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Bioaccumulation of Heavy Metals by Grasshoppers and a Mantid Along a Pollution Gradient

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Abstract. The concentrations of Cd, Cr, Cu, Fe, Ni, Pb, and Zn were studied in soil, plants, three grasshopper species (*Aiolopus thalassinus* (Fabricius, 1781), *Calephorus compressicornis* (Latreille, 1804), and *Acrotylus patruelis* (Herrich-Schäffer, 1838)), and a mantid (*Miomantis paykullii* Stål, 1871), to estimate the scale of spatial influence of a point pollution source, to investigate metal bioaccumulation up the soil–plant–grasshopper–mantid food chain, and to assess these insects as ecological bioindicators. Plant, insect, and soil sampling was undertaken simultaneously from grass margins of an arable farmland near an industrial complex in Al-Tebbin. Correlation analyses revealed a pollution gradient, with samples closest to the industrial activities having the highest concentrations of heavy metals. Sigmoid curves indicated that the influence radius of the industrial zone for heavy metal contamination in soil, plants, and grasshoppers was in the range of 3.0–4.0 km. The average concentrations of Cd, Cr, Cu, Fe, Ni, Pb, and Zn in the three grasshopper species were 4.9, 4.0, 73.9, 210, 5.2, 12.0, and 224.2 mg/kg, respectively, while those in the predaceous mantid were 9.0, 7.9, 106.4, 400, 14.8, 23.0, and 463.7 mg/kg, respectively. The metal content did not increase from soil to grasses, but at plant–primary consumer level, Cd, Cu, Pb, and Zn were biomagnified, and, in general, all metal concentrations increased with an increase in the trophic level (i.e., secondary consumer). In light of this site-dependent accumulation of heavy metals and their biomagnification patterns, the investigated acridids can be considered bioindicators of Cd, Cu, Pb, and Zn pollution.

Key words: bioaccumulation, bioindicator, food chain, grasshoppers, industrial activities, influence range.

Introduction

Environmental issues related to heavy metal pollution have become a widespread serious problem (SOFIANSKA *et al.*, 2013). Metals are released into the environment as a consequence of a wide range of industrial activities and combustion of fossil fuels. These different sources contribute to the load of metal pollutants in terrestrial and aquatic food chains (GALL *et al.*, 2015). Dust fall, bulk precipitation, and gas adsorption processes are responsible for the transportation of heavy metals from the atmosphere to the soil and plants (MALIZIA *et al.*, 2012; RAKIB *et al.*, 2014). In several localities, the emission of pollutants into the air by industrial activities has reached levels that are toxic to plant, animals, and humans. The measuring and monitoring of the environmental pollution in rural and urban ecosystems can help in the assessment of air quality and can be used to quantify the effective range of pollution sources (ROSS, 1987). Heavy metal inputs into agricultural soils have gradually increased

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over the last century, as a result of atmospheric deposition, disposal of sewage sludge, and agricultural practices (fertilizers, liming materials, pesticides, manures, and compost) (GALL *et al.*, 2015).

Element concentrations in living organisms are generally low, except in the vicinity of heavy metal pollution sources (ZHANG *et al.*, 2012). Nevertheless, because of the long retention times of toxic substances in the ecosystems, the transport of even small amounts through the food chain increase the concentration of these metals and can cause deterioration of the environment. Some heavy metals (e.g., Cu, Fe, Se, and Zn) are involved in biochemical and physiological functions in plants and animals, and therefore are essential in trace amounts (MARSCHNER, 2012). However, when present in excess, these become toxic to living organisms, and possibly dangerous to human health through their transfer via the food chain (CHAFFAI & KOYAMA, 2011). Other metals (e.g., Cd and Pb) are not known to have any essential function, and are toxic even at low concentrations (NORDBERG *et al.*, 2014). Heavy metals, such as Cd and Pb are responsible for certain diseases and cause a number of health hazards in humans (AGARWAL, 2009). Moreover, they have been classified as carcinogenic for humans and wildlife (BEYERSMANN & HARTWIG, 2008).

Pollution zones with high metal content in soil and biota are often found near industrial areas (DUDKA *et al.*, 1996). The ability of some animals to accumulate these metals, and establish a reliable relationship with the concentration of the metal in their surroundings, makes them useful indicators of environmental pollution.

Grasshoppers are herbivorous insects, feeding mainly on grass leaves, and account for 20–30% of the biomass of all arthropods. Therefore, they play a significant role in accumulating and transferring toxic metals to higher trophic levels through the food chain (DEVKOTA & SCHMIDT, 2000). Several studies have reported excessive concentrations of heavy metals in terrestrial food chains (HSU *et al.*, 2006; HECKEL & KEENER, 2007; ZHUANG *et al.*, 2009; JUNG & LEE, 2012; NICA *et al.*, 2012; BOSHOFF *et al.*, 2015; SIMON *et al.*, 2016; CONTI *et al.*, 2017; DAR *et al.*, 2017). However, few studies have focused only on grasshoppers. Some studies have been undertaken on the accumulation of metals in soil, plants, and grasshoppers, where the concentrations of metals are usually highest at the sites closest to the pollution sources (DEVKOTA & SCHMIDT, 2000; KARADJOVA & MARKOVA, 2009; NATH *et al.*, 2011; ZHANG *et al.*, 2012; ZHANG *et al.*, 2014). In addition, heavy metals could be potentially accumulated in predators that feed on grasshoppers (e.g., mantids), and transported to organisms higher up the food web (ZHENG *et al.*, 2008; ZHANG *et al.*, 2009, 2010).

The main purpose of this study was to specify the effective range of heavy metal contamination from industrial areas, and to investigate Cd, Cr, Cu, Fe, Ni, Pb, and Zn bioaccumulation and transportation up the soil–plant–grasshopper–mantid food chain in grass margins of arable farmland near an industrial complex in Al-Tebbin District of Egypt. We also assessed the grasshoppers (*Aiolopus thalassinus* (Fabricius, 1781), *Calephorus compressicornis* (Latreille, 1804), and *Acrotylus patruelis* (Herrich-Schäffer, 1838)) and a predator (*Miomantis paykullii* Stål, 1871) for suitability as ecological bioindicators for heavy metal contamination.

Materials and Methods

Study area. The study area was located east of the River Nile, 20 km south of Cairo, in Al-Tebbin region in the Helwan Province. The population of Al-Tebbin District is approximately 0.5 million people (2008); it is considered the biggest industrial zone in Egypt, with about 16.5% of the total industrial activities for the country. The main industrial activities include ferrous and nonferrous metallurgical work, coke factory, chemical and cement industry (MOHAMED *et al.*, 2011; SOLIMAN *et al.*, 2017). With the steady increase of industrial emissions, in addition to high temperatures, lack of rain, and predominantly low wind speed, this area has severe air pollution problems.

