) for heavy metals in food, water and soil.** All  
 576 permissible limits are based on human toxicity data. Levels were determined from the  
 577 international standards set by the World Health Organization (WHO) and the Food and  
 578 Agriculture Organization (FAO) of the United Nations. The permissible limits are  
 579 recommended values for: ‘food and drinking water’, as defined in the Codex Alimentarius  
 580 (2015), to deal with ‘contaminants and toxins in food and feed’ and to be ‘applied to  
 581 commodities moving in international trades’ (Codex Alimentarius 2015); guidelines for water  
 582 quality in irrigation (Ayers & Westcot 1994); critical values in soil based on the Organization  
 583 for Economic Co-operation and Development (OECD) risk assessment studies (de Vries *et al.*  
 584 2003) and FAO standards (WHO/FAO 2001). Local guidelines (see Table S2), when they  
 585 exist, can vary across countries and are less conservative (higher thresholds) than the  
 586 international standards, especially for soils and water. We defined three concentration ranges:  
 587 below the minimal estimated permissible limit (green), between the minimal and maximal  
 588 estimated permissible limits (orange), and above the maximal estimated permissible limit  
 589 (red). Whenever only one threshold value was defined, no intermediate range could be  
 590 defined (NA: not applicable). The same color codes for metals and concentration ranges were  
 591 used in Figures 1 and 2.

Matrices	Arsenic (As)			Cadmium (Cd)			Mercury (Hg)			Lead (Pb)		
Food	<0.1	0.1-0.2	>0.2	>0.05	0.05-2	>2	<0.5	0.5-1	>1	<0.01	0.01-3	>3
Water	<0.01	0.01-0.1	>0.1	<0.003	0.003-0.01	>0.01	<0.001	NA	>0.001	<0.01	0.01-5	>5
Soil	<20	NA	>20	<0.9	0.9-3	>3	<0.03	0.03-2	>2	<30	30-50	>50

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595

596 **Figure 1: Diversity of groups of invertebrate species and experimental conditions. A)**

597 Percentage of experiments (sample sizes in black) conducted in the field (dark grey) or in the

598 lab (light grey) per heavy metal. Experiments with mixtures of metals in the lab are displayed

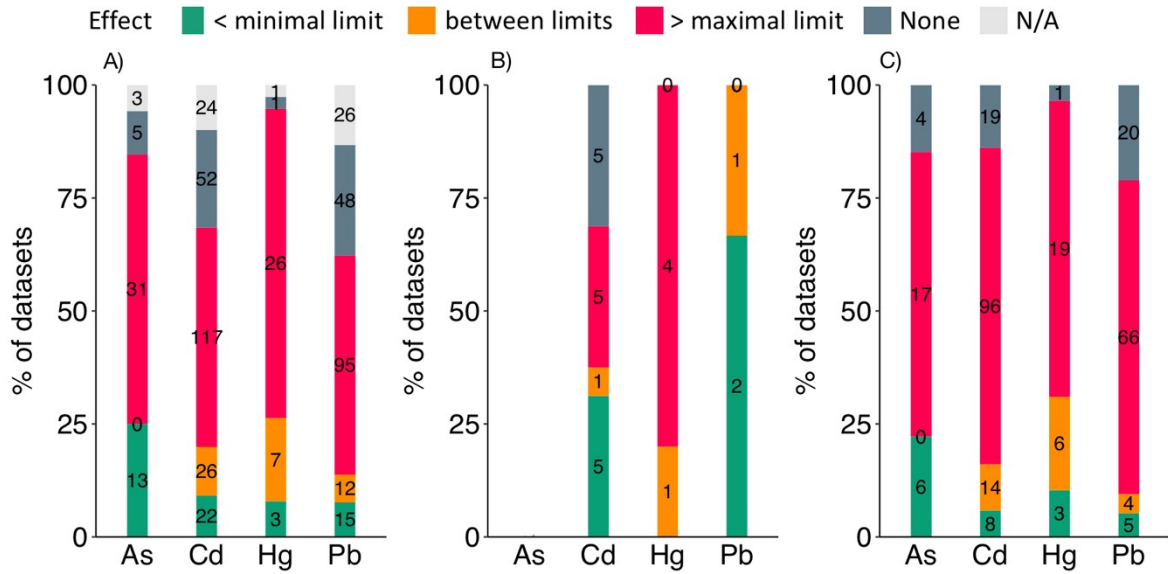
599 in textured light grey. Numbers of datasets are shown in bars. **B)** Diversity of invertebrate

