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ABSTRACT

The recovery of soil in coastal regions that has been contaminated with heavy metals depends heavily on the exotic weeds. In the coastal region of the Nile Delta, the present study assessed the contribution of four exotic invasive weeds to phytoremediation of heavy metals. These were *Xanthium strumarium* L. (Xa), *Trianthema portulacastrum* L. (Tr), *Bassia indica* (Ba), and *Atriplex lindleyi* (At). Results showed that Fe, Zn, Co, Pb, Cd, and Cu in plant samples had average heavy metal concentrations of 0.34, 0.07, 0.009, 0.21, 0.01, and 0.042 mg/g DW, respectively. *X. strumarium* had the maximum bioaccumulation (BCF) of Cd, Cu, Co, and Zn. *A. lindleyi* and *B, indica* displayed the greatest BCF values for Fe and Pb, respectively. BCF of Pb, Co, and Fe were shown to be highly correlated with nitrogen content. Results obtained will be helpful for using these exotic weeds as a tool for phytoremediation of heavy metal-polluted soil. Therefore, the chosen exotic weeds could be considered as the one of best candidates for heavy metal phytoremediation in contaminated soils.

Keywords: Atriplex, Bassia, Bioaccumulation (BCF), Heavy metals, Invasion, Phytoremediation, Trianthema, Weeds, Xanthium.

INTRODUCTION

Plant invasion has a significant impact on biodiversity, the flow of materials and disrupting ecosystem (Rai and Singh, 2020). Daz and Cabido (2001) and Priya (2022) gave important details regarding the ecological value of the exotic weeds. On adaption of certain traits of the invasive plants in their new habitats due to prevailing environmental conditions, they most prominently disrupted the cycling of nutrients and the composition of the soil (Wang *et al.*, 2015), negatively affecting native plant communities and endangering plant species (Milanovi *et al.*, 2020). The disruption of the ecosystem's goods and services by plant invasion has significant effects on biodiversity (Rai and Singh, 2020).

With the growth of urbanization and industrialization, heavy metal pollution has become a significant issue (Noman *et al.*, 2017; Jiang *et al.*, 2021, 2022). Heavy metals are regarded as major environmental contaminants because they contaminate the air, water, and soil. Numerous anthropological factors influence the growing amount of heavy metal toxicity (Rhind, 2009). Pb contaminated soil is mostly caused by mining and smelting, automobile emissions, Pb-based paints, and industrial activity. Despite the fact that heavy metals are naturally present in soil, anthropogenic and geological activities have increased their concentration to levels that may be detrimental to plants. Exotic weeds have hyper-accumulator properties, and they can survive in highly polluted soils and exclude metals from the soil. Compared with crops, weeds often possess stress resistant properties and can maintain their growth under adverse environmental stresses like heavy metal polluted soils (Wei *et al.*, 2005).

Using a plant as a pollution absorbing agent is one strategy or technique to address the heavy metal pollution in the ecosystem. This process is known as phytoremediation. Phytoremediation is enhanced by plants with large biomass, rapid growth, wide adaptability, and have the ability to absorb one or more heavy metals (Xue *et al.*, 2022). One method for cleaning up pollution using flowering plants is phytoremediation (Yaashikaa *et al.*, 2022). With regard to cost and environmental friendliness, this technology is frequently utilized. Numerous research have shown that a number of plants have the ability to clean up the environment.

The present study aims at to assess heavy metals status, develop and establish database required to monitor the main environmental pollution using four exotic and invasive weeds as bio-indicators for heavy metals pollution in the coastal area of Nile Delta.

MATERIALS AND METHODS

Study area

Plant and soil samples were collected from Damietta Governorate at the Northern area of Nile Delta (Fig. 1). Damietta Governorate is located between latitude 31.3626°N and longitude 31.6739°E. It has a hot arid summer with temperature range from 24 to 35°C and low rainy winter with temperature range from 8 to 20°C. The total annual rain is around 167 mm and usually falls from October to March.