Sample collection and preparation. During the summers (July and August) of 2012 and 2013, soil, plant, and insect samples were collected from the grass strips along the edges of farmland in Al-Tebbin region.

Samples were taken from nine sites at different distances, up to 10 km downwind and 3.5 km upwind, from the main sources of industrial pollution (Fig. 1). *A. thalassinus* (356 specimens), *C. compressicornis* (219), *A. patruelis* (131), and *M. paykullii* (41) were collected, euthanized with alcohol, and preserved in a refrigerator at 4 °C until used. Three common grasses in the study area, *Paspalum distichum* L., *Brachiaria deflexa* (Schumach.) Robyns, and *Hyparrhenia hirta* (L.) Stapf, were sampled at the same sites and stored in polyethylene bags. The leaves and stems of the three species were washed with ultrapure water to remove metals attached to the surface. For each site, five soil samples were randomly collected from the upper soil layer (0–15 cm) and thoroughly mixed, with about 1 kg of soil used for lab analysis. The soil, plants, and arthropods were oven dried (70 °C) until they reached a constant weight, and were then ground to a homogenous powder and preserved in polythene bags. The sampling positions were recorded using GPS technology, and ArcGIS 9.3 was used to develop the location map.

Chemical analysis. The soil digestion was based on the protocol described by SOLIMAN *et al.* (2017). A soil sample (0.5 g, accurately weighed) was placed in a digesting flask and a pre-digestion step was run at room temperature for 24 hrs with 12 mL of 37% HCl: 65% HNO_3 (3: 1) mixture. Then, in a fume cupboard, the suspension was digested to near dryness on a thermostatically controlled hot plate at 90 °C. To the hot residue, 2.5 mL of 37% HCl and 2.5 mL of 30% H_2O_2 were added to complete the digestion, and the resultant mixture was heated again and then cooled to ambient temperature. The flask walls were washed with 10 mL of ultrapure water and the obtained suspension was filtered through Whatman filter paper (No. 41) in a volumetric flask, diluted to 50 mL, and stored in polyethylene bottles at 4 °C for analyses. The digestion of plant and insect samples were prepared in a similar way, but were allowed to stand for 24 hrs with 10.0 mL of 65% HNO_3, and only 2.0 mL of 30% H_2O_2 was added to aid the digestion of the organic matter. The filtered solutions were diluted up to 25 mL. The concentrations of Cd, Cr, Cu, Fe, Ni, Pb, and Zn were determined by inductively coupled plasma (ICP-AES-Jobin Yovin ultima2, France). Wavelengths and detection limits of the ICP for the analyzed metals were: 226.502 nm and 0.0023 mg L^{-1} for Cd; 220.353 nm and 0.028 mg L^{-1} for Pb; 324.754 nm and 0.0036 mg L^{-1} for Cu; 213.856 nm and 0.0012 mg L^{-1} for Zn; 231.604 nm and 0.01 mg L^{-1} for Ni; 205.552 nm and 0.0041 mg L^{-1} for Cr; and 259.940 nm and 0.0041 mg L^{-1} for Fe, respectively. Certified reference materials (NIST 2709, 1547, and 1577) and a reagent process blank were performed for each analytical batch to assess the analytical accuracy. In addition, mean values of three replicates were taken for each determination. Metal concentrations were expressed as mg/kg dry weight. Chemicals, stock solutions, and reagents were all of analytical grade and were obtained from Merck. All glassware and plastic materials were washed with distilled water before use, soaked in 2N nitric acid overnight, and then rinsed thoroughly with ultrapure water.

Statistical analyses. Only a limited number of data sets revealed a good fit to normal distribution and there were large differences in variance between treatment groups. Therefore, Kruskal-Wallis H-test was used to test for significant differences in metal concentrations between sites, while Mann-Whitney U-test was used to compare the total metal concentrations between the soil, plants, grasshoppers, and mantid, and mean metal bioaccumulation factors between trophic levels (plants, grasshoppers, and mantid) (SPSS STATISTICS, 2008). Correlation analyses of heavy metal concentrations with the distance from the industrial area, and between soil, plant, and grasshopper were conducted using Spearman's rank correlation (SPSS STATISTICS, 2008). In all tests $p \leq 0.05$ was considered significant. All treatments were replicated three times in the experiment. In this study, the bioaccumulation factor for plants was calculated as the ratio of the metal concentration in shoots to that in the soil (CUI *et al.*, 2007), and the bioaccumulation factor (B) for grasshoppers and the mantid was defined as:

$$B = Cn/Cn\text{-}1$$

where Cn is the total metal concentration in the organism from trophic level n, and Cn-1 is the total metal concentration in the food of this organism from trophic level n-1 (LASKOWSKI, 1991).

Results and Discussion

Metal content in the soil, plants, grasshoppers, and mantid. Metal concentrations in the soil–plant–insect food chain at different sampling sites are shown separately in Fig. 2 and collectively, with range of variation, in Table 1. The heavy metal content varied significantly among sample sites. Cu (H = 24.942, p = 0.002 for soil; H = 25.079, p = 0.002 for *C. compressicornis*; H= 24.335, p = 0.002 for *A. patruelis*) and Zn (H = 25.153, p = 0.001 for plant; H = 24.677, p = 0.002 for *A. thalassinus*) had the highest significant differences among the sites, while Fe (H = 22.783, p = 0.004 for soil) and Cr (H= 19.691, p = 0.012 for plant; H = 17.010, p = 0.030 for *A. thalassinus*; H = 19.744, p = 0.011 for *C. compressicornis*; H = 20.179, p = 0.010 for *A. patruelis*) had the lowest significant differences among the sites.

