Original Research

# Trace Elements Accumulation and Maternal Transfer in Critically Endangered Sea Turtle, *Eretmochelys Imbricata*

Farideh Ahmadi<sup>1</sup>, Abdolrahim Pazira<sup>1\*\*</sup>, Tayebe Tabatabaei<sup>1</sup>, Majid Askari-Hesni<sup>1, 2\*</sup>

<sup>1</sup>Department of Environmental Engineering, Bushehr Branch, Islamic Azad University, Bushehr, Iran <sup>2</sup>Department of Biology, Faculty of Science, Shahid Bahonar University of Kerman, Kerman, Iran

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## Abstract

In this study, concentrations of Fe, Zn, As, Cd, Hg, Pb, and Cu in blood and muscle samples and maternal transfer of these elements to eggs and hatchlings were investigated for hawksbill turtles on Ommolgorm, Nakhiloo, and Kharko Islands from the Persian Gulf. The results indicate a significant difference ( $p \le 0.05$ ) between the accumulations of the elements between the rookeries. On three islands, the highest and lowest concentrations of Fe were observed in the yolk and albumin, respectively. The highest concentrations of Zn were found in yolk and maternal muscle from Ommolgorm and Nakhiloo islands, and the lowest concentration of Zn was found in hatchling and abnormal albumin at Kharko Island. The highest amount of As was present in maternal blood, and the lowest was in an abnormal egg on three islands. The highest amount of Hg was in maternal blood and muscle, and the lowest was in normal and abnormal eggs. The highest Pb concentration was in yolk and the lowest was in maternal muscle and blood in Ommolgorm and Nakhiloo Island; however, at Kharko Island, the highest and lowest concentrations were in yolk and normal albumin, respectively. Cd and Cu were ND in the most tissues. Fe, As, Hg, and Pb were higher than Cd and Cu in the maternal blood. The accumulation of heavy metals in the yolk, blood, and hatchling was higher than in other tissues. The accumulation of some elements in samples from Kharko Island was higher than on the other two islands. Based on the results, a significant relationship was found among the concentrations of elements in the maternal blood, yolk, and hatchling tissue. Our results showed a positive correlation between the concentrations in the maternal blood and yolk and hatchling tissue, which confirms maternal transfer of the elements.

Keywords: pollution, heavy metals, maternal transfer, sea turtles

<sup>\*</sup>e-mail: mahesni@gmail.com, mahesni@uk.ac.ir

<sup>\*\*</sup> e-mail: abpazira@gmail.com

### Introduction

The Persian Gulf is a shallow and semi-closed ecosystem that regularly receives large volumes of pollutants annually, including oil and heavy metals. It is known as one of the world's largest oil and gas terminals [1-3], and along with several other anthropogenic activities (e.g., marine transportation, contractions, coastal development, and industries), the likelihood of heavy metal pollution has increased [4-6]. In addition, hot weather conditions, high evaporation, high salinity, shallow depth, and limited water circulation have caused a high rate of pollutant accumulation in sediments and the water column. The environmental issues of the Persian Gulf are having negative effects on biodiversity, including marine turtles, and influencing the survival of this ecosystem [7, 8].

There are seven species of sea turtles living in tropical and subtropical waters around the world, and five of them, including Eretmochelys imbricata, Chelonia mydas, Caretta caretta, Lepidochelys olivacea (family Cheloniidae), and Dermochelys coriacea (family Dermochelyidae), have been observed in the Persian Gulf. Among these five species, nesting by E. imbricata, C. mydas, and L.olivacea has been reported from southern Iranian islands in the Persian Gulf [9-12]. The majority of nesting sites related to E. imbricata in Iranian waters in the northern part of the Persian Gulf were reported on Kish, Nakhiloo, Ommolgorm, Shidvar, Qeshm, Hengam, Hormoz, and Hendourabi Islands, and Naiband Marine-Coastal National Park [8-12, 14].

In recent years, the E. imbricata population in the world has declined, and since 2010, this species has been listed as an endangered species on the IUCN Red List of the World Organization for Conservation of Nature [13]. Several factors are contributing to the decline in marine turtle populations. Destruction of habitat by coastal development, fishing activities, waste disposal (city, urban, agricultural, and industrial), oil extraction and production activities, chemical pollutants, tourism activities, travel, and collisions of boats and ships are among the most important factors in reducing their populations [12, 14-16]. Heavy metal pollution occurs because of many of these activities, and the accumulation of metals in aquatic ecosystems creates a serious environmental issue. Heavy metal pollution can have devastating effects on the ecological balance of the environment and aquatic diversity [6]. These pollutants are among the most dangerous and threatening groups of pollutants because of their toxicity, stability, transferability, and bioaccumulation in organisms [15, 17]. The bioaccumulation of heavy metals in subsequent generations may have acute effects on the health and biological survival of organisms [1, 6]. Because the different species of sea turtles occupy different ecological niches in the marine ecosystem, they may accumulate heavy metals and other toxic chemicals from their food. For example, green turtles forage

on seagrasses and hawksbill turtles forage on sponges, which may accumulate pollutants from the sea bed or water column [16].

