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Bioaccumulation of heavy metals in earthworms from field contaminated soil in a subtropical area of China

Kun Wang[a], Yuhui Qiao[a,*], Huiqi Zhang[a], Shizhong Yue[a], Huafen Li[a], Xionghui Ji[b], Longsheng Liu[c]

[a] College of Resources and Environmental Science, China Agricultural University, Beijing 100193, China
[b] Hunan Academy of Agricultural Sciences, Changsha 410125, China
[c] Hengyang Academy of Agricultural Sciences, Hengyang 421151, China

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ABSTRACT

Bioaccumulation factors for heavy metals (Cd, Zn, Cu, and Pb) were examined for selected earthworm species (*Metaphire californica, Amynthas homochaetus, Amynthas pecteniferus*, and *Amynthas heterochaetus*) that inhabit metal-polluted soils in a subtropical area (Hunan Province) of South China. The earthworms had high plasticity in inhabiting *in situ* contaminated areas and showed high uptake of the heavy metals and capability for their accumulation in the tissues. The bioaccumulation factor (BAF) were greatest for cadmium and ranked as follows: Cd (10.6–18.8) > > Zn (1.15–1.75) > Cu (1.01–1.35) > Pb (0.56–0.95). Earthworm species with the similar BAF of heavy metals ($p > 0.05$) belong to the same ecological group. Within individual groups, Cd, Cu, Zn, Cu, and Pb concentrations in earthworms are consistently predicted by total and extractable fraction (DTPA-extractable) in soil. Our results provide insights into the ecological relationships and variations in the uptake and accumulation of heavy metals in different earthworm species in contaminated soils in China.

1. Introduction

Soil heavy metal contamination is a common problem in terrestrial ecosystems that are contaminated by atmospheric deposition and industrial activities (Wei and Yang, 2010; Xu et al., 2017). In China, it has been estimated that nearly 20% of farmland soil has been polluted by heavy metals (Zhao et al., 2015; Shen et al., 2017; Liu et al., 2017), with particularly severe pollution occurring in the Hunan Province of South China. In addition to concerns over heavy metal uptake by crop plants, better understanding of the potential for food chain (Zhao et al., 2015) transfer via earthworms and other soil fauna has become a top priority. Numerous studies have recently examined the potential risks of heavy metals and metalloids (Schreck et al., 2012; Sołek-Podwika et al., 2016), and the evaluation of methods for the remediation of contaminated soils are being tested to ensure food safety (Williams, 2009; Austruy et al., 2013; Liu, 2013). However, relatively few studies have been conducted to examine the relationships between heavy metal levels in contaminated soils and different earthworm species within different ecological groups having different habitats and feeding strategies.

Earthworms are considered to be important bioindicators for risk assessment and have been used in ecotoxicological studies to assess the potential for bioaccumulation and food chain transfer of heavy metals (Brown et al., 2004; Vijver et al., 2005; Suthar et al., 2008; Pérès et al., 2011). Due to their intimate contact with the soil, both externally and internally, earthworms readily take up and bioaccumulate many heavy metals including Cd, Zn, Cu, and Pb (Li et al., 2010; Nannoni et al., 2014), with several species showing high tolerance to contaminated soils, suggesting differences related to ecological groups or physiological tolerance to heavy metals (Nahmani et al., 2007; Holmstrup et al., 2011; Luo et al., 2014; Demuynck et al., 2014; Sivakumar, 2015; Nirola et al., 2016). Differences in bioaccumulation of various metals also appears to be related to differences in metal bioavailability (Lanno et al., 2004; van Vliet et al., 2005; Hobbelen et al., 2006; Li et al., 2010) as affected by environmental factors affecting solubility (pH and redox) and metal complexation with soil organic matter (Nahmani et al., 2007; Nannoni et al., 2014; Liu et al., 2017). A common method for estimating bioavailable concentrations of heavy metals uses soil extraction with the metal chelator, diethylenetriaminepentaacetic acid (DTPA), which has been developed into a formal test method to address the availability of heavy metals to earthworms (Lebourg et al., 1996; Dai et al., 2004).

In general, accumulation of metals by earthworm species occurs through two pathways (Nannoni et al., 2016), which include absorption

* Corresponding author.
E-mail addresses: qiaoyh@cau.edu.cn, qiaoyh0095@163.com (Y. Qiao).