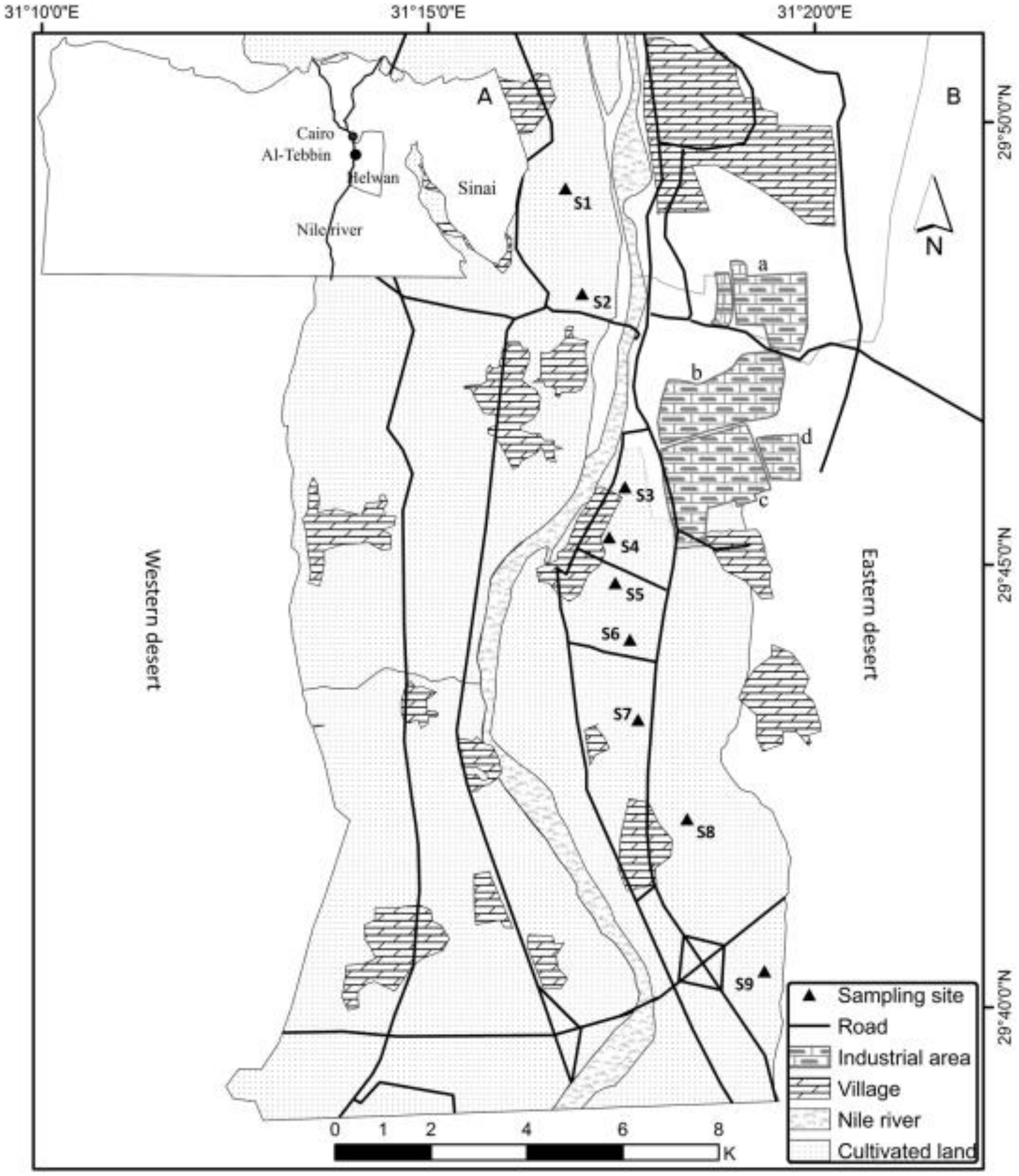


Fig. 1. A- Location of Al-Tebbin area southeast of Cairo. B- Location of industries and sampling sites within the Al-Tebbin area; (a) cement industry; (b) iron and steel industry; (c) coke and chemical industry; (d) metallurgical work.

The metal content in the soil, plants, and grasshoppers from sites S2, S3, S4, and S5 was higher than elsewhere, especially for S3, which was the site nearest the industrial area. In addition, Spearman's rank correlation analysis showed significant ($p < 0.05$) negative relationships between metal content in the soil, plants, and grasshoppers and the distance from the industrial area (Table 2) for Cd, Fe, Pb, Ni, and Zn in the soil and plants; Cd in the three grasshoppers; Pb in *A. thalassinus*; and Cu in *C. compressicornis*. The data showed that Cd, Fe, Pb, Ni, and Zn were the main heavy metal pollutants in this region, and, in general, the levels of heavy metals decreased with increasing distance from the pollution sources. This result is in agreement with HELIÖVAARA & VÄISÄNEN (1990), who found a continuous decrease in metal levels in the European pine sawfly adult, immature stages, larval food plants, and feces, with increasing distance from industrial plants. In our study, the relationships between distance from the industrial area and each of the heavy metals (except Cr) in the soils, plants, and grasshoppers were fit using a negative sigmoid model (Fig. 3); the metal concentrations decreased greatly with increasing distance until 3.0 km, when the curve began to flatten, with metal content stable after the distance exceeded 4.0 km. These results are similar to those of ZHANG *et al.* (2012), who found that the influence radius of zinc smelting on Cd and Pb contamination in soil, plants, and grasshoppers was in the range of 2.0–4.0 km. It should be noted that the predominant wind direction in our study area was northerly. This resulted in the contamination of the soil–plant–insect system in the area south of the industrial complex with excessive levels of heavy metals recorded in the soil: 67.5 mg/kg for Cd; 97.8 for Cr; 292 for Cu; 58900 for Fe; 160 for Pb; 80.0 for Ni; and 605 for Zn. S1 and S2, the areas mainly affected by the cement industry, showed relatively lower concentrations than S3, S4, and S5, which were the sites affected by the other emission sources. This may be because cement kiln dust does not contain high concentrations of heavy metals (ALI *et al.*, 1992). However, these areas were still enriched with relatively higher concentrations of toxic metals than those detected in the sites far from pollution sources. This might be explained by the southerly winds, which carried emissions from the metallurgical (iron and steel) and coke industries. When compared with the background levels of heavy metals in unpolluted soil in Egypt (ELSOKKARY & LAG, 1980; ELSOKKARY, 1996; ABOULROOS *et al.*, 1996; ABDEL-SABOUR & ABDEL-BASSET, 2002; ABDEL-SABOUR & ZOHNY, 2004), sites that were considered far away from industrial sources in our study were also found to be enriched with heavy metals. Our results indicated that this area is highly polluted by heavy metals, and the main source of pollution is the industrial complex.