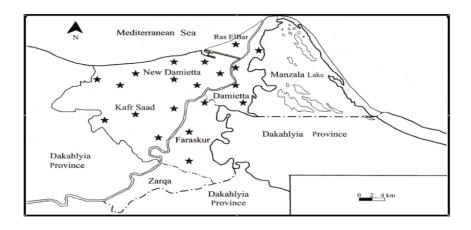


Fig. 1. Location map of Damietta showing the different sites where the four weeds and soil samples were collected.

Study species

Four exotic invasive weeds were selected for the present study namely: *Xanthium strumarium* (Xa), *Trianthema portulacastrum* (Tr), *Bassia indica* (Ba) and *Atriplex lindleyi* (At). *X. strumarium* L. is belonging to e family Asteraceae (Fig. 2a) which is one of the emerging invasive plant species currently invading croplands and non-cropped areas (Marwat *et al.* 2010). *T. portulacastrum* L. of family Aizoacae is native to South Africa. It has become noxious due to substantial yield reduction on account of competition in several cultivated crops (Fig. 2b). *B. indica* (Wight) A.J. Scott is an annual halophyte, native to India and widespread on disturbed

habitats throughout Mediterranean coast of Egypt. It exhibits a unique phenomenon of "halotropism" in which the root seems to search for high salinity in the soil to enable better conditions for enhanced growth (Fig. 2c). *A. lindleyi* is an exotic pioneer herbaceous shrublet, native to Australia, where it is widespread, especially in dry areas (Fig. 2d).

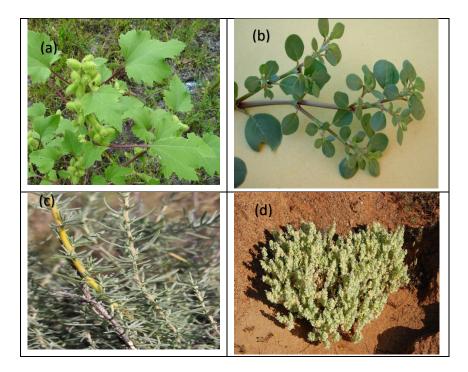


Fig. 2. Four Exotic plant species namely *Xanthium strumarium* (a), *Trinthema portulacastrum* (b), *Bassia indica* (c) and *Atriplex lindleyi* (d) from the coastal soil at Damietta Governorate.

Sample collection and analysis

Four Exotic weeds and their supporting soil were collected from three sites in Damietta Governorate. The plant samples were collected and kept in clean plastic bags. The dry weight of the collected plant samples was estimated after drying them in an oven at 85°C until constant weight (El-Liethy *et al.*, 2022). The collected soil samples were transported to the laboratory where they were air dried for 14 days at lab temperature (18–22°C). After that, the air-dried soil were sieved through a 2-mm aperture sieve and kept for the further analyses (Ibrahim and Selim, 2022).

Determination of some heavy metals

The dried samples were grind into powders, then one gram was absorbed in a 20 mL triacid mixture of HNO_3 : $HCIO_4$:HF (1:1:2 V:V:V), filtered, and diluted in 25 mL double deionized water. Six heavy metals including iron (Fe), zinc (Zn), cobalt (Co), lead (Pb), cadmium (Cd) and cupper (CU) were determined by the Atomic Absorption Spectrophotometer (Shimadzu AA-6200). The accuracy and precision of the metals measurement was estimated using certified reference material 1643e, from the National Institute Standards and Technology (NIST). Results were expressed by mg for each gram dry weight (mg/g DW).

Statistical analysis

ANOVA was achieved using SPSS 21.0 software (IBM, 2012). Forward selectionconstrained canonical correspondence analysis (CCA) was performed to determine some nutrients (Mg, Na, K, N and P) and some heavy metals (Fe, Zn, Co, Pb, Cd and Cu) characteristic that have the most significant effect on the plants and soils samples. CCA was performed using CANOCO 5.0 (Braak and Šmilauer, 2012). Pearson Correlation Coefficient and the Bioaccumulation Factor (BCF) of target nutrients and heavy metals were also calculated using the "corrplot" package in R 3.6.350.

