



Transfer and accumulation of trace elements in seawater, sediments, green turtle forage, and eggshells in the Xisha Islands, South China Sea

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Abstract

Chemical pollutants present a substantial threat to the survival of the green turtle (*Chelonia mydas*). In this study, the concentrations of 12 trace elements (TEs) in seawater, sediments, and green turtle forage and eggshells from the Xisha Islands in the South China Sea, along with their patterns of transfer and accumulation, were identified. The results revealed that the median TE concentrations in seawater and sediments were lower than the first-grade limit values of the national standard in China, indicating a low ecological risk. The concentrations ($\mu\text{g}\cdot\text{g}^{-1}$) of TEs in forage ranged from 0.05–0.69, 3.43–14.4, 157–2391, 27.9–124, 2.05–9.39, 0.30–9.78, 2.01–80.50, 0.18–5.76, 0.06–0.98, 2.00–18.4, 0.02–0.24, and 0.01–0.09 for Cr, Mn, Sr, Fe, Ni, Cu, Zn, Se, Cd, As, Pb, and Hg, respectively. Seawater, sediments, turtle forage, and eggshells exhibited different TE profiles, which were driven by Hg, Sr, Cr, and Pb in seawater and sediments; Fe and Ni in sediments; Cd and As in forage; and Zn, Se, and Cu in eggshells. The contents of Cu, Zn, and Se increased slightly with trophic level, indicating that they were transferred through dietary pathways. Although Cd and As appeared to bioaccumulate in green turtle forage, it was not transferred to their eggshells, which may be related to the excretion and metabolism process in the mother's body. Thus, eggshells may be a poor bioindicator for the exposure of female green turtles to these toxic elements.

Keywords Seagrass · Algae · Bioconcentration factor · Cd · As · Essential elements

Abbreviations

TE	Trace element
BAF	Bioaccumulation factor
BSAF	Biological sediment accumulation factor
PCA	Principal component analysis
OM	Organic matter
AB	Arsenobetaine
DMA(V)	Dimethylarsinic acid

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Introduction

Due to population decline, green turtles (*Chelonia mydas*) have been listed as globally endangered species on the International Union for Conservation of Nature (IUCN) Red List (Semionoff 2004). Among other risks, chemical pollution poses a substantial threat to the survival of sea turtles. Some of trace elements (TEs) with toxic and chronic effects threaten the health and survival of marine organisms and coastal wetlands. Sea turtles are long-lived vertebrates with high fidelity to particular foraging areas; long-term exposure to some TEs (especially heavy metals) may impact their health and fecundity (Camacho et al. 2014; Villa et al. 2017).

Adult green turtles inhabit nearshore and inshore neritic zones and principally feed on seagrass and macroalgae (Bjørndal, 1997). Trace element exposure in sea turtles is strongly associated with the specific forage habitat used and individual food preferences (Gardner et al. 2006). The dietary composition of green turtle depends on the forage species present and their physiological stage. In addition to seagrass, Rhodophyta, Chlorophyta, and Phaeophyta are important dietary items of gravid green turtles (Stokes

et al. 2019; Esteban et al. 2020). However, seagrass and macroalgae have the capacity to selectively absorb Zn, Cd, and Cu and accumulate them in concentrations up to thousands times higher than their respective levels in environmental media (Zheng et al. 2018). Moreover, TE accumulation in seagrass and macroalgae can vary widely across geographic regions (e.g., different pollutant concentrations or background values) and species (e.g., different uptake capacities and retention) (Sánchez-Quiles et al. 2017; Bonanno and Orlando-Bonaca, 2018). Therefore, it is important to monitor TE pollution in the forage species and habitat media of these endangered animals and to identify bioaccumulation patterns.

Recent studies have shown that some TEs (e.g., As or Co) levels in sea turtle tissues (e.g., blood and carapaces) are consistent with those in their forage (Komoroske et al. 2012; Thomas et al. 2020). It is known that female birds and reptiles may eliminate TEs by depositing them into eggs; thus, eggs become suitable bioindicators of pollution. In particular, Cu, Cd, and Pb contents in bird (*Corvus frugilegus*) eggshells are significantly correlated with those of their breeding habitat (Orłowski et al. 2016). The eggshells of sea turtles are secreted one week before spawning (Miller, 1985). Thus, eggshells may be a reliable and non-invasive indicator of recent pollutant exposure in female sea turtles in their breeding habitats.