In the present study, grasshoppers clearly showed site-dependent metal accumulation patterns for Cd, Cu, and Pb, and these results may support the thought that the body burden of heavy metals in animals reflects site pollution. Several studies have reported that there are physiological mechanisms that aid in the regulation of the essential elements in insect metabolism and prevent toxic levels (NEWMAN & UNGER, 2003; NFON *et al.*, 2009; BJERREGAARD *et al.*, 2014). This may explain the site-independent accumulation for Fe, Ni, and Zn in grasshoppers despite their significant inverse relation in soil and plants for the distances far from the industrial area. The soil Cd, Cu, and Pb concentrations showed significant positive correlations with those in plants and the herbivore insects, while the Ni was only correlated with *A. thalassinus* and *C. compressicornis*. Similarly, a strong relationship was also detected for metal concentrations in plants and grasshoppers, particularly for Cd, Pb, and Cu, which were significantly correlated with the three species of grasshoppers; in the case of Cr, Fe, Ni, and Zn, the relationship was only significant for one or two species (Table 3). The values for Zn in soil also showed significant relationships with plant Zn. The correlation analysis suggested that Cd, Cu, Pb, and Zn in the soil–plant–grasshopper system in Al-Tebbin District might originate from the same source; metal content in grasshoppers came mainly from their plant food.

Acridid grasshoppers are exposed to heavy metals via different paths, including from plants through their feeding habit and directly from the soil through oviposition behavior (DEVKOTA & SCHMIDT, 2000). In the Biesbosch floodplains, NOTTEN *et al.* (2005) suggested that metal transfer from plant leaves to the snail, *Cepaea nemoralis* (Linnaeus, 1758), is more important than is transfer from the soil. Likewise, in our study, strong relationships were found between the plants and grasshoppers for all metals, and this may confirm the expectation that food is the primary factor determining metal bioaccumulation in animals (NOTTEN *et al.*, 2005; CHARY *et al.*, 2008; ZHANG *et al.*, 2012).

Three sites were selected for measuring total heavy metals in the mantid (S4, S5, and S8). Although Cd, Cu, Pb, and Zn content was comparatively higher in *M. paykullii* collected from S4 and S5 than in mantids collected from S8 (Fig. 2), metal levels were not significantly different at the different sites ($p > 0.05$, H-test). Apart from Cr and Ni, metal concentrations in soil, plants, grasshoppers, and mantids from all the sampling sites were in the following order: Fe > Zn > Cu > Pb > Cd (Table 1). Based on the average concentrations, there were no significant differences between grasshopper taxa in the concentrations of Cd, Pb, or Ni (Table 1). Cu and Zn concentrations were significantly lower, and the Cr concentration was significantly higher in *C. compressicornis* than in *A. thalassinus* and *A. patruelis*. The results also showed that significantly more Fe was accumulated in *A. thalassinus* than in the other two grasshoppers (Table 1).

Bioaccumulation of metals through the food chain. Table 4 shows the average and range of metal bioaccumulation factors for plants and arthropods. Significantly lower concentrations of heavy metals were noted for grasses than for soil (Table 1). The bioaccumulation factors for heavy metals in grasses were less than 1.0 (varied between 0.04 and 0.59), which indicated that metals did not accumulate in grasses. Metals that were present were in the following order: Cu > Zn > Cd > Pb ≈ Ni > Cr > Fe. From soil to plant leaves, the bioaccumulation factor was significantly higher for Cu and significantly lower for Fe than noted for the other heavy metals ($p < 0.05$, U-test). The bioaccumulation factors of the other metals (except Cr) were not significantly different ($p > 0.05$, U-test).

The bioaccumulation factors were higher for herbivorous insects than for plants. Although the trend of bioaccumulation factors for metals in each grasshopper was different, all three species had significantly higher Zn bioaccumulation factors (3.8, 2.7, and 3.4 for *A. thalassinus*, *C. compressicornis*, and *A. patruelis*, respectively) and lower Fe bioaccumulation factors (0.32, 0.19, and 0.20 for *A. thalassinus*, *C. compressicornis*, and *A. patruelis*, respectively). Cd, Cu, and Pb in the three grasshoppers were found in higher concentrations than in plants, and their bioaccumulation factors were > 1 and < 2. In the case of Cr, Fe, and Ni, the accumulation in grasshoppers never exceeded that of plants, and the bioaccumulation factors were < 1. Except for Cu, our results are broadly consistent with earlier reports on the European pine sawfly, *Neodiprion sertifer* Geoffroy, 1785, (Harjavalta, Finland) where HELIÖVAARA & VÄISÄNEN (1990) found higher accumulation of Cd in *N. sertifer* than in pine needles, while the levels of Cu, Fe, and Ni were higher in plant food than in *N. sertifer*. Although Cd and Pb concentrations decreased along the soil–plant–grasshopper food chain in a study conducted by ZHANG *et al.* (2014) in South China, their results also agreed with our data in the case of Cu and Zn. Our study showed that Cd, Cu, Pb, and Zn accumulated in the grasshoppers, with Zn having the highest bioaccumulation factor, while, Cr, Fe and Ni were not accumulated. The influence of homeostatic regulation on metal concentrations in organisms may explain the grasshoppers' low accumulation of Fe and Ni and the biomagnification of Cu and Zn. In general, uptake, accumulation, and excretion rates of these essential metals in terrestrial animals are regulated by metallothionein and metallothionein-like proteins (NEWMAN & UNGER, 2003; NFON *et al.*, 2009; BJERREGAARD *et al.*, 2014), and, based on the physiological requirements of these elements in the organisms, their bioaccumulation in the food chain is detected. Usually, naturally occurring concentrations of Ni seem to be sufficient to meet intake requirements because of their low physiological requirement (Phipps *et al.*, 2002). However, essential metals, such as Zn, are usually biomagnified up the food chain through invertebrates that are known to be efficient accumulators or that lack the necessary regulatory and detoxification mechanisms of higher-order animals (Nfon *et al.*, 2009).

Of the three grasshoppers investigated, the highest metal concentrations were found in *A. thalassinus* and the lowest in *C. compressicornis* (Table 1), which suggested slightly larger metal absorption abilities for *A. thalassinus* than for *C. compressicornis* and *A. patruelis*. Even so, the highest accumulations of Ni and Cr were found in *A. patruelis* and *C. compressicornis*, respectively, and the lowest accumulation of Cr was found in *A. thalassinus*. Based on the aforementioned results, *A. thalassinus* may perform better as a test animal for the evaluation of metal pollution. In this sense, heavy metal accumulation in herbivorous insects may differ among metals and species. For example, in previous studies, Cd bioaccumulation factors for grasshoppers (Devkota & Schmidt, 2000; Zhang *et al.*, 2009, 2012) were more than 1.0 for all grasshoppers, but Pb accumulation factors were less than 1.0 for the same species; however, Pb concentration was 1.3 times higher in *Acrida chinensis* (Westwood, 1842) than in its food plant, *Echinochloa crusgall* (L.) P. Beauv (Zhang *et al.*, 2009).