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Table 1
Soil properties and heavy metal concentrations of the sampling sites (n = 5, SE).

Sites	pH	CEC (cmol kg^{-1})	SOC (g kg^{-1})	TN (g kg^{-1})	Cd (mg kg^{-1})	Zn (mg kg^{-1})	Cu (mg kg^{-1})	Pb (mg kg^{-1})	DTPA-Cd (mg kg^{-1})	DTPA-Zn (mg kg^{-1})	DTPA-Cu (mg kg^{-1})	DTPA-Pb (mg kg^{-1})
S1	6.0	18.1 ± 0.94	21.2 ± 0.97	2.96 ± 0.21	0.81 ± 0.17	123 ± 7.31	24.9 ± 1.39	66.8 ± 2.10	0.18 ± 0.05	25.3 ± 3.32	3.09 ± 0.23	4.38 ± 0.26
S2	5.3	16.1 ± 0.39	20.8 ± 0.99	3.05 ± 0.27	1.31 ± 0.16	130 ± 16.0	28.8 ± 3.17	78.6 ± 6.90	0.22 ± 0.25	23.3 ± 3.96	2.37 ± 0.21	5.16 ± 1.10
S3	5.3	16.0 ± 0.33	18.2 ± 2.08	3.17 ± 0.10	1.75 ± 0.06	131 ± 10.4	55.0 ± 8.98	67.2 ± 2.24	0.26 ± 0.14	25.7 ± 3.91	8.37 ± 2.20	4.22 ± 0.83
S4	6.2	18.9 ± 0.72	18.5 ± 1.47	2.87 ± 0.36	2.39 ± 0.17	140 ± 7.08	19.5 ± 1.83	64.8 ± 4.95	0.57 ± 0.15	22.8 ± 5.73	1.71 ± 0.17	5.09 ± 0.26
S5	6.1	18.8 ± 0.82	18.5 ± 1.05	2.92 ± 0.26	3.45 ± 0.70	164 ± 32.9	207 ± 13.8	101 ± 10.5	1.44 ± 0.53	23.2 ± 9.85	16.4 ± 1.76	22.5 ± 8.16
S6	5.8	17.4 ± 0.98	18.3 ± 0.94	2.11 ± 0.29	4.03 ± 0.96	127 ± 10.7	20.1 ± 1.38	58.4 ± 3.95	1.27 ± 1.40	12.7 ± 3.58	2.93 ± 0.43	5.84 ± 0.35
S7	6.7	15.4 ± 0.33	17.0 ± 1.40	2.18 ± 0.29	4.30 ± 0.96	227 ± 22.4	24.4 ± 1.10	78.7 ± 5.34	2.21 ± 0.70	86.6 ± 13.3	1.61 ± 0.14	6.31 ± 0.56
S8	5.6	17.1 ± 0.35	15.8 ± 1.43	2.39 ± 0.25	4.53 ± 0.37	129 ± 1.99	23.9 ± 1.82	61.3 ± 4.38	2.08 ± 0.36	19.1 ± 1.78	2.49 ± 0.67	3.16 ± 0.58
S9	6.3	19.4 ± 0.58	19.5 ± 0.69	2.71 ± 0.12	4.65 ± 0.78	131 ± 4.34	39.8 ± 1.65	127 ± 18.3	1.75 ± 0.77	11.9 ± 1.54	5.04 ± 0.40	22.9 ± 2.21
S10	6.0	18.3 ± 0.48	21.4 ± 0.74	3.02 ± 0.22	5.11 ± 1.01	276 ± 26.6	55.8 ± 3.16	158 ± 8.87	1.16 ± 0.71	127 ± 18.3	6.01 ± 1.39	22.7 ± 11.7
S11	6.5	20.1 ± 0.28	20.8 ± 0.42	2.95 ± 0.26	7.49 ± 1.47	160 ± 10.9	216 ± 22.7	429 ± 32.2	2.17 ± 1.10	28.8 ± 8.18	17.5 ± 2.95	32.3 ± 5.76
S12	6.7	15.4 ± 0.39	18.6 ± 2.07	2.07 ± 0.21	17.8 ± 2.57	374 ± 42.4	26.6 ± 1.56	139 ± 16.4	9.73 ± 4.00	53.6 ± 14.1	2.57 ± 0.24	15.2 ± 1.88
S13	6.0	18.60 ± 2.02	25.2 ± 2.68	2.97 ± 0.21	20.4 ± 7.00	571 ± 126	105 ± 20.8	334 ± 43.4	10.6 ± 5.03	179 ± 25.5	7.74 ± 1.78	63.5 ± 5.29
Threshold[a]					0.3	200	50	250				