The South China Sea hosts the largest sea turtle population in China, 90% of which are green turtles (Chan et al. 2007). In particular, the Xisha Islands are typical tropical islands that host diverse reef ecosystems and serve as key breeding habitats for green turtles (Jia et al. 2019). Some studies suggest potential anthropogenic input of various TMs (e.g., Pb, Cd, and Hg) to the seawater and coral sands in the Xisha Islands (Zhou et al. 2007; Wang et al. 2017). To the best of our knowledge, information about TE accumulation in local green turtle forage is lacking. Moreover, only Komoroske et al. (2012) used tissue (carapace)–forage biological magnification factor to discuss the TEs bioaccumulation patterns in green sea turtle food web. In this study, we investigated the concentrations and profiles of TEs in seawater, sediments, green turtle forage, and eggshells in the Xisha Islands. Using the eggshells of green turtles, we could give information the bioaccumulation patterns and transfer pathway for a suite of essential and nonessential TEs in the food chain of the green turtle.

Materials and methods

Study area

The Xisha Islands (15°47'–17°08'N, 110°10'–112°55'E) are located in the South China Sea and comprise over 100

small coral islands, sandbanks, and reefs. The islands can be organized into two groups: the eastern Xuande Archipelago and the western Yongle Archipelago. The temperatures vary seasonally between 13 °C and 25 °C (Chen et al. 2012). It is an important waterway that connects China with the Malay Archipelago, Indo-China Peninsula, and Indian Ocean rim states. The islands have a complex and unique tropical marine ecosystem and serve as a critical habitat for green turtles. The Qilianyu cluster in the Xuande Archipelago hosts the largest currently known nesting population of green turtles in China (Jia et al. 2019).

Sample collection

Surface water and sediments (coral sands) from 38 stations were collected from eight islands and sandbanks in the Qilianyu cluster (July–October, 2020), including West Sand ($n=4$), Zhao Shu Island (inhabited, $n=6$), North Island ($n=7$), Middle Island ($n=6$), South Island ($n=6$), North Sand ($n=3$), Middle Sand ($n=3$), and South Sand ($n=3$) (Fig. 1). Coastal surface seawater and sediments were collected at intervals of 100 m in the direction perpendicular to the coast. The sample from each station was composed of three subsamples. Surface sediments were sampled with a stainless steel grab sampler and transferred to airtight polyethylene bags. Seawater was collected using a water sampler and stored in a polyethylene plastic bottle. The pH of the seawater and sediments (distilled water [mL]–dry sediment [g]=2.5:1) was measured in the laboratory using a pH meter (FE28 – Standard, Mettler Toledo, USA). Seawater samples were filtered through a 0.45- μm microfiltration membrane and treated with concentrated nitric acid to $\text{pH}<2$. Seawater samples were preserved in fresh conditions (4 ± 1 °C), and sediment samples were frozen (-18 ± 2 °C) until further laboratory analysis.

Three samples of the aboveground tissues of forage taxa, namely, *Caulerpa racemosa*, *Caulerpa serrulata*, *Caulerpa taxifolia*, and *Codium fragile* (Chlorophyta); *Ishige sinicola*, *Colpomenia sinuosa*, *Padina australis*, and *Sargassum crassifolium* (Phaeophyta); *Asparagopsis taxiformis*, *Hypnea pannosa*, *Ceratodictyon spongiosum*, and *Turbinaria ornate* (Rhodophyta); and *Thalassia hemprichii* (Hydrocharitaceae), were randomly collected from the nearshore area of the Qilianyu cluster. The thalli and leaves were carefully cut with stainless steel scissors, and the weight of the samples varied from 0.25 to 1 kg. Post-hatch eggshells were collected from 40 nests in the Qilianyu cluster in our previous work (Jian et al. 2021). In the laboratory, biological samples were washed with ultrapure water to remove any particulate matter and then closed in hermetic polyethylene plastic bags, and frozen (-18 ± 2 °C) until further laboratory analysis. All required glassware and plastic materials were soaked in dilute nitric acid (1:3) for more than 24 h.