Physiological studies indicate internal changes in individuals or populations versus environmental changes can affect biomass, activities, nesting behaviors, reproduction, and ultimately survival [15, 18, 19]. Heavy metals are one of the factors affecting the health and physiology of sea turtles because these pollutants can accumulate and transfer throughout the food chain. In addition to being toxic, the possibility of transferring them from the parent to the offspring is very high. But few comprehensive studies have been done on the accumulation of heavy metals in sea turtles, especially E. imbricata in the Persian Gulf. Several studies on the accumulation of heavy metals in the liver, heart, fat, kidney, gonad, and brain tissues of C. caretta on the Sicilian coast of the Mediterranean Sea [18], kidney, liver, and muscle tissues of C. mydas and C. caretta on Japanese shores [19], muscle tissues of E. imbricata and C. mydas on the northern shores of Bahia in Brazil [20], eggs of Chelonia mydas and Lepidochelys olivacea in the Pacific Ocean [21], and adipose and muscle tissues of C. caretta on the coast of Spain [22]. Because no comprehensive study has been conducted on the transfer of heavy metals from breeders to eggs and embryos in the Persian Gulf, the present study was conducted to investigate the accumulation of heavy metals in eggs and hatchlings of Eretmochelys imbricata.

## **Materials and Methods**

#### Sampling Location

The sampling operation of the two islands of Ommolgorm and Nakhiloo, located in Dayer-Nakhiloo National Park and Kharko (Khargo) Island Fig. 1, took place during April 2019. Nesting sites were monitored at night for sampling. 18 nesting turtles were sampled on each island after they had completed nesting. The carapace curve length (CCL) and the carapace curve width (CCW) were measured. After nesting, a piece of the muscle from the right flipper was removed using a muscle puncture and immediately placed on ice. After transfer to the laboratory, it was kept at -24°C until heavy metals were measured. Blood samples were taken from the dorsal sinus of the neck using a sterile syringe and a needle. Before extracting the blood, the neck was rinsed with deionized water and ethanol. About 5 to 10 ml of blood was taken from each turtle and immediately placed in a heparinized polyethylene tube. After egg lying, 5 normal eggs and 5 abnormal eggs (yolkless eggs) were randomly removed from each nest before contact with the sand, and then the weight and diameter of each egg were measured by using calipers with 0.1mm accuracy and a digital laboratory scale of 0.1gr. The nests that were sampled were marked, fenced, and numbered. At the time of emergence for

	Ommolgorn	n	Nakhiloo		Kharko	
Element	X <sup>2</sup> (chi-square)	p-value	X <sup>2</sup> (chi-square)	p-value	X <sup>2</sup> (chi-square)	p-value
Fe	41.083333	0.001	44.286417	0.001	42.477891	0.001
Cu	а	a	20.134577	0.005	а	a
Zn	44.748624	0.001	41.734645	0.001	45.453147	0.001
As	44.998871	0.001	41.279877	0.001	43.315728	0.001
Cd	37.949126	0.001	46.032413	0.001	45.785370	0.001
Hg	38.943829	0.001	34.018221	0.001	41.403351	0.001
Pb	42.577609	0.001	31.318353	0.001	21.643213	0.03

Table 1. Heavy metal concentrations in adult, egg, and hatchling hawksbill turtles in Ommolgorm, Nakhiloo, and Kharko Islands according to the Kruskal-Wallis Nonparametric test. The same Cu concentration in all samples (less than 001 ppm), the test did not show any significant difference between the groups.

the nest, 5 dead embryos were selected from each nest, and analyzed for heavy metals.

To analyze the heavy metals in the eggs, whole eggs are washed with deionized water to remove any foreign matter. The shell, yolk, and albumin were then separated and stored at -24°C. To measure the heavy metals in the blood, 3 ml of blood was used. To measure the heavy metals in the tissues, 0.2 gr of tissue samples were concentrated in glass containers in 5 ml of concentrated nitric acid and digested on the heater. The digested samples were diluted with 50 ml of deionized water and stored in polyethylene containers for further digestion. The prepared samples were then measured by ICP-MASS. To measure the amount of heavy metals in the studied tissues, 0.2 gr of each tissue was inserted into a teflon tube, and then gently 1 ml of hydrochloric acid and nitric acid solution (with a volume ratio of  $HNO_3$ : HCL, 1:3 V/V) was added, and the sample was placed at laboratory temperature for one hour.