Abbreviations: CEC, cation exchange capacity; SOC, soil organic carbon; TN, total nitrogen; DTPA-Cd, DTPA-Zn, DTPA-Cu and DTPA-Pb represent the DTPA-extractable Cd, Zn, Cu and Pb concentrations in soils, respectively.
[a] Threshold based on Chinese Environmental Quality Standard for Soils (GB 15618-1995)(State Environmental Protection Administration of China,1995).

following dermal contact or else by ingestion of organic matter and adsorption through the gut tissues (Spurgeon and Hopkin, 1996; Saxe et al., 2001; Peijnenburg et al., 1999; Vijver et al., 2003; Morgan et al., 2004; Hobbelen et al., 2006; Lanno et al., 2004). Because of the diversity of ecological characteristics and feeding habits of different earthworm species, the relative importance of dermal contact versus gut adsorption may vary (Nannoni et al., 2011). The earthworm species are generally classified into three broad ecological groups. These include epigeic earthworms that both feed and burrow in the litter layer on the surface of soils, endogeic earthworms that primarily inhabit the organic matter rich top soil profile (Suthar et al., 2008), and anecic that move both vertically and horizontally in the soil and that form burrows in lower compartments of the soil profile (Ireland and Richars, 1977; Bouché, 1977; Ash and Lee, 1980; Morgan and Morgan, 1999; Curry and Schmidt, 2007). Due to differences in heavy metal distributions and bioavailabilities in soil fractions, it can be hypothesized that earthworms in different ecological groups will have different exposures to heavy metals and different potentials for metal bioaccumulation (Suthar et al., 2008; Li et al., 2010). Given these differences in ecological niches, it is of interest to determine the relationship between total soil or DTPA-extractable levels of heavy metals and the internal metal concentrations in earthworm tissues, and to investigate the bioaccumulation factors for individual metals by different earthworm species.

Meanwhile, many field studies have been conducted to examine the bioaccumulation of metals by individual earthworm species (Hobbelen et al., 2006; Nahmani et al., 2009; Pérès et al., 2011; Lévêque et al., 2013; Nannoni et al., 2014), variations in the uptake and accumulation of metals by different earthworm species in the same soil have rarely been studied. In this case, it is especially to relevant to use field-contaminated soils where metal bioavailability to different ecotypes reflects long term natural processes that affect the bioavailability of metals in soil.

To examine the relationships between heavy metal concentrations and bioaccumulation, the study conducted here examined the different species and bioaccumulation levels of cadmium (Cd), zinc (Zn), copper (Cu) and lead (Pb) in soils having different levels of contamination. The specific aims of the study were: (i) to determine the bioaccumulation of Cd, Zn, Cu and Pb in the predominant earthworm species. (ii) to characterize the eco-physiological patterns of bioaccumulation of heavy metals in different earthworm species; (iii) to determine the relationships between tissue metal concentrations in earthworms and total and DTPA extractable metal concentrations across a range of contaminated sites in South China.

2. Materials and methods

2.1. Site description

The study area has a typical subtropical monsoon climate, with mean annual temperatures ranging between 2 °C and 35 °C and an average annual precipitation ranging between 1200 and 1700 mm. The sources of pollution in the study area were mainly related to the mining industry which was active in the area for ten years, but which has since been halted in recent years. Heavy metals were introduced into the soils in this area mainly from wet and dry atmospheric deposition from smelters, and land contamination from waste water and mine tailings. The sites chosen in this study for evaluating the effects of *in situ* soil pollution and any possible local deposition of heavy metals. A total of 65 samples were collected from 13 sites that showed variations in soil metal concentrations. Earthworm species counts and soil samples were collected from each site in early November 2015.

2.2. Soil properties

Five soil samples were collected (0–25 cm) at each site to determine the soil properties and metal concentrations. The soil samples were air