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Occurrence and patterns of toxic metals in Mangrove Forests from the Oman Sea, Iran

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Abstract

Concentrations of selected toxic metals were investigated in roots, stems, leaves and sediments from mangrove forests situated along the coast of the Oman Sea, Iran. Results showed that the overall average concentrations of lead, nickel, copper, and zinc in sediments were 47.90, 54.12, 42.13 and 44 µg/g dry weight (dw) and 3.81, 16.41, 29.23 and 25 µg/g dw in plant tissues, respectively. In addition, the bioconcentration factors (BCFs) of root, stem and leaf ranged from 0.5 to 1.7, 0.2 to 1.5, and 0.4 to 1.3, respectively. Calculated bioconcentration factors showed that all plant tissues were able to uptake copper from the sediments, making them suitable biological indicators for this metal. Similarly, the roots were found a suitable indicator for nickel and lead, while leaves and stems were better indicators for zinc contamination. Pollution indices showed that the sediments of mangrove forests along the coast of the Oman Sea were in the low ecological risk category (risk index < 150), and that all investigated sites were in the category of low to moderate pollution (pollution load index: 1.5-0.11), with a 21% probability of biological toxicity.

Key words: Mangrove ecosystem, Toxic metals, geochemical indicators, bioavailability, Iran

1. Introduction

Toxic metals are natural earth elements; in trace amounts, certain metals are necessary to support life but in larger amounts they may build up in biological systems and become a significant health hazard. As non-degradable and widespread contaminants, toxic metals can enter water bodies and consequently the food web, causing adverse effects on the aquatic environment and finally human health (Das et al., 2014; Shah, 2021).

Mangroves are plants or shrubs typically growing on low-sloping banks with fine-grained sediments. Mangrove trees have been shown to be able, to some extent, to accumulate certain (toxic) metals. This was evidenced in a previous study on toxic metals in the mangrove trees of Gowater bay, Chabahar, southeastern coast of Iran, where nickel was found in mangrove plant roots, likely resulting from the accumulation of this metal naturally present in ophiolite stones in the beach bed. Conversely, the presence of other toxic metals, like cadmium, copper and zinc, in other plant tissues was the result of accumulation from anthropogenic sources like upstream runoff (Einollahipeer, 2012). Due to this accumulation potential, mangrove forests play an important role in aquatic ecosystems, making them suitable to be used as biological indicators (Smical et al., 2008). To a certain extent, mangroves can tolerate an uptake of metals. This is because they possess aerial roots (i.e. pneumatophores), which allow for gas exchange in soggy soils, and facilitate the uptake of oxygen, helping to alleviate potential negative effects of metal toxicity. In addition, mangroves can sequester and store metals within certain tissues (e.g. roots) which helps protecting the vital metabolic processes and physiological functions of the trees. Mangroves also possess several detoxification mechanisms to mitigate the harmful effects of metals, e.g. by producing metal-binding ligands or through their enzymatic system (Kumari& Rathore, 2021; Alongi, 2021; Sruthi et al., 2017), and are known to benefit from symbiotic associations with microorganisms in the root systems which contribute to metal tolerance and detoxification by facilitating metal immobilization, precipitation, or transformation into less toxic forms (Harguinteguy et al., 2014; Sawidis et al., 2011; Sarwar et al., 2017; Salam et al., 2016). Finally, the mangrove physiological adaptations which allow them to maintain water and ion balance in saline environments can help mitigate the toxic effects of metals.

Studies conducted on marine ecosystems by Arumugam et al (2018) show that the concentrations of toxic metals in the sediments of mangrove forests were 3 to 5 times higher than in the surrounding water, and that the sediments of mangrove forests in tropical and subtropical regions have a high potential to store toxic metals (Shi et al., 2019). This shows the importance of such ecosystem in maintaining and possibly restoring the environmental quality of the coastline (Baharvand et al., 2022). For these reasons, monitoring the concentrations of metals in coastal and mangrove forests and evaluating their environmental quality can be considered an essential management tool to protect these ecosystems (Zhang et al., 2022).

Due to their richness in biodiversity, strategic location at the threshold of the ecological range of environmental conditions, and sensitivity to pollution, Iranian mangrove forests are highly environmentally relevant study habitats (Meena, 2018). Among them, the mangrove forest of the Oman Sea, located between latitude 25° 11' N and 27° 52' N, is considered a special ecosystem due to its a rich diversity of plant and animal life. Unfortunately, in the last few decades, it has been exposed to several anthropogenic stressors, including oil extraction and refinery and urban sewage discharge, which threaten its biodiversity (Ghayoumi et al., 2019). Because of this, this area has been the object of a few studies monitoring the metal contamination, but the quality and quantitative assessment of its ecological status has been so far poorly researched (Einollahipeer et al., 2012; Pakzadtoochaei et al., 2013).

This study aimed at investigating the presence and concentrations of trace metals, including copper, zinc, nickel, and lead, in several mangrove plant tissues and relative surface sediments collected from this Iranian area. The ability of mangroves to uptake metals from the sediments and transfer them to different plant tissues was also explored. Finally, various ecological risk assessment indicators were used to investigate the current environmental quality of the study area.