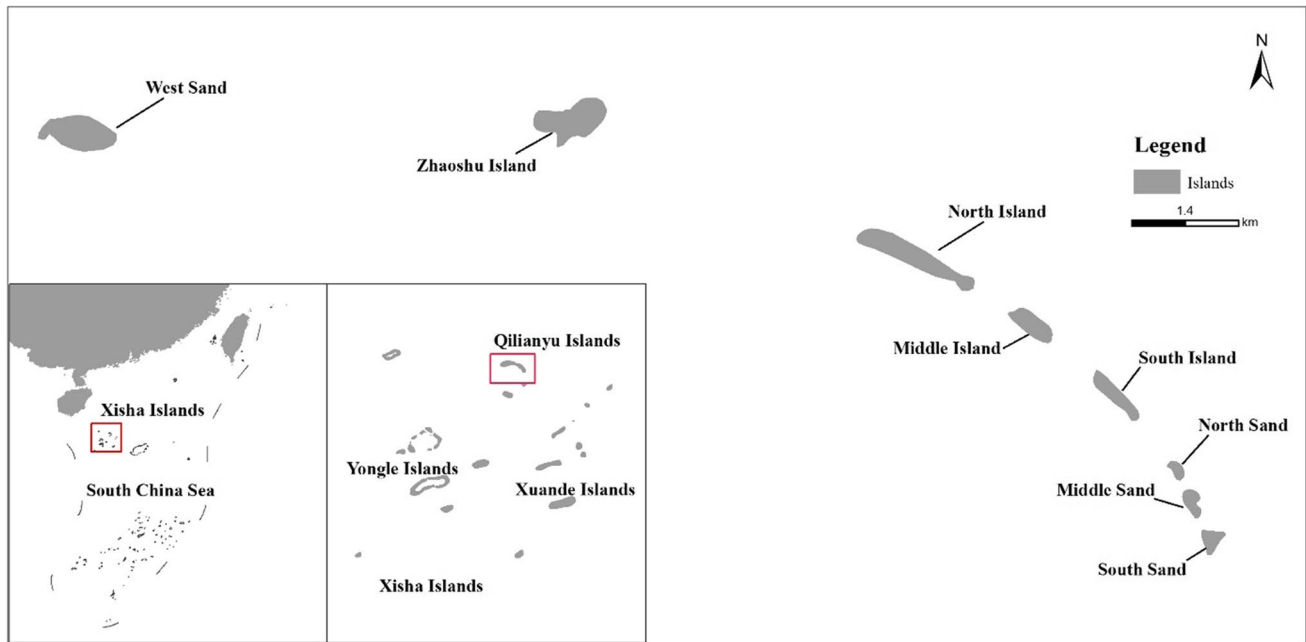


Fig. 1 Location of the collection site of environmental media, green turtle eggshells and forage from Xisha Islands, China

TE analysis

In the laboratory, sediment samples were dried naturally (3~5 days) and then ground to <math><0.125\text{ mm}</math>. Biological samples were freeze-dried and cut into tiny pieces. The analytical procedure was similar to that described by Bonanno et al. (2020). The resulting dry material was weighed at \text{HNO}_3 and 2 mL of 30% H_2O_2). Sample digestion was conducted at room temperature (25 °C) overnight, and the samples were then further digested with a microwave digestion instrument (MARS6, CEM Corp., USA). The digestion process was as follows: 0–15 min at 120 °C, 15–30 min at 200 °C, and 30–60 min at 200 °C. The digestion solution was evaporated at 95 °C to ~1 mL and then diluted to 25 mL with ultrapure water for analysis.

Zinc, Fe, Mn, Se, Cr, Cu, Pb, Ni, Cd, and Sr contents were measured using inductively coupled plasma–mass spectrometry (ICP-MS, X Series 2, Thermo Fisher Scientific, USA). As and Hg were analyzed using atomic fluorescence spectrometry (AFS-3000, Beijing Haiguang, China). Ten of the TEs in eggshells have been analyzed in our previous study (Jian et al. 2021), while Ni and Hg contents were determined in the present study. An online injection technique (including online dilution and online preconcentration) combined with ICP-MS was used to analyze dissolved TEs in seawater samples (Mu et al. 2015). The detection limits were calculated as three times the standard deviation for digestion blanks (sediments and biological sample) and acid blanks (2% HNO_3 , seawater) ($n > 20$). The detection limits of TEs for sediment

and biological samples were 0.005, 0.005, 0.05, 0.05, 0.002, 0.01, 0.01, 0.01, 0.001, 0.002, 0.02, and 0.001 $\mu\text{g}\cdot\text{g}^{-1}$ for Cr, Mn, Sr, Fe, Ni, Cu, Zn, Se, Cd, Pb, As, and Hg, respectively. The detection limits of TEs for seawater were 0.01, 0.05, 0.5, 0.5, 0.01, 0.05, 0.1, 0.4, 0.005, 0.003, 0.3, and 0.01 $\mu\text{g}\cdot\text{L}^{-1}$ for Cr, Mn, Sr, Fe, Ni, Cu, Zn, Se, Cd, Pb, As, and Hg, respectively.