Heavy metals in plant species

RESULTS AND DISCUSSION

Metal and nutrient transfer between soil and plants occurs naturally and is a component of the nutrient cycle (Yaashikaa *et al.*, 2022). Plants absorb metals at varying quantities, most frequently through soil solutions, and greater metal accumulation indicates higher soil metal levels. Certain heavy metals, such as Zn, Pb, Co, Fe, Cd, and Cu, are necessary in low amounts despite their potential toxicity and pollution to plants. Nevertheless, accumulation levels that are higher than the threshold limits might seriously harm plants and other biota (Ijaz *et al.* 2020).

Table (1) showed the average values of some heavy metals including Fe, Zn, Co, Pb, Cd and Cu were 0.34, 0.07, 0.009, 0.21, 0.01 and 0.042 mg/g DW, respectively. However, the maximum values of Fe (0.894 mg/g DW), Zn (0.13 mg/g DW), Co (0.017 mg/g DW), Pb (0.37 mg/g DW), Cd (0.029 mg/g DW) and Cu (0.21 mg/g DW) were detected in At11, Tr6, Tr4, At11, Tr4,5 and Xa1 plant species, respectively (Table 1). Whereas, the minimum values of Fe, Zn, Co, Pb, Cd and Cu were 0.02 mg/g DW in Tr4 plant, 0.011 mg/g DW in At10 plant, 0.001 mg/g DW in At10 plant, 0.104 mg/g DW in Ba7 plant, 0.001 mg/g DW in Ba8 plant and 0.001 mg/g DW in At11 plant, respectively. The order of the highest values of the detected heavy metals in the tested plants were Fe > Pb > Cu > Zn > Cd > Co .

Table 1. Heavy metals concentrations in the four exotic plant species, Nile delta. *Xanthium strumarium* (Xa), *Trinthema portulacastruma* (Tr), *Bassia indica* (Ba) and *Atriplex lindleyi* (At).

Heavy metals in soil samples

On the other hand, the average values of some heavy metals in the collected soil samples were 4.43, 0.055, 0.005, 0.031, 0.003 and 0.033 mg/g DW for Fe, Zn, Co, Pb, Cd and Cu, respectively (Table 2). However, the maximum values of Fe (5.30 mg/g DW), Zn (0.073 mg/g DW), Co (0.014 mg/g DW), Pb (0.058 mg/g DW), Cd (0.01 mg/g DW) and Cu (0.063 mg/g DW) were detected in soil surround the plant species Tr4, Tr5, Ba8, Xa1, At10 and Tr5, respectively (Table 2). Whereas, the minimum values of Fe, Zn, Co, Pb, Cd and Cu in soil samples were 3.59 mg/g DW in Ba9, 0.034 mg/g DW in Xa3, 0.001 mg/g DW in Tr6, 0.011 mg/g DW in Tr6, 0.001 mg/g DW in Tr4,5 and 0.007 mg/g DW in Ba9, respectively. The order of the highest values of the detected heavy metals in the tested plants were Fe > Zn > Cu > Pb > Cd > Co (Table 2).

There was normal distribution for the bioaccumulation factor (BCF) of the detected heavy metals (Fe, Zn, Co, Pb, Cd and Cu) between the tested four weeds (*X. strumarium*, *T. portulacastrum*, *B. indica* and *A.* lindleyi) (Table 3).

According to ANOVA results, it was showed that, there was highly significant positive correlation between the four weeds and bioaccumulation of Zn, Cd and Cu whereas P < 0.01. While, there was no significant correlation (P > 0.05) for bioaccumulation of Pb, Co and Fe. The

F-values of the detected heavy metals were relatively high. The F-value of Cd was the highest and significant (F value=9.10) followed by Zn and Cu with F value =8.77 and 8.32, respectively. This means that the bioaccumulation variation was mostly high (Table 3).