Praying mantids are predators feeding on other insects and arthropods (Hurd, 1999), living solitary on trees, shrubs, and grasses. *M. paykullii*, the secondary consumer in the studied food chain, belongs to the group that prefers green grasses and small plants (Sawaby *et al.*, 2010), and thus, mainly feeds on *A. thalassinus*, *C. compressicornis*, and *A. patruelis*, the dominant grasshopper species in the study area. The metal concentrations were significantly higher in *M. paykullii* than in the three grasshoppers, particularly for Zn and Cu, which were high enough to exceed the concentrations of Zn and Cu in the soil (Table 1). Nonetheless, in *M. paykullii*, the bioaccumulation factors were comparatively lower in Zn and Cu than in the other elements. In the present work, Cd and Pb concentrations in mantid were, on average, 9.0 and 23.0 mg/kg, respectively. The concentrations for Cd and Pb were much lower in our mantid than in the mantids and spiders in a similar study from Zhang *et al.* (2009). Cd, Cu, Pb, and Ni concentrations were much lower in the mantid than in other predators (waterstriders, antlions, ants, and dragonfly larvae) collected from sites close to an iron and steel factory in Koverhar, Hanko Peninsula, Finland (Nummelin *et al.*, 2007). In contrast, Fe concentrations were much higher in the other predators than in our mantid, while Zn concentrations were relatively the same between the two studies. Diverse biological factors and environmental conditions might contribute to these differences. Furthermore, organisms have various physiology and metabolic requirements, and consequently, methods for tolerating high concentrations of metals are different (Phipps *et al.*, 2002), encompassing avoidance of contaminated substrates, metabolic processes limiting metal uptake, and the uptake and compartmentalization of metals within the body (Bengtsson & Travnik, 1989; Kabata & Pendias, 2001).

The metal bioaccumulation factors of mantid differed among grasshopper species (Table 4), but as a whole (average), all metals had remarkably high accumulation in this predator: 5.0 for Cd; 4.6 for Cr; 4.1 for Fe; 3.5 for Pb; 2.3 for Ni; 2.2 for Zn; and 1.4 for Cu. Biomagnification was higher for Cd than for other metals, and thus biotransfer of Cd through the food chain, via the mantid, was probably more efficient for Cd than for the other metals. According to Roth-Holzapfel (1990), only Cd and Ni biomagnified along the food chain. In addition, Hunter *et al.* (1987) stated that Cd and Cu were highly mobile in the invertebrate food web. Because of their related chemical properties, Cd can be accumulated by replacing Zn in the enzyme carbonic anhydrase, resulting in highly effective absorption of Cd into the bodies of animals (Hopkin, 1989; Roth-Holzapfel, 1990; Devkota & Schmidt, 2000).

Table 1. Mean heavy metal content in soil, plants, and arthropods near the industrial area in Al-Tebbin.

Metal	Soil	Plant	Grasshopper			*Miomantis*
			Aiolopus	*Calephorus*	*Acrotylus*	
Cd	16.2 ± 4.1a (1.9–67.5)	3.5 ± 0.70b (0.34–10.8)	5.4 ± 1.3b (0.47–19.6)	5.0 ± 0.98b (0.40–14.1)	4.4 ± 1.0b (0.28–14.3)	9.0 ± 1.3a (5.4–14.0)
Pb	49.3 ± 9.6a (6.0–160.0)	9.3 ± 1.6b (1.8–27.6)	12.7 ± 1.7b (3.9–29.5)	10.9 ± 2.2b (1.1–28.7)	12.5 ± 3.0b (1.3–43.5)	23.0 ± 5.1a (4.6–36.3)
Cu	104.8 ± 15.8ad (29.0–292.0)	61.3 ± 9.2b (22.1–160.4)	83.9 ± 7.1ad (33.6–146.0)	62.4 ± 5.8bc (29.3–112.7)	75.6 ± 7.2cd (30.0–142.0)	106.4 ± 12.9a (63.0–137.0)
Zn	266.5 ± 36.1ac (98.0–605.0)	96.2 ± 14.7b (20.0–256.0)	288.5 ± 15.9a (136.7–410.0)	153.6 ± 9.7c (106.0–263.2)	230.4 ± 15.4a (123.0–389.0)	463.7 ± 38.3d (326.2–556.0)
Ni	41.5 ± 4.7a (12.0–80.0)	9.9 ± 1.6bd (2.7–23.4)	5.2 ± 0.61bc (1.0–8.3)	4.7 ± 0.74c (1.3–12.7)	5.8 ± 0.97bc (0.92–17.0)	14.8 ± 3.2d (4.8–23.6)
Cr	60.0 ± 5.5a (27.0–116.0)	7.9 ± 1.3b (2.0–23.2)	2.2 ± 0.30c (0.88–5.8)	6.4 ± 4.2b (2.0–20.0)	3.6 ± 0.58d (1.0–8.6)	7.9 ± 1.6b (5.0–12.2)
Fe	29.9 ± 2.6a (11.5–58.9)	1.3 ± 0.20b (0.35–3.5)	0.32 ± 0.03c (0.14–0.63)	0.15 ± 0.01d (0.07–0.23)	0.16 ± 0.01d (0.09–0.27)	0.40 ± 0.08e (0.11–0.66)

Data are presented as mean ± SE in mg/kg dry weight (mg/g for Fe) (range). Different letters within the same row denote significant differences (Mann–Whitney U test, $p < 0.05$).

Table 2. Spearman's rank correlation coefficients for comparisons between the distances from the industrial area and the concentration of each heavy metal in soil, plants, and grasshoppers.