2. Materials and methods

2.1 Sampling area

Gowater protected area is located on the northern shores of the Oman Sea, in the Sistan and Baluchestan province, Iran (Zahed et al., 2010). The sampling area included five sites in the mangrove forests between the port city of Chabahar and the protected area of Gowater, which has the highest density of mangrove trees and is representative of all mangrove forests in this area (Fig. 1). This area is characterized by a dry and desertic climate, with average annual rainfall below 150 mm, an average monthly temperature of 27.5 °C (which fluctuates from a minimum of 21.2 °C in January to a maximum of 32.6°C in June), a monthly average relative humidity of 70%, and a prevalent wind direction from the southwest. The mangrove forests in the southeast of Iran extend from the Sarbaz River and continue to the coasts of the Oman Sea (Dahmardeh bBhrooz, 2022). Sampling points were chosen to represent the entire region and different habitat conditions, spanning from inland to coastal ecosystems.

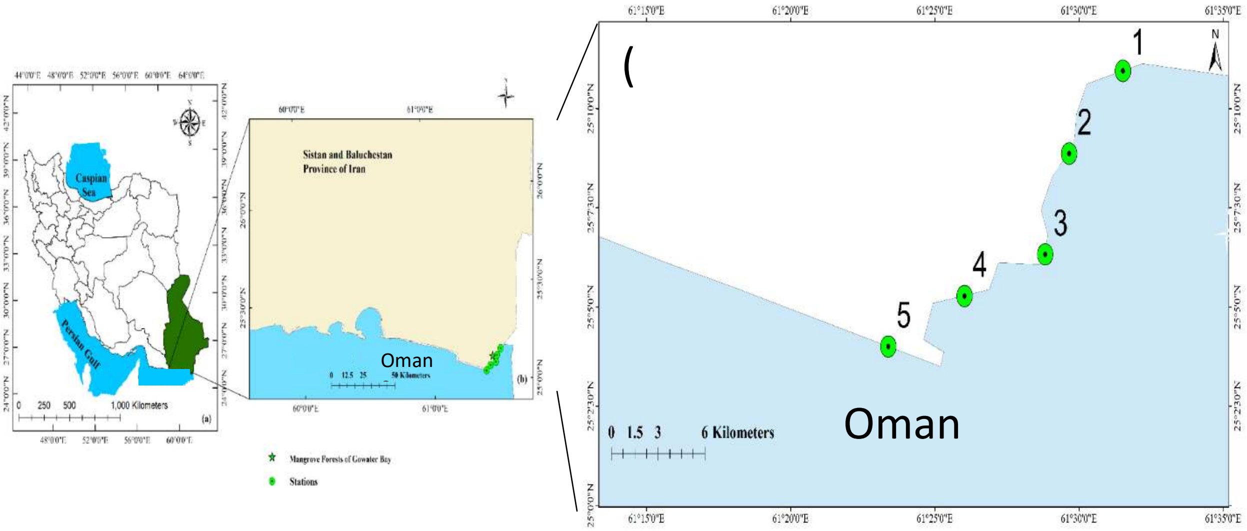

Fig 1. Location of sampling stations in the mangrove forests of Gowater Bay in the Oman Sea, Iran

2.2 Sample collection

Only "true mangroves" were considered for inclusion, defined by their adaptation to intertidal environments, salt regulation mechanisms, and taxonomic separation from terrestrial elements (Tomlinson, 1986). Samples of sediment (n=15), roots (n=15), stems (n=15) and leaves (n=300) of *Avicennia marina* were collected in triplicate from the above-mentioned five sites in spring 2022. At each sampling site, the surface sediment layer (about 500 g) was collected with a plastic shovel at 10 cm depth. For root tissue sampling according to the method of Lawton et al. (1981), sampling was done from the nutritious roots of the mangrove plant, and the harvesting of larger respiratory roots was avoided (Lawton et al., 1981). Following the methods of Lindsey et al. (2005) and MacFarlane et al. (2003), 20 leaves were collected from each plant and with three replicates at each site, carefully separating them from the petiole by horticultural scissors (Lindsey et al., 2005; MacFarlane et al., 2003). These samples were selected from 5 to 10 trees taller than 3 meters and in such a way to cover the whole tree crown. Finally, the stem samples were obtained from the transverse and thin sections of the stem tissue with a diameter of 4 millimeters by pruning shears (Davari et al., 2010).

All plant tissues were collected from trees having a healthy appearance, with no signs of disease or pest activity on the leaves. Sediment and roots were collected in the vicinity of the same marked tree. After collection, all samples were placed in zip-lock polyethylene plastic bags, transported to the laboratory, and kept at -20°C.

2.3 Sample preparation and analysis

Once at the laboratory, sediment samples were dried in an oven at 80 °C for 3 days, pulverized, sieved with a 63 µm mesh steel sieve to separate waste materials and coarser particles, transferred in pre-coded zip-lock bags and stored in the dark at room temperature until analysis (Hashim & Nazli, 2010). Plant tissues, including root, stem, and leaf samples, were carefully rinsed with distilled water, dried in an oven at 60 °C for 24 hours until the weight of the samples reached a constant value, pulverized, and finally stored in the dark at room temperature until analysis.

Sediment samples (1 g) were extracted by acid digestion through addition of a mixture of 65% nitric acid and concentrated perchloric acid (4:1, v/v), and kept at a temperature of 140 °C for 2 hours and then repeated for 3 hours. After digestion, the samples were filtered by Whatman 42 µm filter paper and finally diluted with double distilled water to a volume of 50 mL (Abdul-Wahab and Jupp, 2009). Plant tissues (1 g) were digested with 10 mL of 65% nitric acid and hydrogen peroxide (4:1, v/v) at 90 °C for 2 hours on a hot plate. After cooling at laboratory temperature, the digested samples were filtered with 0.45 µ filter paper and diluted with double distilled water to a volume of 20 mL (MacFarlane et al., 2007).