Species/site	Heavy metals (mg/g DW)							
	Fe	Zn	Со	Pb	Cd	Cu		
Xa1	4.672	0.04	0.008	0.058	0	0.02		
Xa2	4.525	0.041	0.002	0.055	0	0.028		
Xa3	3.803	0.034	0.002	0.033	0	0.02		
Tr1	5.303	0.067	0.008	0.029	0.001	0.053		
Tr2	4.805	0.073	0.014	0.015	0.001	0.063		
Tr3	4.561	0.053	0.001	0.011	0.005	0.033		
Ba1	4.485	0.054	0.008	0.022	0.004	0.034		
Ba2	4.777	0.059	0.006	0.014	0.004	0.03		
Ba3	3.593	0.055	0.002	0.019	0.003	0.007		
At1	4.3	0.065	0.005	0.043	0.01	0.04		
At2	4.211	0.064	0.005	0.043	0.009	0.039		
At3	4.204	0.064	0.005	0.041	0.01	0.039		
Min	3.593	0.034	0.001	0.011	0.001	0.007		
Max.	5.303	0.073	0.014	0.058	0.01	0.063		
Average	4.436583	0.05575	0.0055	0.031917	0.003917	0.033833		

 Table 2. Heavy metals concentrations in the soils supporting the growth of four exotic plant species in the Nile Delta.

Table 3. Descriptive statistics of the bioaccumulation factor (BCF) of the target heavy					
metals in the four exotic plant species in the Nile delta.					

	Ν	Range	Min	Max	Mean		Std. Dev	Variance	Skewness		Kurtosis	
Zn	12	2.84	.17	3.01	1.4603	.2785	.96498	.931	.369	.637	999	1.23
Pb	12	15.03	2.70	17.73	8.0096	1.380	4.78279	22.875	1.184	.637	.744	1.23
Co	12	11.03	.17	11.20	3.1988	1.0110	3.50429	12.280	1.569	.637	1.748	1.23
Fe	12	.21	.00	.21	.0782	.0186	.06472	.004	.726	.637	122	1.23
Cd	12	3.93	-1.49	2.44	.5615	.3422	1.18559	1.406	045	.637	714	1.23
Cu	12	2.64	-1.60	1.04	2007	.2103	.72851	.531	081	.637	.039	1.23

Bioaccumulation factor (BCF)

Figure (3) showed the minima, maxima and average of the bioaccumulation factor (BCF) of *A. lindelyi, B. indica, T. portulacastrum* and *X. strumarium* for Cd, Co, Cu, Fe, Pb and Zn. *X. strumarium* had the highest bioaccumulation for Cd followed by *T. portulacastruma*. Similarly, *X. strumarium* had the highest bioaccumulation for Co followed by *B. indica*. Moreover, it was observed that Cu was easily accumulated in *X. strumarium* species. On contrary, *A. lindelyi* has the highest accumulation rate for Fe followed by *X. strumarium* and *B. indica* (Figure 3). Furthermore, *B. indica* then *T. portulacastrum* had the highest accumulation for Pb. It was found that, *X. strumarium, T. portulacastrum* and *B. indica* had ability to bio-accumulate Zn (Figure 3).

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Figure (3) showed a positive correlation with significance (0.667^*) between accumulation of Cu in soil with the accumulated Fe in the soil. A high positive correlation with significance (0.998^*) was found between accumulation of Cu in soil support *A. lindleyi* with the accumulated Fe in the soil. Also, a strong positive correlation (0.930) between accumulations of Cu in soil surrounding of *B. indica* species with the accumulated Fe in the soil has been observed. Additionally, there was a strong negative correlation between accumulation of Cu in *T. portulacastruma* (-0.947) with the accumulated Fe in the soil. However, there was positive correlation (0.667) between the accumulated Cu in *X. strumarium* with the accumulation of Fe in its soil (Figure 4a). Regarding to the accumulation of Zn in the soil, it was observed a strong positive correlation with a significance (0.998^*) between Zn in the surrounding soil of *A. lindleyi* plant species and strong positive correlation of Zn in the surrounding soil of *X. strum* (0.958), *B. indica* (0.542) and *T. portulacastruma* (0.527) with the accumulated Fe in the soil (Fig. 3).