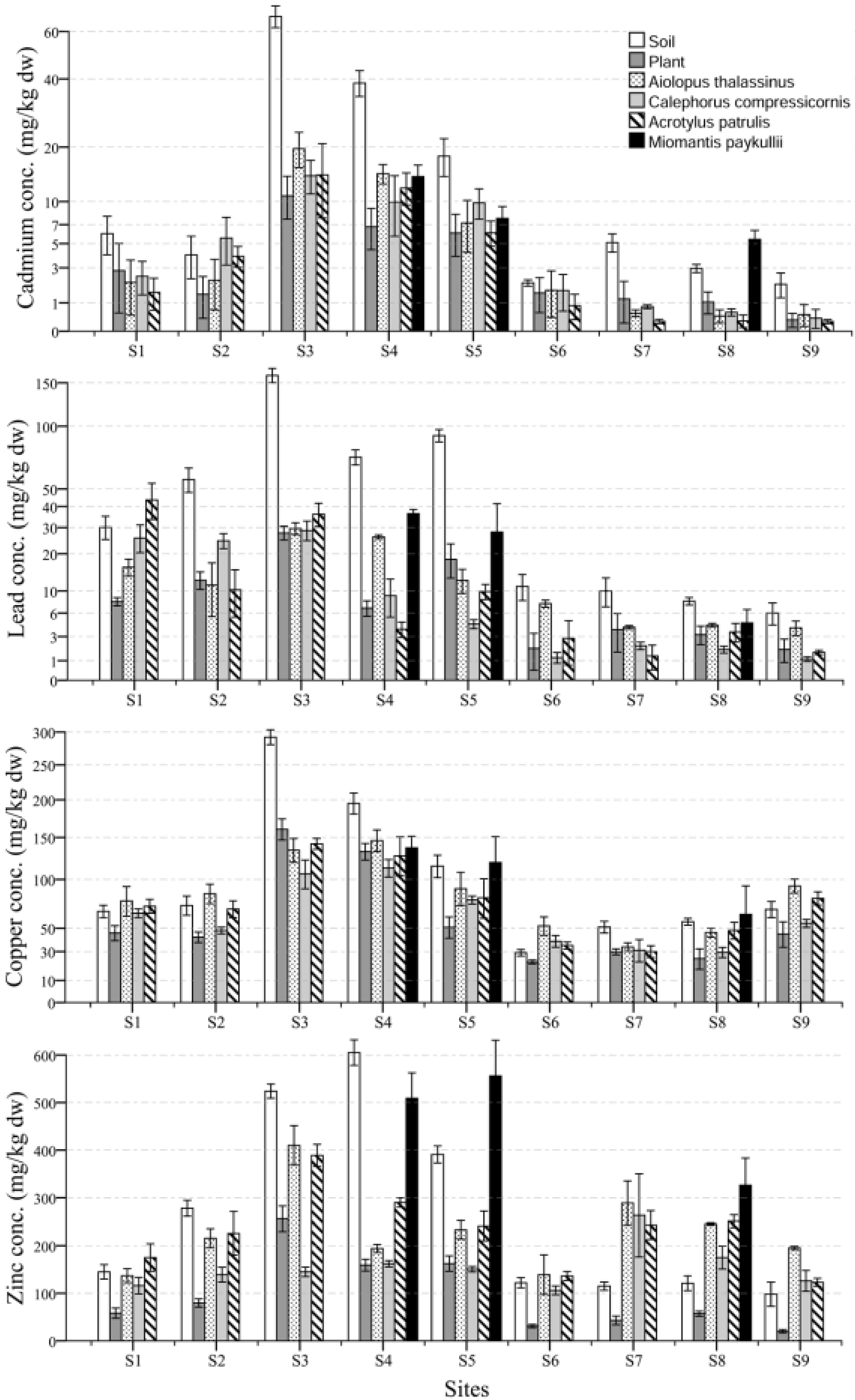
Taxon	Heavy metal						
	Cd	Pb	Cu	Zn	Ni	Cr	Fe
Soil	- 0.783*	- 0.933**	- 0.633	- 0.917**	- 0.433	- 0.159	- 0.767*
Plant	- 0.950**	- 0.700*	- 0.617	- 0.800**	- 0.667*	- 0.500	- 0.750*
Aiolopus	- 0.933**	- 0.867**	- 0.567	- 0.083	- 0.017	- 0.050	- 0.250
Calephorus	- 0.950**	- 0.617	- 0.717*	0.033	- 0.500	- 0.217	0.067
Acrotylus	- 0.933**	- 0.517	- 0.600	- 0.533	0.350	0.600	0.100

Statistically significant at $p < 0.05$ (*) or $p < 0.01$ (**).

Table 3. Spearman's rank correlation coefficients for comparisons between soil, plants, and grasshoppers for heavy metal concentrations.

Metal	Soil–Plant	Soil–Grasshopper			Plant–Grasshopper		
		Aiolopus	*Calephorus*	*Acrotylus*	*Aiolopus*	*Calephorus*	*Acrotylus*
Cd	0.883**	0.850**	0.883**	0.850**	0.933**	0.950**	0.933**
Pb	0.900**	0.900**	0.783*	0.700*	0.783*	0.883**	0.783*
Cu	0.917**	0.883**	0.850**	0.933**	0.833**	0.933**	0.933**
Zn	0.900**	0.000	0.050	0.583	0.333	0.250	0.700*
Ni	0.583	0.817**	0.733*	0.567	0.383	0.917**	0.233
Cr	0.602	0.502	0.368	0.377	0.683*	0.883**	0.150
Fe	0.367	- 0.100	- 0.350	- 0.150	0.667*	0.300	0.000

Statistically significant at $p < 0.05$ (*) or $p < 0.01$ (**).


Soil
Plant
Aiolopus thalassinus
Calephorus compressicornis
Acrotylus patrulis
Miomantis paykullii
Cadmium conc. (mg/kg dw)
Lead conc. (mg/kg dw)
Copper conc. (mg/kg dw)
Zinc conc. (mg/kg dw)
S1
S2
S3
S4
S5
S6
S7
S8
S9
Sites