The tubes were then closed and placed in an aluminum cover on a hot plate at 120°C for 2 hours and 30 minutes. Then, 2.7 g of boric acid was weighed in 50 ml polypropylene tubes or a volumetric flask, and then about 20 ml of deionized water was added. The sample is cooled at room temperature in the laboratory, and then the tubes are opened and the sample is shaken to complete the dissolution of boric acid. The solutions were then diluted to 50 ml with deionized water, and finally, the solution was transferred to plastic tubes, and the amount of metals was measured with an ICP-MASS device.

Data analysis was performed by SPSS software version 24. The normality of the data was assessed by the one-sample Kolmogorov-Smirnov test. The T-test was used to analyze the mean of the data and a significant difference between the data at the 95 confidence level.

## **Results and Discussion**

Based on the monitoring, 76, 84, and 19 nesting turtles were observed on the Ommolgorm, Nakhiloo, and Kharko islands, respectively. The means of CCL were 71.4±3.67, 70.8±4.21, and 70.1±2.7 cm, respectively. The means of CCW were 65.5±8.19, 64.8±9.21, and 64.4±4.79 cm, respectively. The average number of normal eggs on each island was  $74.5\pm3.93$ ,  $81.4\pm3.12$ , and 92.5±3.19, respectively. Also, the number of abnormal eggs was 31.3±8.93, 34.1±6.73, and 22.7±3.19, respectively. The mean CCL and CCW hawksbill turtles from Kish Island in the Persian Gulf reported 71.6 and 65.1 cm, and total, normal and abnormal egg numbers were reported at 92.9, 75.2, and 17.7 eggs in this island (10). Razaghian et al. (2019) calculated the mean CCL and CCW of E. imbricata at 70.1±2.7 and 64.77±3.2 cm, respectively, in the Persian Gulf (11).

## Elements Concentration in Different Tissues in Three Islands

The amount of elements in the tissue of nesting turtles (blood and flipper tissue), eggs, and hatchlings in Ommolgorm, Nakhiloo, and Kharko indicates a significant difference ( $p \le 0.05$ ) between the studied tissues of each population in terms of the accumulation of elements (Table 1).

The results showed that the concentrations of iron differed significantly among the studied tissues of the Ommolgorm Island population (X2 = 41.08, p<0.001) (Table 1). This element was significantly different in yolk samples compared with other tissues such as normal egg albumin, abnormal eggshell, and others, and also with hatchling muscle tissue (p $\leq$ 0.05). The highest and lowest concentrations of Fe were present in the egg's normal yolk and albumin, respectively (Table 2). But the concentrations of this element was significantly different (p $\leq$ 0.05) among the samples from Nakhiloo Island (Table 3). The highest amount of iron was

				Sample	;			
Element	Yolk	N. E. Albumin	Ab. E. Albumin	Normal shell	Abnormal shell	Blood	Muscle	Hatchling
Fe	75.58±1.64	9.66±0.78	8.41±0.59	24.16±4.82	21.71±3.27	30.38±2.49	30.18±3.8	45.61±5.85
Cu	ND	ND	ND	ND	ND	ND	ND	ND
Zn	26.8±0.29	0.03±0.02	0.04±0.01	0.01±0.01	ND	17.6±1.46	2.48±0.56	20.76±1.33
As	1.93±0.16	$0.02{\pm}0.01$	ND	ND	ND	9.38±3.2	1.12±0.81	3.04±0.42
Cd	ND	ND	ND	0.03±0.001	0.01±0.001	ND	ND	ND
Hg	2.31±0.24	3.4±0.64	2.01±0.38	0.36±0.09	1.45±0.21	9.67±1.86	7.84±0.67	1.43±0.32
Pb	0.87±0.16	0.01±0.001	0.01±0.001	0.15±0.05	0.52±0.05	0.78±0.21	0.41±0.09	0.75±0.18

Table 2. Heavy metal concentrations (Mean±SD) in adult, egg, and hatchling for the hawksbill turtle population in Ommolgorm Island.

Table 3. Heavy metal concentration (Mean±SD) in adult, egg, and hatchling for the hawksbill turtle population in Nakhiloo Island.