Figure (4a) illustrated the correlation matrix and General Linear Models (GLMs) between Fe, Cu and Zn heavy metals in the four weeds and in their supporting soils. It was observed that, there was a strong positive correlation between accumulations of Fe and Cu in the soil by *B. indica* (0.919 and 0.709) and *X. strummarium* (0.889 and 0.745) with the presence of Fe in plant species. Moreover, a strong negative correlation between accumulation of Cu in *A. lindleyi* species (-0.959) with the presence of Fe in plant species has been observed. While, there was a strong positive correlation for bioaccumulation of Cu in *T. portulactruma* (0.988) with Fe presence in the collected plant species, it was found that, there was a strong positive correlation of Zn in the soil of *B. indica* (0.830) and *X. strumarium* with the Fe presence in plant species. Moreover, there was a strong positive correlation between accumulation of Zn in *A. lindeyi* (0.747), *B. indica* (0.837), *T. portulacastruma* (0.995) and *X. strumarium* (0.955) with the presence of Fe in the four tested weeds (Figure 4).

Moreover, there was a strong negative correlation with significance between the accumulated Zn in *T. portulacastruma* (-0.998*) and without significance with *A. lindleyi* (-0.732) with accumulated of Fe in their surrounding soil. On the contrary, a strong positive correlation of Zn accumulation in *B. indica* (0.985) and *X. strumarium* (0.985) with the accumulated Fe in their soil (Figure 4a).

There was a negative correlation between accumulation of Cu in the four tested weeds; *A. lindleyi* (-0.189), *B. indica* (-0.407), *T. portulacastrum* (-0.189) and *X. strumarium* (-0.458) with the accumulated Cu in their surrounding soil (Figure 4a). There was a strong positive correlation with significance (0.742**) between the accumulated Zn in the soil with the accumulated Cu in the soil sample as well. Also, there was a strong positive correlation with highly significance between the accumulated Zn in soil of *A. lindleyi* (1.000***), *T. portulacastrum* (0.999***) and X. *strumarium* (0.610) with the accumulated Cu in soil samples. In addition to this, there was a strong positive correlation between the accumulated Zn in *B. indica* (0.979) and *X. strumarium* (0.513) with the accumulated Cu in the soil samples (Fig. 4a).

There was positive correlation between the accumulated Zn in the soil samples that surrounding *B. indica* species (0.817), *X. strumarium* (0.425) with accumulated Cu in the plant species. Moreover, there was a positive correlation (0.509) between the accumulated Zn in the plant species with the accumulated Cu in the plant species. Whereas, a positive correlation between the accumulated Zn in *T. portulacastrum* (0.967) and *X. strumarium* (0.528) with the accumulated Cu in the plant species (Fig. 4a). There was only a positive correlation between the accumulated Zn in *X. strumarium* (0.993) with the concentration of Zn the soil (Fig. 4a).

Correlation matrix and General Linear Models (GLMs) between Co, Pb and Cd heavy metals in the four collected plants species and in their supporting soils were illustrated in figure (4b). There was a positive correlation between accumulation of Pb in *A. lindleyi* (0.982), *B. indica* (0.874), *T. portulacastrum* (0.986) and X. strumarium (0.756) with the accumulated Co in the four collected weeds (Figure 4b). Moreover, there was a positive correlation between accumulation of Cd in *B. indica* (0.680), *T. portumarium* (0.910) and *X. strumarium* (0.743) with the accumulated Co in the collected plant species. Furthermore, there was a positive correlation between the accumulated Co in the soil samples that surrounded plant species; *T. portulacastrum* (0.616) with the accumulated Co in the collected plant. A positive correlation between Pb in the soil surrounded *T. portulacastrum* (0.919) with the accumulated Co in plant species. On contrary, a negative correlation have been shown between the accumulated Co in the plant species (Figure 4b).