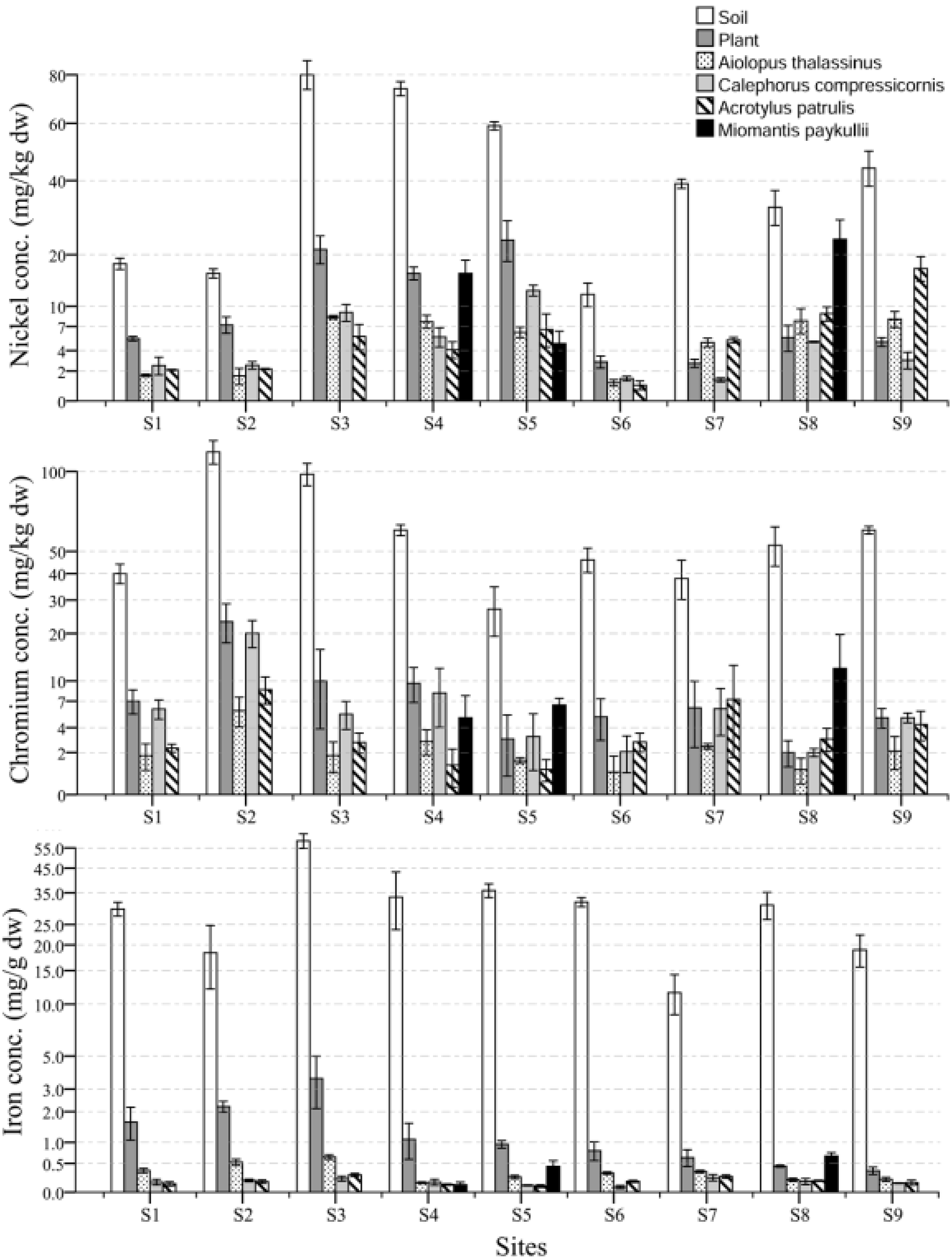


Fig. 2. Heavy metal concentrations in soil, plants, and arthropods at sampling sites near the industrial area in Al-Tebbin. The error bars indicate the standard error and each bar represents the mean of three replicates. (Miomantis was collected only from three sites: S4, S5, and S8).

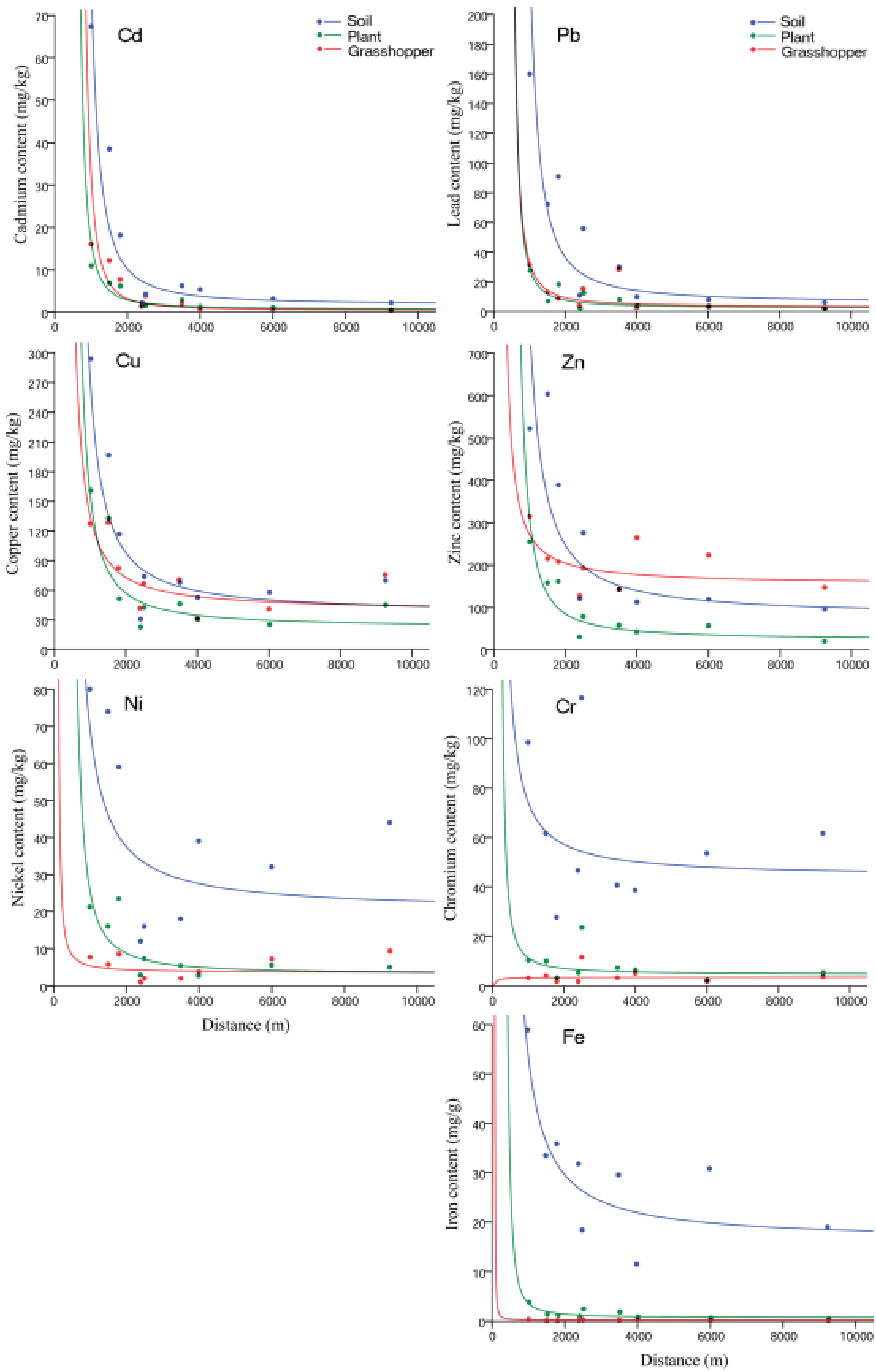


Fig. 3. Metal content in soil, plants, and grasshoppers against distance from the industrial area.