The concentrations of copper, nickel, lead and zinc in sediment and plant samples were measured using a Konic atomic absorption device model NOVAA 300 and expressed in µg/g dry weight (dw).

The sediment physicochemical properties of the study sites are presented in Table 1. The texture of the sediment samples among different sites was sandy clay loam (sites 2, 4 and 5), clay loam (site 1), and loam (site 3), with pH varying between 2.45 and 3.25 in all sites. Sediment acidity may also have resulted from decomposition of mangrove litter. All sediment samples had medium cation exchange capacity (CEC). The lowest and highest average content of organic matter in the samples was 4.76 and 11.10%, respectively. The values of organic carbon ranged from 1.07 to 6.43% with the lowest and the highest values recorded at sediments of study area.

Table 1: Physiochemical characteristics in mangrove sediments at the surface soil.

Sites	pH	OM	OC	CEC	% Soil composition			Soil Texture
		(%)		(cmol/kg)	Sand	Silt	Clay	
S1	3.15	9.20	5.38	24.15	34.08	36.50	29.57	clay loam
S2	3.25	8.12	4.35	9.19	58.05	15.96	26.05	sandy clay loam
S3	2.97	9.37	5.96	20.27	43.58	27.80	28.61	loam
S4	2.45	11.10	6.43	18.76	52.90	17.40	29.65	sandy clay loam
S5	2.97	4.76	1.07	9.69	64.95	8.35	26.82	sandy clay loam

OM**:** organic matter; OC: organic carbon

Three measures of metal uptake were used for interspecific and intraspecific comparisons: root bioconcentration factor (BCF), leaf BCF, and transfer factor (TF). However, when combining data from different studies, validity issues arise. Metal uptake may vary due to sediment conditions such as anoxic sediments with high sulfur and organic content, which can reduce metal bioavailability (Harbison, 1986). Metal availability is influenced by sediment factors like cation exchange capacity, pH, redox status, metal speciation, nutrient availability, and salinity. Limited data availability for highly contaminated areas can skew observed patterns (Greger, 2004). Varying sample sizes within and across studies can also impact interpretation by potentially including anomalous data.

2.4 Quality Control

Instrument calibration was performed with a NIST-traceable std solution (AccuTrace Single Element Standard; AccuStandard Inc., New Haven, CT, USA). The precision and accuracy of the applied analytical method were determined by means of seven replicate analyses of standard reference materials SRM 1633b (constituent elements in coal fly ash), SRM 2709 (San Joaquin soil baseline trace element concentrations), and SRM 2711 (Montana II soil). Blank samples were prepared as the samples but without matrix and average blank levels per batch were subtracted from the sample results, and a value equal to 3 times the standard deviation of the blank measurement was used as the limit of quantification (LOQ). For compounds absent in the blanks, LOQs were based on a signal/noise ratio of 10 (S/N = 10). LOQs were 0.09, 0.06, 0.05, and 0.10 µg/g dw in Cu, Ni, Pb, and Zn respectively. In each sample batch, procedural blanks and SRMs were included. The certified values for the reference materials amounted to Zn = 86 ± 2.5, Pb = 0.23 ± 0.3, Cu = 21.8 ± 5, and Ni = 1.6 ± 0.12, and the certified values for the used material amounted to Zn = 89 ± 60, Pb = 0.24 ± 0.4, Cd = 22.58 ± 3, and Ni = 1.7 ± 0.13 (6 replicates with recoveries between 88 and 105% and a relative standard deviation (RSD) of 6%).

2.5 Statistical Analysis

In this study, SPSS version 19 software was used for statistical analysis. At first, the normality of the data was checked using the Kolmogorov Smironov test, and after confirmation, one way ANOVA and Tukey statistical tests were used to compare the mean and differences between selected metal concentrations.

2.6 Indices

The following indices were used to assess the environmental quality and metal pollution status of the investigated ecosystem.

The *bioconcentration factor* (BCF) is the ratio between the concentration of a toxic metal in a living organism and in a non-living environment (water and sediment). Species with BCF > 1 can be considered as element-stabilizing species (Almahasheer, 2019).

The *metal transfer factor* (TF) is used to evaluate the ability of the mangrove plant to transfer metals from the underground tissues (roots and rhizomes) to the upper ones (stems and leaves) via the ratio between metal concentration in aerial tissues and in the roots (Hilmi et al., 2023). The higher the rate of TF, the faster the ecosystem purification process happens and the coastal and marine micro-ecosystems are less exposed to pollution (ELTurk et al., 2018).

The *geochemical accumulation index* (Igeo), introduced by (Muller, 1969), is a common method for estimating the intensity of contamination of sediments with toxic metals and is calculated based on equation 1.

$$\text{Igeo} = Log_2 \frac{Cn}{1.5 \times Bn} \tag{1}$$

Where Cn is the measured concentration of a toxic metal in the sediments and Bn is the concentration of the same element in the earth's crust (background concentration, or element concentration in shale). The index goes from class 0 (unpolluted) to class 6 (strongly polluted, where the values of the elements are at least 100 times the reference values) (Table S1).