600 groups classified by economic importance (based on Skaldina and Sorvari (2019)). Datasets

601 with different metals are marked by the same color code as Table 1 (As: brown, Cd: beige,

602 Hg: light green, Pb: dark green).

603



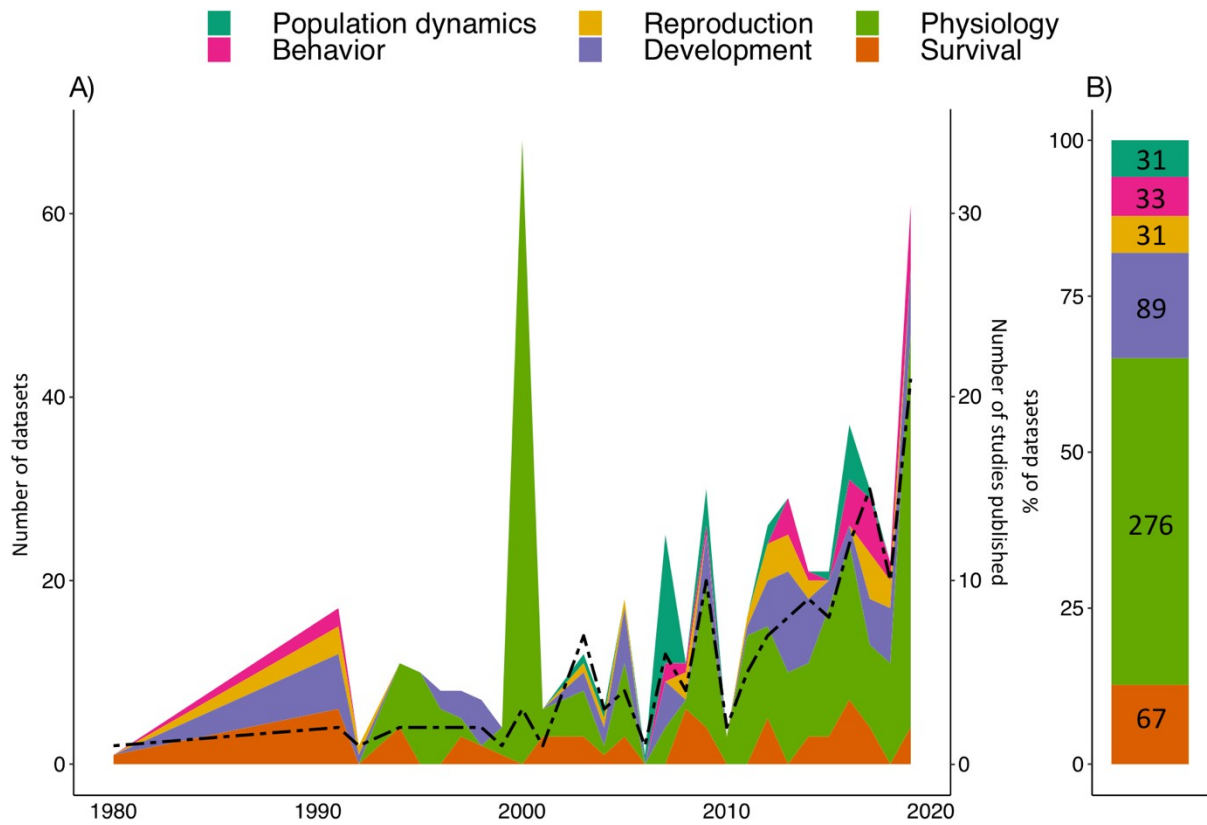
604

605 **Figure 2: Effects observed according to permissible limits.** We defined the following  
 606 ranges below the minimal estimated limit, between the minimal and the maximal estimated  
 607 limits, or above the maximal estimated limit. **A)** All studies (N=527). **B)** Laboratory studies  
 608 with acute exposure (N=24) and **C)** chronic exposure (N=288). None: no observable effect,  
 609 N/A: or no conclusion available. Sample sizes are in black. Concentration ranges were  
 610 marked by the same color code as Table 1.