				Samp	le			
Element	Yolk	N. E. Albumin	Ab. E. Albumin	Normal shell	Abnormal shell	Blood	Muscle	Hatchling
Fe	72.78±1.72	7.33±3.1	8.13±0.34	19.76±8.14	22.92±8.89	30.51±1.45	42.35±6.86	61.98±8.21
Cu	ND	ND	0.01±0.003	1.36±2.16	ND	ND	ND	ND
Zn	39.02±6.02	0.03±0.01	$0.04{\pm}0.001$	0.5±0.34	0.3±0.01	21.64±0.55	$1.91{\pm}0.78$	22.87±4.75
As	1.61±0.35	0.01±0.01	0.01±0.001	$0.03 \pm 0.02$	$0.01 \pm 0.01$	23.29±4.79	3.49±0.3	2.36±0.67
Cd	ND	ND	ND	$0.06 {\pm} 0.05$	0.02±0.01	ND	ND	ND
Hg	5.76±1.56	0.96±0.81	0.79±0.8	0.92±1.8	$0.11 \pm 0.09$	4.52±0.32	4.37±0.52	1.86±0.33
Pb	0.34±0.06	0.01±0.001	0.01±0.002	0.44±0.35	0.25±0.03	0.42±0.03	0.12±1.02	0.33±0.14

Table 4. Heavy metal concentration (Mean±SD) in adult, egg, and hatchling for the hawksbill turtle population in Kharko Island.

				Sampl	e			
Element	Yolk	N. E. Albumin	Ab. E. Albumin	Normal Shell	Abnormal Shell	Blood	Muscle	Hatchling
Fe	89±2.66	15.73±0.65	7.18±1.73	29.86±20.08	27.52±16.92	28.26±2.47	31.38±4.34	126.14±16.75
Cu	ND	ND	ND	ND	ND	ND	ND	ND
Zn	27.11±0.48	0.07±002	0.04±0.011	0.28±0.09	0.41±0.001	16.52±1.62	5.76±0.26	29.1±7.40
As	1.5±0.23	$0.01 \pm 0.001$	ND	$0.02{\pm}0.001$	ND	12.25±3.07	1.22±0.09	2.23±1.24
Cd	ND	ND	ND	0.03±0.02	0.04±0.01	ND	ND	ND
Hg	3.95±1.18	ND	ND	ND	ND	2.99±0.22	6.81±3.75	2.32±1.49
Pb	0.71±0.44	0.01±0.001	0.01±0.001	0.37±0.25	0.77±0.53	0.33±0.37	0.55±0.12	0.33±0.36

present in the yolk, and the lowest in normal albumin (Table 3).

The concentration of this element on Kharko Island was significantly different between all samples and tissues ( $p \le 0.05$ ). The highest amount of iron was in the yolk, and the lowest amount was in the abnormal albumin (Table 4).

The concentration of zinc among samples from Ommolgorm Island between egg, adult muscle, and hatchling was significantly different ( $p \le 0.05$ ) (Table 2).

The concentration of this element in Nakhiloo Island between the tissues of adult and yolk, normal egg yolk and albumin, normal albumin and adult blood, hatchling and adult muscle, adult muscle and normal albumin, adult muscle and blood, hatchling and albumin, normal and yolk, and abnormal albumin showed a significant difference ( $p \le 0.05$ ). On this island, the highest zinc concentration was measured in the yolk and the lowest in hatchling muscle (Table 3). The concentration of this element in Kharko Island among the samples of yolk and abnormal albumin, abnormal albumin and hatchling, abnormal albumin and adult blood, normal albumin and yolk, normal albumin and hatchling, hatchling and normal eggshell were significantly different ( $p \le 0.05$ ). The highest and lowest amounts of this element were observed in hatchling and abnormal albumin, respectively (Table 4).

The arsenic concentration in different samples from Ommolgorm Island showed a significant difference ( $p \le 0.05$ ). The highest arsenic accumulation was observed in the blood, and the lowest amount was in the abnormal eggshell (Table 2). The concentration of this element was significantly different between samples on Nakhiloo Island ( $p \le 0.05$ ). The highest and lowest arsenic concentrations were observed in adult blood and abnormal eggshells, respectively (Table 3). The concentration of this element on Kharko Island were significantly different between different samples ( $p \le 0.05$ ). The highest and lowest arsenic concentrations were presented in adult blood and abnormal albumin, respectively (Table 4).

There was no significant difference between normal eggshell and abnormal eggshell samples from Ommolgorm, Nakhiloo, and Kharko ( $p \ge 0.05$ ). In other tissues, the Cd concentration was not detectable (Tables 2-4).

The concentration of mercury element between adult blood tissue with normal eggshell, muscle, hatchling, and abnormal eggshell, adult muscle with normal eggshell, abnormal eggshell and hatchling showed a significant difference on Ommolgorm Island ( $p \le 0.05$ ). The highest Hg concentration was observed in adult blood and muscle. The lowest Hg concentration was in normal eggshells (Table 2). However, the Hg concentration on Nakhiloo Island was significantly different between an abnormal eggshell with adult muscle, yolk and adult blood, adult blood and tissue, yolk and normal eggshell ( $p \le 0.05$ ). The highest amount of mercury was in adult blood and muscle, and the lowest was in the normal eggshell (Table 3).