A strong positive correlation was found between the accumulated Cd in *T. portulacastrum* (0.828) with the accumulated Pb in plant species. However, a strong negative correlation was observed between the accumulated Co in the soil of *B. indica* (-0.946) with the accumulated Pb in plant species. While, a strong positive correlation was found between the accumulated Pb in the surrounding soil of *T. portulacastrum* (0.972) and with the accumulated Pb in the collected plant species. On the other side, there was no any positive correlation between the accumulated Cd in soil samples with the accumulated Pb in the plant species (Fig. 4b).

There was a strong positive correlation between the accumulated Co in the soil surrounding *T. portulastrum* (0.887) with the accumulated Co in plant species. Also, a strong positive correlation was found between the accumulated Pb in the surrounding soil of *B. indica* (0.724) and *T. portulastrum* (0.672) with the accumulated Cd in the plant species. Moreover, a strong positive correlation with high significance was found between the accumulated Cd in the soil surrounding *A. lindeyi* (1.000**) with the accumulated Cd in the plant species (Fig. 4b). On the other hand, there was a positive correlation only between the accumulated Pb in the soil surrounding *X. strumarium* (0.592) with the accumulated Co in the collected soil samples. Also, there was a strong positive correlation between the accumulated Cd in the soil surrounding *B. indica* (0.945) with the accumulated Co in the collected soil samples.

Canonical Correspondence Analysis (CCA) was achieved to define the attribution of nutrients including Mg, Na, K, N and K in bioaccumulation of six heavy metals namely, Fe, Cu, Zn, Co, Pb and Cd in the collected exotic weeds and their soil (Figure 5). Results showed that, there was a high association between the BCF of Pb, Co and Fe and nitrogen (N) content in *A. lindleyi* (At) and *B. indica* (Ba) plant species and sodium (Na) in the soil. This indicated the high contribution plant nitrogen (P_N) and soil sodium (S_Na) in the variation of BCF of Pb, Co and Fe for plant species At and Ba (Fig. 5). On the other hand, *T. portulacastrum* (Tr) plant species showed high BCF of Cd in association with the phosphorous (P) in both soil and plant samples and also with Mg in both soil and plant species. This explanation is based on CCA1 (Axis 1) which showed high contribution (i.e. 0.55) (Fig. 5).

Heavy metals, released from both natural and anthropogenic activities contaminating water, soil and air are harmful for biological systems (Ghosh and Roy, 2019). According to many studies, they exhibit risk to the surrounding environment and human health (Huang *et al.*, 2019). Heavy metals like arsenic, cadmium, chromium, lead, mercury, and nickel get deposited to the pollutant level (Streets *et al.*, 2018).

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Due to a variety of anthropogenic activities as well as natural processes, the accumulation of heavy metals in soil has rapidly grown. Because they cannot biodegrade, heavy metals remain in the soil, where they may eventually accumulate in people due to bio-magnification. They also have the potential to go into food chains through crop plants. Heavy metal pollution poses a severe hazard to both human health and the ecology because of its poisonous nature (An Yan *et al.*, 2020). Since neither microbiological nor chemical processes can decompose heavy metals including Cd, Cu, Fe, Pb, and Zn, they tend to accumulate in sediment. The issue extends beyond soils with high metal concentrations, like those in mining regions, and affects those with moderate to low metal contamination as well (Ali *et al.*, 2004).

The effectiveness of a number of exotic weeds that have been discovered in the past two decades in absorbing and accumulating various hazardous trace elements is being examined for its potential use in phytoremediation (Liao and Chang, 2004). Through cation exchange, filtration, absorption, and chemical changes through the root, plants may be crucial in the removal of metals (Yadav and Chandra, 2011). The exotic weeds have the ability to restore ecosystem services and functions through phytoremediation, scavenging, and accumulation of excess heavy metals (Prabakaran *et al.* 2019, Naikoo *et al.*, 2020).