The *contamination factor* (CF) provides a description of the pollution related to the investigated toxic elements and the pollution of the sediment environment (Hakanson, 1980). More specifically, the CF is derived using equation 2:

$$\mathrm{CF} = \frac{\mathrm{Ci}}{\mathrm{Cn}} \tag{2}$$

Where C_i is the concentration of the element in the sediments and C_n is the concentration of the same element in shale sample (Table S2), (Hakanson, 1980).

The comprehensive ecological risk assessment of selected toxic metals in sediments is determined using the *potential ecological risk index* (E^{i}_{r}) (Hakanson, 1980) and it is calculated according to equation 3.

$$E_{ir} = T_r \times C_f \tag{3}$$

Where C_f is the contamination factor and Tr is the toxicity coefficient, whose values are 1, 5, 5, and 5 for zinc, lead, copper, and nickel metals, respectively (Table S3).

Further, the *risk index* (RI) is determined as the sum of E_{ri} (equation 4) and generally indicates the sensitivity of living organisms to toxic metals and the environmental risks associated with toxic metal pollution (Kusin et al., 2018).

$$RI = \sum_{\mathrm{i=1}}^{\mathrm{n}} E_r \tag{4}$$

The sediment quality is also calculated through the *pollution load index* (PLI) based on equation 5 (Tomlinson et al., 2014).

$$\mathrm{PLI} = [\mathrm{CF1} \times CF2 \ \times CF3 \times \ldots\ldots \times CFn]\frac{1}{n} \tag{5}$$

Where n is the number of metals (=4, i.e. lead, nickel, copper and zinc), CF_n is the contamination factor of a metal (see equation 2). PLI indicates how many times the metal content in sediments exceeds the natural background concentration of metals and is a cumulative indication of the overall level of toxicity of a sample. The classification of the PLI index is as follows: PLI<1: uncontaminated; PLI≥1: contaminated.

Finally, the *mean probable effect level quotient index* (mPELq) is used to measure the biological effect of toxic metals on mangrove shrubs (Long et al., 2006) (equation 6).

$$\mathrm{mPELq} = \frac{\sum_{m}^{n} n{=}1(\frac{c_m}{PEL_m})}{n} \tag{6}$$

Where c_m is the metal concentration in the sediments, PELm values for zinc, lead, copper and nickel metals are 270, 110, 110 and 50, respectively, and n is the number of metals (Table S4).

3. Results and Discussion

3.1 Concentrations of metals in sediments

The content of trace metals in the sediment samples collected in 5 sites of the Oman Sea mangrove forests are showed in Table 2, together with a comparison of concentrations measured in this study and in other studies worldwide.

Table 2. Content of trace metals in the sediments from the Oman Sea mangrove forests ((µg/g) (mean±SD) and in other studies worldwide and in international standards

Sites	Zn	Pb	Ni	Cu	Ref
S1 (n=3)	47.45±19.45	52.25±1.95	60.20±0.95	49.10±0.95	
S2 (n=3)	30.80 ±3.90	26.30±3.70	49.15±0.95	49.90±3.60	
S3 (n=3)	58.35±1.95	57.10±1.70	59.30±3.95	44.50±8.75	
S4 (n=3)	38.60±11.20	54.50±2.95	54.20±14.60	36.90±2.35	
S5 (n=3)	47.80±7.00	54.20±2.95	58.30±0.80	37.60±0.70	
Total	44.60±12.65	48.90±3.85	56.20±24.65	43.60±6.25	**Prsesent Study**
Mangalavanam, India	139.15	27.91	41.34	33.70	Puthusseri *et al.,* 2021
Futian, South China	32.80	40.30	14.90	29.40	Wang *et al.,* 2013
Gabrik Creek (Jask), Iran	69.63	67.63	86.53	-	Zarezadeh & Rezaee, 2016
Klang, Malaysia	163.60	46.94	18.22	38.24	Yap& Al-Mutairi, 2022
Mahshahr, Iran	75.98	15.02	100.96	25.13	Cheraghi *et al.,* 2015
Sirik, Azini Creek, Iran	109.05	99.40	132.70	-	Zarezadeh *et al.,* 2014
American Sediment Quality Guidelines (NOAA)	150	46.70	20.90	34	ERL (Effect Range Low)
	410	218	51.60	270	ERM (Effect Range Medium)
Canadian Sediment Quality	120	16	31	16	LEL (Lowest Effect level)

Guidelines (SQGs)	820	75	250	110	SEL (Severe Effect level)
New York Sediment Quality Guidelines	120	32	16	16	LEL (Lowest Effect level)
	270	110	50	110	SEL (Severe Effect level)

Among all analyzed samples, sediments were the most contaminated with toxic metals, with the following pattern: nickel (56.22 µg/g dw), lead (48.86 µg/g dw), zinc (45 µg/g dw), copper (43.62 µg/g dw). The slightly higher concentration of nickel in sediments compared with the other metals was likely caused by anthropogenic sources represented by traffic of ships, boats and tankers, crude oil, and urban and industrial wastewater (Vieira et al., 2008), and by runoff from the upstream rivers of this area. In addition, the presence of nickel in sediments can be directly related to the type of bed and the prevailing morphological conditions, like the proximity of the estuary and its shallow depth (Zarezadeh et al., 2017). Similarly, lead contamination in the analyzed sediments might have derived from its presence in gasoline of ships and boats, and from runoff of inland car traffic (El Tokhi et al., 2008).