The Hg accumulation in different tissues in Kharko Island showed a significant difference between tissues ( $p\leq0.05$ ). The highest amount of mercury occurred in adult blood and muscle, and the lowest was observed in normal and abnormal albumin and normal eggshells (Table 4). The concentration of this element on Kharko Island was significantly different between different tissues ( $p\leq0.05$ ). The highest Hg accumulation was in blood and adult muscle, and the lowest was in normal albumin, abnormal albumin, and normal eggshell (Table 4). The Hg concentration in Ommolgorm Island showed a significant difference between adult blood with abnormal eggshells, muscle, yolk and hatchling, hatchling and adult muscle, normal albumin and adult muscle, yolk and adult muscle, adult muscle and abnormal eggshells ( $p \le 0.05$ ).

The highest concentration of lead was in yolk, and the lowest was in adult muscle and blood (Table 4). The Pb concentration in Nakhiloo Island was significantly different between adult blood with yolk and hatchling, and adult muscle with normal albumin and yolk ( $p \le 0.05$ ). The highest concentration of lead was in yolk, and the lowest was in adult blood (Table 3). The concentration of lead in Kharko Island was significantly different between yolk and normal albumin, normal albumin, and abnormal eggshell ( $p \le 0.05$ ). The highest and lowest Pb accumulation were in yolk and normal albumin, respectively (Table 4).

The concentration of copper in the islands of Ommolgorm and Kharko in all tissues was not detectable and was measured only in Nakhiloo Island in the normal eggshell and abnormal egg albumin ( $p\leq 0.05$ ). In other tissues (similar to the other two islands), it was not detectable (Tables 2-4).

Heavy metals are one of the most important pollutants in the Persian Gulf, which enter the ecosystem through industrial activities, oil extraction and transportation, and other polluting activities [1, 23, 24]. In recent years, various industrial activities such as water desalination, fishing activities, coastal destruction, and the entry of pollutants have had many negative effects on the nutritional habitat, reproduction, nesting habitats, biological survival, and eggs and hatchlings of sea turtles in the Persian Gulf and other parts of the world [10, 25]. Different pollutants have many negative effects on biodiversity and aquatic survival, such as marine turtles [15-22, 25]. One of the unique and critically endangered species in the Persian Gulf is the Hawksbill turtle, Eretmochelys imbricata [9-14]. In the present study, the presence of heavy metals such as iron, zinc, arsenic, mercury, cadmium, lead, and copper obtained from nesting turtles, their eggs, and hatchlings from three islands (Ommolgorm, Nakhiloo, and Kharko islands) in the northern Persian Gulf was investigated. The results showed that the levels of iron and zinc at all three islands and in all tissues sampled were higher than other metals. The lowest metal bioaccumulation in tissues was copper. Various factors play a role in the accumulation of elements in the bodies of aquatic animals, such as temperature, environmental pollutant concentration, and time of contact with pollutants [17-26].

The amount of different metals accumulated in tissues depends on their physiological role. Because of its physiological role in the transfer of various substances from the gastrointestinal tract to other parts of the body, blood plays an effective role in carrying pollutants to other tissues. Blood is considered an important tissue for measuring pollutants [15, 18, 25]. In the present study, the accumulation of iron, zinc, arsenic, mercury, and lead metals in the blood was high, but the levels of cadmium and copper were low. The kidney tissue is the target tissue for the accumulation



Fig. 1. Sampling sites, Ommolgorm, Nakhiloo and Kharko Islands in The Persian Gulf.



Fig. 2. Iron concentrations in different tissues of hawksbill turtle adults and hatchlings from the Ommolgorm, Nakhiloo and Kharko Islands in the Persian Gulf.



Fig. 3. Zinc concentrations in different tissues of hawksbill turtle adults and hatchlings from the Ommolgorm, Nakhiloo and Kharko Islands in the Persian Gulf.

of cadmium. According to a study by Yipel et al. (2017), the amount of cadmium in the blood of *Caretta caretta* and *Chelonia mydas* was less than measurable in blood.

The amount of copper in the blood of both species was much lower than in the kidney [15]. Some heavy metals, such as mercury, lead, and cadmium are highly toxic to living organisms (including sea turtles). Some heavy metals, such as copper, nickel, iron, zinc, manganese, and aluminum are essential for the body, but high concentrations are also harmful [28, 29].