Quality control and quality assurance

Quality control and quality assurance procedures for TEs in seawater, sediments, and seagrass/algae were estimated using duplicate samples, with three replicates for every 12 samples, blank samples, as well as with standard reference materials (SRM–GBW 07,314, offshore marine sediments, for sediments; SRM–GBW 080,040, for seawater; and SRM–GBW 08,517, standard for compositional analyses of kelp, seagrass, and algae). These certified reference materials were provided by the Second Institute of Oceanography (Zhejiang, China). For seawater, sediment, and biological samples, relative recoveries ranged from 90.4 to 108.9%, which was within 10% of the certified values.

Data analysis

The bioaccumulation factor (BAF) and the biological sediment accumulation factor (BSAF) of green turtle forage are calculated to evaluate the concentration of accumulated TEs in seagrass and algae samples based on their measured concentrations in seawater and sediments:

$$BAF = \frac{C_a}{C_w} \tag{1}$$

$$BSAF = \frac{C_a}{C_s} \tag{2}$$

where C_a , C_w , and C_s are the median concentrations of TEs in seagrass/algal species, seawater, and sediments, respectively.

Statistical calculations were performed using SPSS v. 23.0 (IBM Corp., USA). Normality of the TE concentration data in the different samples for all stations was checked using the Shapiro–Wilk test. Descriptive statistics, including median, maximum, minimum, and relative standard deviation (RSD), were used to report the amount of TEs in seawater, sediment, and seagrass/algae. Data of TE concentrations in eggshells from our previous studies (Jian et al. 2021) were used to analyze TE transfer. Non-parametric tests of multiple independent samples (Jonckheere–Terpstra) were used to analyze the differences in TE concentrations among the marine macrophytes. Spearman correlation analysis was also conducted to determine the relationships among the elemental concentrations in the sediments, and correlations were considered statistically significant if $p < 0.05$. Principal component analysis (PCA) was used to explore the relationships among the concentrations of TEs in seawater, sediments, and the forage and eggshells of green turtles.

Results

TE concentrations in seawater and sediment

The median concentrations ($\mu\text{g}\cdot\text{L}^{-1}$) of the 12 TEs in surface seawater decreased in the following order: Sr (5520) > Fe (6.70) \approx Zn (6.65) > Mn (1.91) > As (1.51) > Se (1.06) > Cr (0.77) > Cu (0.63) > Pb (0.36) > Ni (0.34) > Cd (0.05) > Hg (0.03) (Table 1). The median concentrations of TEs in seawater were lower than the first-grade limit values of the GB (3097–1997) national standard in China (SEPA, 1997) and the biological chronic toxicity criteria of heavy metals in seawater used by the United States National Oceanic and Atmospheric Administration (Buchman 2008) (Table 1). However, the concentration of Zn at one of the sampling sites in West Sand and those of Pb and Hg at two sampling sites in South Sand slightly exceeded the first-grade limit values (20.0, 1.0, and 0.05 $\mu\text{g}\cdot\text{L}^{-1}$ for Zn, Pb, and Hg, respectively). In addition, the median concentrations of Zn, Pb, and Cu in seawater almost reached 10–80 times the background values (BV) in the South China Sea (Yu, 2003) and exhibited greater variability, indicating that these elements were affected by human inputs.

The median concentrations ($\mu\text{g}\cdot\text{g}^{-1}$) of TEs in the sediments were in the order Sr (4680) > Fe (75.0) > Ni (0.34) > Mn (1.91) > Zn (6.65) > Cr (0.77) > Pb (0.36) > As (0.66) > Cu (0.63) > Se (0.16) > Cd (0.07) > Hg (0.05) (Table 1). The median concentrations of Cr, Ni, Cu, Zn, Cd, As, Pb, and Hg in sediments were lower than the first-grade limit values of the GB (18,668–2002) national standard in

Table 1 Trace element concentrations in seawater and sediments from Qilianyu cluster. *Min* minimum value, *max* maximum value, *median* the median concentration, *RSD* relative standard deviation, *BV* background value, *NA* no data, *SWQ-1* sea water quality of primary standard criteria (GB 3097–1997), *MSQ-1* marine sediment