The solubility and accumulation of metals in mangrove sediments and tissues are influenced by various factors, and caution should be exercised when comparing findings to other studies. Oxygen exuded by roots fixes iron (Fe) and co-precipitates metals as oxyhydroxides in the rhizosphere, reducing trace metal availability and mobility (de Lacerda et al., 2022). Natural processes, like sea level rise, erosion, saline intrusion, tidal forcing, sediment remobilization, porewater salinization, sulfide oxidation, and metal release affect metal dynamics. (Aragon and Miguens, 2001; Lacerda et al., 1988; Nguyen et al., 2020). Metal-chloride complex formation, sulfate reduction, and changes in rainfall and environmental stress can increase metal bioavailability and toxicity (Lacerda et al., 1988). Suspended particles and particulate metals are transported by floods to the continental shelf, decreasing metal bioavailability in sediments (Nguyen et al., 2020). Acidification increases the solubility of trace metals by dissolving carbonates. These factors influence the solubility, availability, and toxic effects of metals in mangrove ecosystems.

Previous studies have shown that mud sediments can be good accumulators for both organic and inorganic pollutants due to the larger ratio between surface and volume of the particles. Because most sediments of mangrove forests are made of silt and clay, which are very small in size, their relatively high concentration of metals is justified (Zahed et al., 2010). Still, such concentrations were in the same order of magnitude as measured in India (Puthusseri et al., 2021), lower than in other Iranian locations (Zarezadeh et al., 2014; Zarezadeh & Rezaee, 2016), and higher than in China (Wang et al., 2013). In addition, to determine the degree of metal contamination of the surface sediments of the studied area, their average concentrations were compared with international sediment quality standards including the American Sediment Quality guidelines (NOAA), the Canadian Sediment Quality guidelines (SQGs), and the New York Sediment Quality guidelines, which can be used to classify polluted sediments and predict the possibility of adverse biological effects in aquatic organisms that are in contact with these sediments (Table 1). The NOAA categorizes the level of pollution in effect range low (ERL) and effect range medium (ERM), the Canadian SQGs and the New York sediment quality standard express the pollution levels as lowest effect level (LEL) and severe effect level (SEL) (Yazdan Panah et al., 2019) (Long et al., 1995). The average concentration of nickel in the sediments from the Coasts of the Oman Sea mangroves (56.22 µg/g) resulted slightly above the ERM limit for this element (51.60 µg/g) and the SEL according to the New York sediment quality standard (50 µg/g), but lower than the SEL based on the Canadian SGQ (250 µg/g). Also the average concentrations of lead (48.86 µg/g) and copper (43.62 µg/g) were higher than ERL and LEL levels but lower than SEL. Finally, the average concentration of zinc (44.60 µg/g) was lower than all standards levels.

3.2 Concentrations of metals in plant tissues

The concentrations of lead, nickel, copper, and zinc in plant tissues (root, stem and leaf) are presented in Table 3.

Table 3. Content of trace metals in tissues of stem, root and leaf of *A. marina* (µg/g) Mean±SD and in other studies worldwide.

Sites	Root				
	Zn	Pb	Ni	Cu	
S1 (n=3)	26.xx±2.44	10.31±1.16	40.22±0.97	26.37±4.39	
S2 (n=3)	28.xx±3.87	11.28±3.62	38.52±0.94	27.94±3.50	
S3 (n=3)	9.xx±4.54	9.20±0.27	39.00±1.74	24.71±1.39	
S4 (n=3)	11.xx±3.90	10.17±0.74	40.27±3.47	26.37±0.64	
S5 (n=3)	26.00±0.63	11.70±1.28	42.78±0.21	27.53±1.14	
Total	20.xx±7.35	10.53±1.84	40.15±4.30	26.58±2.59	**Prsesent Study**
	Stem				
S1 (n=3)	43.xx±7.99	1.12±0.36	7.88±0.97	28.20±0.92	
S2 (n=3)	18.xx±1.84	1.66±0.16	8.85±0.97	25.65±0.94	

S3 (n=3)	16.xx±2.21	1.34±0.16	9.91±0.89	24.50±0.85	
S4 (n=3)	23.xx±3.90	0.83±0.11	5.67±0.82	24.25±0.91	
S5 (n=3)	14.xx±2.51	1.38±0.42	3.44±1.36	25.20±1.17	
Total	23.xx±11.58	1.17±0.43	7.15±2.67	25.56±1.74	**Prsesent Study**
Leaf					
S1 (n=20)	31.xx±4.72	0.99±0.24	2.60±0.68	33.57±7.67	
S2 (n=20)	21.xx±3.97	0.97±0.12	2.69±0.77	39.93±22.91	
S3 (n=20)	24.xx±4.60	0.93±0.12	2.27±0.69	35.94±21.80	
S4 (n=20)	51.xx±9.44	0.91±0.17	1.98±0.94	60.43±6.01	
S5 (n=20)	49.xx±6.49	0.91±0.18	3.17±0.68	24.18±1.33	
Total	35.xx±13.90	0.94±0.17	2.54±0.81	38.81±17.69	**Prsesent Study**
Root					
Port Harcourt, Nigeria (*A.marina*)	6.52	3.28	1.76	4.82	Ubong *et al.*, 2018
Hawks Bay Karachi, Pakistan (*A.marina*)	23.34	-	-	5.25	Siddiqui & Saher, 2015
Carambolim, India (Macrophyte)	307.8	6.71	0.68	25.68	Vardanyan and Ingole,2004
Stem					
Guanabara Bay, SE Brazil (*L.racemosa*)	26.70	3.38	-	-	Machado *et al.,* 2002
Natal, Brazil (*R. mangle*)	1.36	-	4.43	0.31	Silva *et al.,* 2006
Hainan Island, China (*Rhizophora apiculata*)	6.20	-	-	2.90	Qiu et al., 2011
Guangdong, Province of China (*A.marina*)	6.48	**-**	2.17	1.33	Zheng et al., 1998
Leaf					
Natal, Brazil (*R. mangle*)	0.46	-	2.04	0.94	Silva *et al.,* 2006
Peninsular Malaysia (*Sonneratia caseolaris*)	5.90	35.5	-	26.80	Nazli and Hashim, 2010