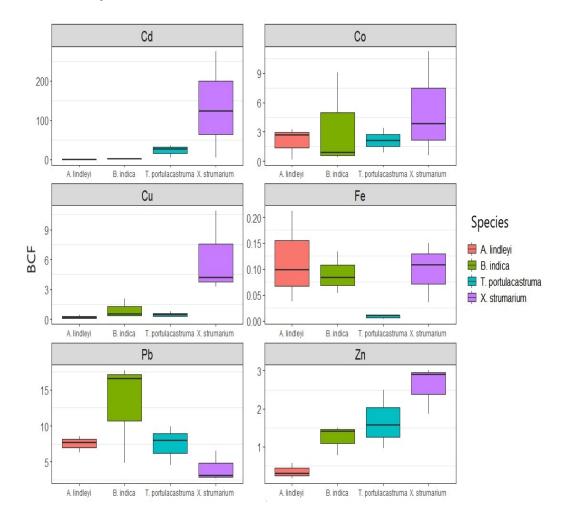


Fig. 3. Box plot of the bioaccumulation factor (BCF) for heavy metals (Cd, Co, Cu, Fe, Pb and Zn) in the four exotic plant species.

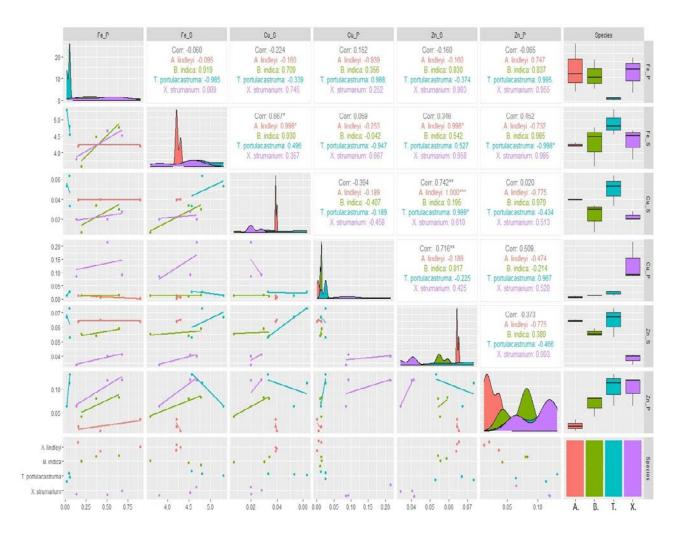


Fig. 4a. Correlation matrix and General Linear Models (GLMs) between Fe, Cu and Zn heavy metals in the four collected plants species and in their supporting soils.

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Fig. 4b. Correlation matrix and General Linear Models (GLMs) between Co, Pb and Cd heavy metals in the four collected plants species and in their supporting soils.

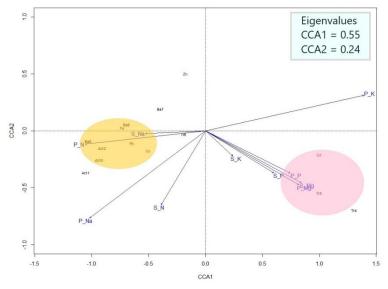


Fig. 4. Canonical correspondence analysis (CCA) shows the effect of nutrients (Mg, Na, K, N and P) presence in the collected plant species and in their surrounding soil samples on bioaccumulation (BCF) of Fe, Zn, Cu, Co, Pb and Cd as heavy metals in the four collected plant species; *X. strumarium* species (Xa1,2,3), *T. portulacastrum* species (Tr 4,5,6), *B. indica* species (Ba7, 8, 9) and *A. lindleyi* species (At10, 11, 12)

Conclusion

Phytoremediation is an eco-friendly approach that could be a successful mitigation measure to remove heavy metals from polluted soil in a low cost-effective way. To improve the efficiency of phytoremediation, a better understanding of the mechanisms underlying heavy metal accumulation and tolerance of the tested weeds is important. Further studies are needed to understand the mechanisms of heavy metal uptake, translocation, and detoxification in the nvestigated four exotic weeds. Finally, it can be concluded that *A. lindelyi*, *B. indica*, *T. portulacastrum* and *X. strumarium* plants can be used as bioindicator for accumulation of heavy metals inn polluted soil.

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التراكم الأحيائي لبعض المعادن الثقيلة بواسطة أربعة حشائش غازية في المنطقة الساحلية من دلتا النيل ، مصر

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