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615 **Figure 3: Biological variables measured.** **A)** Area chart of the number of experiments per  
 616 biological variable (year 2020 was omitted). The peak in 2000 is due to three large studies  
 617 studying physiological effects in the field (38 experiments). The black dashed line represents  
 618 the number of studies published yearly. **B)** Overall proportions of experiments per biological  
 619 variable (numbers of datasets in black).

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622 **SUPPLEMENTARY MATERIALS**

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624 **Table S1: Raw data.** Excel file detailing all parameters took into account for the analyses

625 and the bibliographic sources of the 154 published articles.

626

627 **Table S2: Permissible limits (ppm) for heavy metals in food, water and soil according to**

628 **international and local standards.** International standards used are displayed in bold.

629

Matrices	Area of application	Arsenic (As)	Cadmium (Cd)	Mercury (Hg)	Lead (Pb)	Source
Food	<b>International</b>	<b>0.1-0.2</b>	<b>0.05-2</b>	<b>0.5-1</b>	<b>0.01-3</b>	<b>(Codex Alimentarius 2015)<sup>1</sup></b>
	European Union	0.1-0.2	0.05-3	0.1-1	0.02-3	Commission Regulation (EC) No 1881/2006 2006 <sup>2</sup> ; Commission Regulation (EU) 2015/1006 2015
	USA	NA	NA	NA	NA	The U.S. Food and Drug Administration has not established regulatory limits for trace metals in finished food products other than bottled water.
	China	0.5	0.05	0.01	0.2	Retrieved from (Li <i>et al.</i> 2006) <sup>3</sup>
Drinking water	<b>International</b>	<b>0.01</b>	<b>0.003</b>	<b>0.001</b>	<b>0.01</b>	<b>(Codex Alimentarius 2015)<sup>1</sup></b>
	European Union	0.01	0.005	0.01	0.001	(European Commission 2019) <sup>4</sup>

	USA	0.01	0.005	0.002	0.015	(US EPA 2017) <sup>5</sup>
	China	0.01	0.005	0.001	0.01	(Ministry of Health, China 2006) <sup>6</sup>
Irrigation water	<b>International</b>	<b>0.1</b>	<b>0.01</b>	<b>NA</b>	<b>5</b>	<b>(Ayers &amp; Westcot 1994)<sup>7</sup></b>
	USA	NA	0.005-0.01	NA	5	(US EPA 1978) <sup>8</sup>
	China	0.05	NA	0.01	NA	Retrieved from (Zhao <i>et al.</i> 2018) <sup>9</sup>
Soil	<b>International</b>	<b>20</b>	<b>0.9-3</b>	<b>0.03-2</b>	<b>30-50</b>	(WHO/FAO 2001; de Vries <i>et al.</i> 2003) <sup>10,11</sup>
	European Union	NA	NA	NA	NA	The European Union has not established limits for heavy metals in soils. There is however on-going policy to manage contamination, see (Council Directive 86/278/EEC 1986) <sup>12</sup> which states limit values in sludge for use in agriculture (Cd: 1-3 ppm; Hg:1-1.5 ppm; Pb:50-300 ppm)
	Finland	5-100	1-20	0.5-5	60-750	(Ministry of the Environment, Finland 2007) <sup>13</sup> . The Finnish standard values represent a good approximation of different national systems in Europe have been applied in an international context for

						agricultural soils as well.
	UK	32-640	10-230	1-3600	450-750	(UK Environment Agency 2009) <sup>14</sup>
	USA	0.11	0.48	1	200	(US EPA 2002) <sup>15</sup>  (US EPA 1993) <sup>16</sup> stated limit values in sludge for use in agriculture (As: 75 ppm; Cd: 85 ppm; Hg: 420 ppm; Pb: 840 ppm)
	China	20-40	0.3-0.6	0.3-1	80	(Environmental Protection Ministry of China 2015) <sup>17</sup>

630 References:

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