Tamil Nadu, India (*A. indicum*)	107.80	23.21	-	14.78	Agoramoorthy *et al.,* 2008

The trend of accumulation of toxic metals in the plant roots was Ni (40.15 μg/g) > Cu (26.58 μg/g) > Zn (19.82 μg/g) > Pb (10.53 μg/g), different from the other plant tissues, where the trend Cu (25.56 and 38.81 μg/g), Zn (22.73 and 34.93 μg/g) > Ni (7.15 and 2.54 μg/g) > Pb (1.17 and 0.94 μg/g) could be observed for stems and leaves, respectively.

From this, it appears that essential metals have higher concentrations in aerial tissues, while non-essential metals have higher concentrations in root tissues. Also, our study results showed that concentrations of lead and nickel in the roots were significantly higher than in the other plant tissues, while this was not observed for the other metals. This difference can be due to the fact that the roots are in direct contact with the sediments and thus can directly accumulate the metals. Due to the presence of reduction conditions, frequent tide-related floodings, high levels of organic materials and sulfides, and the fine-grained texture of mangrove sediments wetlands, sediments in particular are considered sink areas for toxic metals (Alharbi et al., 2019). From here, metals can be absorbed by the roots and stored in their tissue or be absorbed and then transferred to aerial tissues. The surface absorption of elements by the root epidermis, the presence of root Casparian bands and the impenetrability of the wall of the wood vessels in the root may be among the factors influencing the elements' fate (Baharvand et al., 2022). In addition, differences in metal concentrations in root and aerial parts of the plants may be due to differences in the physiological structure of the tissues (Zheng et al. (1998)). Roots are perennial and permanent plant organs and have a longer time to accumulate metals, while leaves are subject to seasonal fall (Zheng et al., 1998) (Kabata-Pendias and Pendias in 2001). For example, the slight increase in average concentrations of zinc and copper from roots to leaves, as opposed to the higher concentrations of lead and nickel in the roots than in the stems and leaves, might be because copper and zinc are essential trace elements, necessary to the correct functioning of the plant. This is in accordance with the study by Ingole and Vardanyan (2004) who showed that the concentrations of toxic metals in the tissues of saline plants in Sevan, Armenia and Carambolim, India, were higher in plant root and stem tissues and that the lowest concentrations of metals corresponded to non-essential elements (Ingole & Vardanyan, 2004). This was confirmed by other studies on mangrove forests showing that copper and zinc had the highest concentrations in the root and leaf tissues and that they are both essential elements for plants (Shete et al., 2007) (Victorio et al., 2020) (Wozny and Krzeslowska (1993).

Eynollahipeer et al. (2012) studies looked at toxic metal accumulation in mangrove sediments and tissues in Goater Bay of Chabahar city. The general trend in metal accumulation patterns in sediments and plant tissues were somewhat similar in the study of Eynollahipeer et al. (2012) and the present study. In sediments, Ni showed the highest accumulation in both cases. In plant tissues, Cu and Zn levels tended to be higher in leaves and stems, while Ni and Pb were higher in roots. This suggests mangrove roots take up and concentrate certain metals like Ni and Pb from sediments more readily. In the study of Eynollahipeer et al. (2012), Cd accumulated more readily in plant tissues based on higher BCF values. In this study, we found higher transfer factors for Cu and Zn from roots to shoots. Both studies showed low ecological risk from metal pollution based on risk indices. However, in this research, a have higher potential toxicity based on a 21% probability was estimated (Einollahipeer, 2012).

A comparison of selected metal concentrations in the roots, stems and leaves from mangrove forests in the present study with results obtained from other similar studies worldwide is showed in Table 2. The levels of copper from this study were generally higher than in Brazil (Silva et al., 2006; Machado et al., 2002), China (Qiu et al., 2011; Zheng et al., 1998), Pakistan (Siddiqui & Saher, 2015), and Nigeria (Ubong et al., 2018), but comparable to the concentrations measured in India (Vardanyan and Ingole, 2004) and Malaysia (Nazli and Hashim, 2010). Concentrations of nickel in the roots from this study were higher than in all other selected locations, but the levels of nickel in the other tissues were comparable with results obtained worldwide. Conversely, concentrations of lead were generally similar to or lower than average levels from other locations. Finally, levels of zinc in the mangrove forests from the Oman Sea were lower than measured in India (Vardanyan and Ingole, 2004; Agoramoorthy et al., 2008), comparable with levels in Brazil (Machado et al., 2002) and Pakistan (Siddiqui & Saher, 2015) and higher than Nigeria (Ubong et al., 2018), Brazil (Silva et al., 2006) and China (Qiu et al., 2011; Zheng et al., 1998).Glasby et al. (2019) examined the impact of bushfires on estuarine wetlands, while the current study focuses on toxic metals in mangrove forests. Both studies investigate how toxic metals or bushfires affect the health, organisms, habitats, and overall ecological dynamics of their respective ecosystems. However, they differ in their specific areas of focus. The present study concentrates on mangrove forests, which are coastal wetland habitats dominated by mangrove trees, while Glasby et al.