According to these study observations, some nests have been evacuated by local communities, and the eggs have been used as food, which has negative effects both in terms of conservation of the species and as a source of heavy metal pollutants. However, the amount





Fig. 4. Arsenic concentration in different tissues of hawksbill turtle adults and hatchlings from the Ommolgorm, Nakhiloo and Kharko Islands in the Persian Gulf.



Fig. 5. Mercury concentration in different tissues of hawksbill turtle adults and hatchlings from the Ommolgorm, Nakhiloo and Kharko Islands in the Persian Gulf.



Fig. 6. Lead concentration in different tissues of hawksbill turtle adults and hatchlings from the Ommolgorm, Nakhiloo and Kharko Islands in the Persian Gulf.

of heavy metals measured in hawksbill turtle eggs at all three islands was lower than WHO standards.

According to the results, a significant relationship was observed between the concentration of elements in the adult blood, yolk, and hatchling. As the amount of metals in the blood of the adult increases, the amount in the egg yolk and hatchlings increases. Because there is not sufficient time for transfer from the environment during incubation, this relationship indicates the transfer of heavy elements from the adult to the hatchling through the blood to the yolk. Because the amount of elements in the albumin of abnormal and normal eggs are not significantly different and are low; the main source for the transfer of metals from the adult to hatchling is the yolk. The rate of accumulation of elements in the adult's muscle was in many cases lower than the hatchling tissue, which indicates the transfer of metals from the aquatic environment to the circulatory

Location and Species	Ref	Element				San	aple			
			Yolk	N. E. Albumin	Ab. E. Albumin	Normal shell	Abnormal shell	Blood	Muscle	Hatchling
		Fe	75.58±1.64	9.66±0.78	8.41±0.59	24.16±4.82	21.71±3.27	30.38±2.49	30.18±3.8	45.61±5.85
		Cu	QN	ND	ND	ND	ND	ŊŊ	ŊŊ	ND
Ommolgorm		Zn	26.8±0.29	$0.03 \pm 0.02$	$0.04{\pm}0.01$	$0.01 \pm 0.01$	ND	17.6±1.46	2.48±0.56	20.76±1.33
E.imbricata		As	$1.93 \pm 0.16$	$0.02 \pm 0.01$	ND	ND	ND	9.38±3.2	$1.12 \pm 0.81$	$3.04{\pm}0.42$
		Cd	QN	ND	ND	$0.03 \pm 0.001$	$0.01 \pm 0.001$	QN	ŊŊ	ND
		Hg	2.31±0.24	$3.4{\pm}0.64$	2.01±0.38	$0.36 \pm 0.09$	1.45±0.21	9.67±1.86	7.84±0.67	$1.43\pm0.32$
		Pb	$0.87 \pm 0.16$	$0.01 {\pm} 0.001$	$0.01{\pm}0.001$	$0.15 \pm 0.05$	$0.52 \pm 0.05$	QN	QN	$0.75 \pm 0.18$
		Fe	72.78±1.72	7.33±3.1	8.13±0.34	19.76±8.14	22.92±8.89	30.51±1.45	42.35±6.86	61.98±8.21
		Cu	QN	ND	$0.01{\pm}0.003$	1.36±2.16	ND	ŊŊ	ŊŊ	ND
		Zn	39.02±6.02	$0.03 \pm 0.01$	$0.03 {\pm} 0.001$	$0.5 \pm 0.34$	$0.3 \pm 0.01$	$21.64 \pm 0.55$	$1.91 \pm 0.78$	22.87±4.75
Nakhiloo E imbricata	This study	As	$1.61 \pm 0.35$	$0.01 \pm 0.01$	$0.01{\pm}0.001$	$0.03 \pm 0.02$	$0.01 \pm 0.01$	23.29±4.79	3.49±0.3	2.36±0.67
mmy loun-		Cd	QN	ND	ND	$0.06 \pm 0.05$	$0.02 \pm 0.01$	QN	ŊŊ	ND
		Hg	5.76±1.56	$0.96 \pm 0.81$	$0.79{\pm}0.8$	$0.92{\pm}1.8$	$0.11 \pm 0.09$	4.52±0.32	4.37±0.52	$1.86 \pm 0.33$
		Pb	$0.34 \pm 0.06$	$0.01 {\pm} 0.001$	$0.01 {\pm} 0.002$	$0.44 \pm 0.35$	$0.25 \pm 0.03$	$0.12 \pm 0.03$	$0.42 \pm 1.02$	$0.33 \pm 0.14$
		Fe	81±2.66	15.73±0.65	7.18±1.73	29.86±20.08	27.52±16.92	28.26±2.47	$31.38 \pm 4.34$	126.14±16.75
		Cu	DN	QN	ND	ND	ND	QN	ND	ND
		Zn	27.11±0.48	0.07±002	$0.04 \pm 0.011$	$0.28 \pm 0.09$	$0.41 \pm 0.001$	$16.52 \pm 1.62$	5.76±0.26	29.1±7.40
Kharko E imbricata		As	$1.5 \pm 0.23$	$0.01 \pm 0.001$	ND	$0.02 \pm 0.001$	ND	12.25±3.07	$1.22 \pm 0.09$	2.23±1.24
		Cd	ΟN	QN	ΟN	$0.03 \pm 0.02$	$0.04{\pm}0.01$	ND	ND	ND
		Hg	$3.95 \pm 1.18$	QN	ΟN	ΟN	ND	2.99±0.22	$6.81 \pm 3.75$	$2.32 \pm 1.49$
		Ъb	$0.71 \pm 0.44$	$0.01 \pm 0.001$	$0.01{\pm}0.001$	$0.37 \pm 0.25$	0.77±0.53	$0.33 \pm 0.37$	$0.55 \pm 0.12$	$0.33 \pm 0.36$
		Fe	0.045	3.8		2.3				
		Cu	0.34	0.063		1.3				
		Zn	0.045	0.3		1.2				
Hong Kong C <i>mvdas</i>	[35]	As	2.5	0.17		0.22				
		Cd	ND	ŊŊ		0.0157				
		Hg	0.0015	0.00009		0.0006				
		Ъb	0.049	0.0047		0.11				