Table 4. Heavy metal bioaccumulation factors for plants, grasshoppers, and the mantid.

Metal	Plant/Soil	Grasshopper/Plant			Mantid/Grasshopper		
		Aiolopus	*Calephorus*	*Acrotylus*	*Aiolopus*	*Calephorus*	*Acrotylus*
Cd	$0.33 \pm 0.06_{A}$	$1.2 \pm 0.19_{AC}$	$1.4 \pm 0.33_{AB}$	$1.0 \pm 0.27_{AD}$	$4.5 \pm 3.4_{A}$	$3.7 \pm 2.6_{A}$	$6.7 \pm 5.4_{A}$
	(0.16–0.72)	(0.46–2.1)	(0.59–3.9)	(0.25–2.7)	(0.96–11.4)	(0.79–8.8)	(1.1–17.6)
Pb	$0.24 \pm 0.03_{A}$	$1.9 \pm 0.41_{A}$	$1.1 \pm 0.31_{AE}$	$1.3 \pm 0.52_{AD}$	$1.6 \pm 0.35_{A}$	$4.3 \pm 1.0_{A}$	$4.5 \pm 2.5_{A}$
	(0.09–0.40)	(0.68–3.9)	(0.24–3.2)	(0.35–5.5)	(1.0–2.2)	(2.5–6.3)	(1.3–9.5)
Cu	$0.59 \pm 0.03_{B}$	$1.6 \pm 0.17_{A}$	$1.2 \pm 0.11_{AB}$	$1.4 \pm 0.12_{D}$	$1.2 \pm 0.13_{A}$	$1.6 \pm 0.27_{A}$	$1.3 \pm 0.12_{A}$
	(0.44–0.76)	(0.84–2.3)	(0.66–1.7)	(0.89–1.9)	(0.94–1.3)	(1.2–2.1)	(1.0–1.5)
Zn	$0.35 \pm 0.03_{A}$	$3.8 \pm 0.94_{B}$	$2.7 \pm 0.71_{B}$	$3.4 \pm 0.58_{B}$	$2.1 \pm 0.39_{A}$	$2.9 \pm 0.54_{A}$	$1.8 \pm 0.29_{A}$
	(0.20–0.49)	(1.2–9.7)	(0.57–6.2)	(1.4–6.1)	(1.3–2.6)	(1.8–3.7)	(1.3–2.3)
Ni	$0.24 \pm 0.04_{A}$	$0.76 \pm 0.21_{CD}$	$0.51 \pm 0.05_{C}$	$0.97 \pm 0.37_{AD}$	$1.9 \pm 0.65_{A}$	$2.6 \pm 1.2_{A}$	$2.4 \pm 0.92_{A}$
	(0.07–0.45)	(0.22–1.8)	(0.35–0.90)	(0.26–3.4)	(0.77–3.0)	(0.38–4.7)	(0.73–3.9)
Cr	$0.12 \pm 0.01_{C}$	$0.32 \pm 0.04_{D}$	$0.83 \pm 0.07_{E}$	$0.61 \pm 0.15_{A}$	$6.1 \pm 3.1_{A}$	$2.9 \pm 1.6_{A}$	$4.8 \pm 0.79_{A}$
	(0.04–0.20)	(0.17–0.50)	(0.40–1.0)	(0.13–1.5)	(1.8–12.2)	(0.62–6.1)	(4.0–6.4)
Fe	$0.04 \pm 0.01_{D}$	$0.32 \pm 0.05_{D}$	$0.19 \pm 0.04_{D}$	$0.20 \pm 0.05_{C}$	$3.0 \pm 0.69_{A}$	$4.5 \pm 1.2_{A}$	$4.8 \pm 1.2_{A}$
	(0.01–0.12)	(0.13–0.56)	(0.06–0.39)	(0.07–0.41)	(2.1–4.4)	(2.0–6.2)	(2.7–7.0)

Data are presented as the overall mean metal bioaccumulation factors in all sites ± SE (range). Means within each column followed by the same capital letter are not significantly different (Mann–Whitney U test, $p > 0.05$).

The results of this study indicate that the insectivorous *M. paykullii* can accumulate heavy metals in its body and can probably transfer them to higher trophic levels through the food web. On the whole, the bioaccumulation factors of the three trophic levels (primary producer, primary consumer, and secondary consumer) were: 0.33, 1.2, and 5.0 for Cd; 0.12, 0.58, and 4.6 for Cr; 0.59, 1.4, and 1.4 for Cu; 0.04, 0.23, and 4.1 for Fe; 0.24, 1.4, and 3.5 for Pb; 0.24, 0.74, and 2.3 for Ni; 0.35, 3.3, and 2.2 for Zn, respectively. The results showed that all heavy metals were biomagnified by the carnivorous insect when the terrestrial food chain extended to the secondary consumer. From the herbivorous grasshoppers to the predaceous mantid, average bioaccumulation factors for Cu remained constant, while values for Zn decreased. This was not surprising, as Cu and Zn are essential elements that are regulated by homeostasis, and are probably efficiently excreted by the mantid at the third trophic level.

Conclusions

In our study, different metals accumulated throughout the food chain, with differences between trophic levels. However, in general, the bioaccumulation factors of all metals increased along the food chain. Heavy metal concentrations did not increase from soil to grasses, but at the plant–primary consumer level, Cd, Cu, Pb, and Zn were biomagnified (i.e., metal concentrations were higher in the acridid grasshoppers than in the plants), and most metals were biomagnified with an increase in trophic level (i.e., secondary consumer). Thus, the present work provides some indications of the efficiency of the examined insect species in accumulating different trace elements, and provides additional evidence for metal biomagnification in terrestrial food chain, as has been previously reported (e.g., DEVKOTA & SCHMIDT, 2000; ZHENG *et al.*, 2008; ZHANG *et al.*, 2009; ZHANG *et al.*, 2014). In terms of this site-dependent accumulation of heavy metals and their biomagnification patterns, the investigated acridids can be considered bioindicators of Cd, Cu, Pb, and Zn pollution in polluted ecosystems such as Al-Tebbin region. Although the predator *M. paykullii* had higher levels for all metals than the herbivorous insects, its use as a bioindicator was not clear and needs more detailed research. Finally, we can conclude that the Al-Tebbin region has relatively high levels of potentially toxic metals in soils, plants, and arthropods. Therefore, we suggest that urgent action needs to be taken in order to control the anthropogenic impact.

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