(2019) examines estuarine wetlands, which are transitional zones between rivers and the sea. The present research emphasizes toxic metals from various sources, while Glasby et al. (2019) highlights bushfires caused by natural or human-induced factors (Glasby et al., 2023).

TM Glasby, PT Gibson, R Laird, D S Swadling, G West. 2023. Black summer bushfires caused extensive damage to estuarine wetlands in New South Wales, Australia. Ecological Management&Restoration. 24(1): 27-35.

3.3 Environmental quality assessment

Results of the calculations of the geochemical accumulation (Igeo), Contamination factor (CF), potential ecological risk factor (E^i_r), and the potential ecological risk index (RI) are reported in Table 4. The analysis of metals in sediment varies across studies due to different digestion methods and acids used. HF, which extracts metals from the silicate matrix, was not employed in any of the studies. Various extraction methods were used, including H2O2 + HCl + HNO3 + HClO4 (Silva et al., 1990), microwave digestion in HNO3 + HCl, and hotplate digestion in HNO3 + H2O2, as well as other acids such as HNO3 + HClO3, HNO3 + HSO3, (Che, 1999) and weakly bound fraction examinations with 0.1 N HCl (Chiu and Chou, 1991)or 0.1 M HCl (Lacerda, 1997). Recovery rates varied among metals when comparing different extraction methods. The choice of extraction method can impact estimates of sediment metal loadings, particularly affecting bioconcentration factors (BCFs) more than transfer factors (TFs). Caution is necessary when interpreting the data due to potential biases and variations caused by sampling protocols for vegetation collection and environmental factors like temperature, light, and inundation frequency, which influence metal uptake patterns (Greger, 2004).

Table 4. Averages of geo-accumulation (I_{geo}), Contamination factor (CF), potential ecological risk factor (E^i_r) and potential ecological risk (RI) indexes of the measured toxic metals *A. marina* sediments.

		(I_{geo}) Index		
Sites	**Zn**	**Pb**	**Ni**	**Cu**
1	2.77	2.93	3.63	2.89
2	2.01	3.52	2.85	2.96
3	3.20	2.98	3.61	3.07
4	2.86	2.60	3	2.67
5	3.08	2.91	3.4	2.88
		(CF) index		
Sites	**Zn**	**Pb**	**Ni**	**Cu**
1	1.40	2.21	2.98	1.88
2	1.23	2.80	2.78	2.02

3	2.40	2.10	2.95	2.24
4	1.79	2.40	2.20	1.60
5	2.30	2.05	2.50	1.81

(E^{i}_{r}) index

Sites	Zn	Pb	Ni	Cu	RI
1	1.28	4.18	2.89	7.27	26.33
2	1.90	3.37	2.76	6.85	30.22
3	1.79	3.83	2.10	6.64	28.35
4	1.30	4.26	2.05	7.57	31.11
5	1.55	3.43	2.91	6.89	27.55
Total	**7.82**	**19.07**	**12.71**	**35.22**	**143.56**

(PLI) and (mPELq) indexes

Sites	PLI	mPELq
1	0.45	0.87
2	0.55	0.74
3	0.37	0.63
4	0.42	0.45
5	0.40	0.42

According to the calculated geo-accumulation index, sediments from the studied mangrove forest could be classified as moderately to highly polluted. The intensity of contamination with these metals followed the order: nickel (3.2) > lead, copper and zinc (2.98, 2.89 and 2.78, respectively). A similar pattern of metal pollution was obtained based on the contamination factor (CF) for the investigated metals in the region, for which the average values were between 1 and 3, classified as low to moderate pollution status.

The results of the evaluation of the potential ecological risk factor (E^{i}_{r}) for Pb, Zn, Ni, and Cu were low ($E^{i}_{r} < 40$) and exhibited a low risk. This was confirmed by the calculated ecological risk index (143.5), representing a low ecological risk. Copper had the greatest influence on the value of the index, and zinc had the least influence. This is generally in accordance with the results of another study on toxic metal contamination in coastal sediments from the South Pars Special Economic Zone where most of the investigated sites were classified in the low to medium risk category in terms of ecological risk due to metal contamination (Haghshenas et al., 2017). Fu et al. (2014) evaluated the concentration and ecological risk of mercury, arsenic, chromium, lead, zinc and copper in the sediments of the Jialu river in China. The analysis of the E^{i}_{r} showed that, except for cadmium, classified as high-risk, other metals were in low risk status (Fu et al., 2014). Liu et al. (2014), investigated the E^{i}_{r} of chromium, copper, zinc, cadmium, arsenic, mercury and lead in mangrove ecosystem sediments in south China and found that the sediments were in a relatively severe ecological risk, especially due to the presence of mercury and cadmium (Liu et al. , 2014).