Table 5. Heavy metal accumulation in different tissues of sea turtles from different locations.

Table 5. Continued.										
		Cu	$2.28 \pm 0.35$	$3.81 \pm 1.14$		$6.64 \pm 3.09$		$1.89 \pm 0.78$		
		Zn	$34.13\pm 8.2$	$3.45 \pm 1.18$		5.56±3.91	-	37.6±3.98		
Iran-( Qeshm		As					-	$0.34{\pm}0.08$		
E. imbricata	[0]	Cd	$0.42 \pm 0.09$	$0.59{\pm}0.35$		$0.36 \pm 0.63$	-			
		Hg	$0.007 \pm 0.002$	$0.004 \pm 0.004$		$0.002 \pm 0.002$	1	$0.18\pm0.05$		
		Pb	$3.1 \pm 0.35$	2.27±0.38		4.16±3.15	1	0.56±0.25	-	
		Cu	$1.57 \pm 0.07$	$0.12 \pm 0.08$		$5.57 \pm 0.76$				
,		Zn	$34.4 \pm 3.18$	$0.59\pm0.58$		2.17±0.59				
Japan C <i>caretta</i>	[19]	Hg	12.1±3.41	$0.49 \pm 0.24$		$4.05\pm1.31$				
		Pb	$0.02 \pm 0.007$							
		Cu	$0.91 \pm 0.10$	$2.33 \pm 0.40$		$3.16 \pm 0.80$				
China C <i>mvdas</i>	[36]	Zn	120±9.6	$11.1 \pm 2.2$		$2.9\pm0.6$				
		Cd				$0.04{\pm}0.03$				
		Cd							0.57	
Cyprus C caretta	[37]	Hg							0.48	
		Pb							2.46	
		Fe								
		Cu	2.2±1.47	3.53±2.87		7.48±2.6	-	2.28±0.4		
Mexico L. olivacea	[38]	Zn	72.3±10.9	$33.6 \pm 6.1$	-	12.4±1.5	-	58.4±4.7		
		As								
		Cd	$0.24{\pm}0.1$	$0.22 \pm 0.09$		$0.47 \pm 0.09$		$0.45 \pm 0.2$		
		Cu						$1.34 \pm 0.28$		
		Zn						$11.1 \pm 0.28$		
French Guiana D.coriacea	[27]	Cd						$0.08{\pm}0.03$		
		Hg						$0.011 \pm 0.003$		
		Рb						$0.18{\pm}0.05$		
-		Cd						35.47±9.25		
Australia C. <i>mvdas</i>	[39]	Hg						$2.51{\pm}0.05$		
		Рb						22.18±5.83		

system, maternal liver, transfer to the yolk, and eventually accumulation in the hatchling. It can be an indicator of a pollutant in the aquatic environment. If the amount of pollutants in the sediments, water, and benthic organisms increases, their transfer to the adult increases, which increases the transfer of heavy elements to the hatchling [30, 31]. Studies have shown that excessive accumulation of metals in turtle bodies can destroy some tissues, such as the kidney and liver [15]. An increased concentration of heavy metals can influence sex hormones and the reproductive system [17]. These problems can severely affect the survival of this endangered species. Some elements, such as cadmium and copper, were not detected in the blood, yolk, or hatchlings. However, these metals were found in normal and abnormal eggshells. This suggests that eggshells may be a storage part that can be used as a Cd and Cu indicator for contamination.