The pollution load index (PLI) of the selected metals was below 1 in all analyzed sample sites (Table 3), suggesting that the region can be classified as non-polluted. This is consistent with the results of Yu et al. (2011) and Suresh et al. (2012). Islam et al. (2015) investigated the PLI in urban river sediments in Bangladesh, and found it was > 1, indicating a reduction of sediment quality and the contamination of the studied river sediments with toxic metals. This was likely attributed to the discharge in the river of a urban sewage. This difference with the current study shows that, although human activities and industrialization are spreading in this area, their effects are still not critical on sediment quality. However, due to the growing urban population and industrialization of the region, constant monitoring of the area is highly recommended.

Finally, to determine the possible biological effects of toxic metals in sediments, the mean probable effect level quotient index (mPELq) was calculated between 0.42 and 0.87, classified as low to moderately polluted with a 21% probability of biological toxicity (Table S4). The same index was calculated by Aljahdali and Alhassan (2020) to evaluate the possible biological effects of copper, zinc, cadmium, chromium, lead, nickel and cobalt in the coastal sediments of the Kaduna River, Nigeria. Their results showed that the studied area was in the high pollution class with 94% probability of biological toxicity. The elevated mPELq was attributed to the impact of human intervention in the catchment area caused by industrial activities and atmospheric depositions (Aljahdali, 2020 & Alhassan). Also Rastegari Mehr et al. (2020) investigated the mPELq index to evaluate the possible biological effects of mercury, nickel, zinc, copper, lead and chromium on the coastal sediments of the Musa estuary and found the mPELq index between 0.5-1.51, with a probability of biological toxicity of 49% (Rastegari Mehr et al., 2020).

Finally, the BCF and TF were calculated based on the concentrations of metals in the sediments and in the plant tissues (Table 5).

Table 5. Bioconcentration Factor (BCF) and Transfer Factor (TF) of toxic metal in *A. marina*.

Tissue	Zn	Pb	Ni	Cu
BCF in Root	0.88	0.75	0.77	1.05
BCF in Stem	0.95	0.52	0.43	1.03
BCF in Leaf	0.93	0.68	0.65	1.02
TF in Stem	1.15	0.40	0.47	1.04
TF in Leaf	1.50	0.58	0.45	1.20

To be biological indicators, tissue accumulation of metals should be sediment-dose dependent. Otherwise, BCFs and TFs have very limited utility concluding biological indicator potential.. For zinc, leaf and stem tissues are the best biological indicators, with BCF values higher than in roots. The TF results for lead and nickel were < 1, indicating that the state of accumulation and accessibility in the plant is average. For zinc and copper, the calculated TF was > 1, indicating that, after their absorption from the environment by the roots, the metals were transported to the aerial parts of the plant. MacFarlane et al. (2007) calculated the BCF for copper, lead and zinc in the same mangrove plant species (*A. marina*), obtaining values > 1, and concluded that the root tissue is a suitable bioindicator for these metals. In the same study, the TF was determined for copper and zinc as 1.52 and 1.53. They considered the reduction of the metal transfer factor from the root to the plant as a result of the type of metal consumption for the plant (MacFarlane et al., 2007).

Conclusion

This study found that the concentrations of nickel in sediments was slightly higher compared to other metals, and this was likely due to anthropogenic sources such as ship traffic, crude oil, urban and industrial wastewater, and runoff from upstream rivers. Lead contamination in sediments was likely derived from ships, boats, inland car traffic, and gasoline. The accumulation of toxic metals in plant tissues varied, with nickel having the highest concentration in roots, followed by copper, zinc, and lead. Essential metals tended to have higher concentrations in aerial tissues, while non-essential metals had higher concentrations in root tissues. The roots, being in direct contact with sediments, accumulated higher levels of lead and nickel compared to other plant tissues. Mangrove sediments were considered sink areas for toxic metals due to reduction conditions, frequent floodings, high organic material and sulfide levels, and fine-grained texture. The concentrations of nickel in the roots were higher in the studied area compared to other locations, but levels in other tissues were comparable worldwide. The sediments in the studied mangrove forest were classified as moderately to highly polluted based on the geo-accumulation index. The intensity of contamination followed the order: nickel > lead, copper, and zinc. The contamination factor indicated low to moderate pollution status. The potential ecological risk factor and ecological risk index suggested low ecological risk, with copper having the greatest influence on the index value and zinc having the least. Other studies on metal contamination in coastal sediments have also indicated low to medium ecological risk. The pollution load index indicated that the region can be classified as non-polluted. The mean probable effect level quotient index indicated low to moderate pollution with a 21% probability of biological toxicity. Plant tissues, especially roots, were identified as good biological indicators for lead, nickel, and copper accumulation. Zinc accumulation was higher in leaf and stem tissues. The transportation factor indicated that lead and nickel had average accumulation and accessibility in plants, while zinc and copper had higher accumulation and transportation to aerial parts after absorption by the roots. Overall, the study highlights the presence of toxic metals, their sources, and their accumulation patterns in sediments and plant tissues in the studied mangrove forest. The findings provide valuable information for assessing the ecological risk and potential biological effects of these metals in the ecosystem.

CRediT authorship contribution statement Sanaz Khammar: Conceptualization, Software, Visualization, Investigation. Fatemeh Rajaei: Data curation. Reza Dahmardeh Behrooz: Methodology, Writing- Original draft preparation. Giulia Poma: Writing- Reviewing and Editing. All authors have read and agreed to the published version of the manuscript.

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Ethics approval and consent to participate All procedures performed in this study were in accordance with the ethical standards of the institutional and/or national research committee University of Zabol with reference number 005.1401.REC.UOZ.IR.

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Data availability Data will be made available on request.

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