# Comparison of Heavy Metals between Three Islands

The elemental accumulation was varied in different samples of hawksbill turtle adults, eggs, and hatchlings on three islands. There was a significant difference in Fe concentration in the hawksbill turtle egg yolk of Kharko Island compared to Ommolgorm and Nakhiloo Islands. There was also a significant difference in the acuumulation of heavy metals in hatchling muscle between Nakhiloo and the other two islands (P<0.05). In other tissues, no difference was observed between the studied islands (P>0.05) (Fig. 2). Zn accumulation showed a significant difference in yolk and blood among Nakhiloo Island and the other islands. There was a significant difference between the hatchling and adult muscle tissue from samples collected at Kharko Island and the other two islands (P<0.05). There were no significant differences in other tissues between the three islands (P>0.05) (Fig. 3). The As accumulation showed significant differences in the adult blood tissue from the three islands. The adult muscle tissue from Nakhiloo Island was significantly different compared to samples from the other two islands. There was no difference in other tissues (P>0.05) (Fig. 4). There was a significant difference in Hg accumulation in all tissues except the abnormal eggshell (Fig. 5). There were no differences between normal and abnormal egg albumin Pb accumulation in three islands (P>0.05). The adult blood tissue from Kharko was significantly different from the other two islands (P<0.05) (Fig. 6). The Cu concentration in turtle blood on Kharko Island was significantly different from the other two islands (P<0.05).

In the present study, the accumulation of heavy metals in the samples from Kharko Island was relatively higher than that from the other two islands, which could be a result of the oil terminals and petrochemical industries on Kharg Island near Kharko Island. Based on the comparison of the amount of heavy metals between hawksbill turtles nesting on the three islands and other species of sea turtles from different parts of the world, there were significant differences based on the type of tissue and species (Table 5). The accumulation of heavy metals in the yolk, blood, and hatchling was higher than in other samples. The difference in the distribution of metals between different tissues of aquatic animals depends on diet, aquatic type, age, the concentration of metals in water and sediment, the type of metal, the type of tissue, and the amount of adipose tissue [15, 18, 27]. Many factors, including habitat, diet, sex, body length, age, and tissue type, are important in the accumulation of pollutants in living organisms [16-29].

The type of nutrition and habitat are the effective factors in the biological accumulation of heavy metals in adults [32]. Hawksbill turtles in the Indo-Pacific region prefer sponges and soft corals but also feed on mollusks and crustaceans [33, 34]. So the contamination is greater than for the species that use different types of foods. The significant presence of heavy metals in the eggs indicates that the adult has been exposed to heavy metals for a long time and also indicates the presence of heavy metals in the foraging areas used by the aquatic organism's adults [28, 31]. Although the food consumed by the female turtles before they produce eggs is the primary source of contamination, the contamination of the beach sand may also contribute to the contamination of the developing embryo.

## Conclusion

In the present study, the accumulation of heavy metals in the blood, muscle tissue, yolk, albumin, normal and abnormal eggshells of adult and hatchling of hawksbill sea turtles was measured on three islands located in the north of the Persian Gulf. This is the first study of this species relating to the transfer of essential and non-essential metals in the Persian Gulf. Iron, arsenic, mercury, and lead were in high concentrations in the blood, but cadmium and copper were low. Turtles that had high concentrations of metals in their blood exhibited high levels in their egg yolks and hatchlings, indicating the transfer of metals from adults to hatchlings through the yolk (i.e., maternal transfer). The amount of accumulation of elements in adult muscle tissue was in many cases lower than in the tissue of the hatchlings, indicating the transfer of metals from the food consumed via digestion to the circulatory system, the mother's liver, the yolk, and eventually to the hatchling. Some elements, such as cadmium and copper, were not detected in the blood, yolk, or hatchlings. These elements were found in the shells of normal and abnormal eggs, which serve as the storage tissue for these elements and may be useful as an indicator of these metals. Because the concentration of some metals in the yolk was higher

than in albumin, it is recommended that turtle eggs not be used for human consumption in the Persian Gulf. The element concentrations were different in samples of hawksbill turtle adults, eggs, and hatchlings in Kharko, Ommolgorm, and Nakhiloo islands. The adult blood tissue from Kharko was significantly different from the other two islands, which could be a result of the oil terminals and petrochemical industries on Kharg Island near Kharko Island, where it is the biggest oil terminal in Iran.

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## **Conflict of Interest**

The authors declare no conflict of interest.

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