TE		Cr	Mn	Sr	Fe	Ni	Cu	Zn	Se	Cd	As	Pb	Hg
Seawater ($\mu\text{g}\cdot\text{L}^{-1}$)	Min	0.18	0.95	4848	0.70	0.15	0.40	2.21	0.59	0.01	1.17	0.05	0.03
	Max	2.84	5.93	5725	20.8	0.70	5.66	26.6	1.34	0.07	3.64	4.34	0.09
	Median	0.77	1.91	5520	6.70	0.34	0.63	6.65	1.06	0.05	1.51	0.36	0.03
	RSD/%	59.2	46.3	3.98	66.7	38.6	102	61.1	25.0	46.2	24.9	132	37.1
	BV	NA	NA	NA	NA	NA	0.08	0.08	NA	0.004	NA	0.05	NA
	SWQ-1	≤ 50.0	NA	NA	NA	≤ 5.0	≤ 5.0	≤ 20.0	NA	≤ 1.0	≤ 20.0	≤ 1.0	≤ 0.05
	CTC	50	NA	NA	NA	8.2	3.1	81	71	9.3	36	8.1	0.94
Sediments ($\mu\text{g}\cdot\text{g}^{-1}$)	Min	0.89	4.49	3699	61.1	6.35	0.14	0.80	0.07	0.02	0.39	0.35	0.01
	Max	2.24	20.3	5804	143	9.28	2.19	4.86	0.35	0.39	1.60	1.12	0.16
	Median	1.43	6.91	4860	75.0	7.60	0.28	2.16	0.16	0.07	0.66	0.72	0.05
	RSD/%	20.8	38.5	8.11	25.2	9.2	92.7	43.9	33.7	72.0	35.8	27.2	64.6
	BV	39.3	NA	NA	NA	NA	7.43	54.4	NA	0.18	9.71	15.6	0.02
	MSQ-1	≤ 80.0	NA	NA	NA	NA	≤ 35.0	≤ 150	NA	≤ 0.50	≤ 20.0	≤ 60.0	≤ 0.20
	TEL	52.3	NA	NA	NA	15.9	18.7	124	NA	0.676	7.24	30.24	0.13

quality of primary standard criteria (GB 18,668–2002), respectively. CTC is the biological chronic toxicity criteria of heavy metals in seawater used by National Oceanic and Atmospheric Administration (NOAA), and TEL is the threshold effect levels of heavy metals in sediment used by NOAA

China (SEPA, 2002) and the threshold effect levels used by Buchman (2008), thereby indicating a low ecological risk. However, the concentrations of Cd in sediments at two sampling sites and those of Hg in sediments at 80% of the sampling sites were 2–8 times higher than the BV (shelf area of South China Sea sediment) and exhibited greater variability. Copper and Zn concentrations were also more variable in the sediment samples (RSD > 36%), and there was a significant association between these elements ($r=0.38$, $p<0.05$). Large RSD values generally indicate artificial sources (Zhang et al. 2019). Boats always have Cu-based antifouling paints, and Zn-based sacrificial anodes may serve as artificial sources of Cu and Zn (Egardt et al. 2018).

TEs in seagrass and algae

The median concentrations ($\mu\text{g}\cdot\text{g}^{-1}$) of the 12 TEs in seagrass and algae decreased in the following order: Sr (1248) > Fe (68.0) > Zn (13.6) > Mn (6.24) > As (5.95) > Ni (5.31) > Cu (1.41) > Se (0.72) > Cd (0.33) > Cr (0.25) > Pb (0.10) > Hg (0.01) (Table 2). Apart from Sr, Fe, Zn, and Mn were the most abundant elements in all seagrass and algae samples, suggesting their important roles in plant metabolism. Meanwhile, less Cr, Cd, Pb, and Hg accumulated in these samples. Compared with other sea turtle habitats worldwide, the concentrations of Cr, Mn, Fe, and Pb in seagrass and algae from the Xisha Islands were considerably lower (10–76x, 4–12x, 7–30x, and 37–289x, respectively), while those of Cu, Zn, As, and Cd were essentially in the same range (Çelik et al. 2006; Lewis et al. 2007; Riosmena-Rodríguez et al. 2010; Sánchez-Quiles et al. 2017; Thomas et al. 2020) (Table 2).

Trace element concentrations in seagrass and algae from the Xisha Islands are shown in Fig. 2. The highest median concentrations of Mn, Fe, Zn, Ni, Se, and Cd were observed in rhodophytes, but only Se differed significantly ($p<0.05$) from the other clades. The highest concentrations of Fe, As, Pb, and Cr were observed in Phaeophyta, and the median contents of Cr in this clade were significantly higher ($p<0.05$) than those in seagrass and chlorophytes. The highest median As concentrations were observed in chlorophytes, although they did not differ significantly from those found in other clades. The median concentrations of Cu in seagrass and rhodophytes were higher than those in other algae, and the Sr contents in seagrass and Phaeophyta were higher than in other clades.

Distribution of TEs in algae, seawater, sediments, and green turtle eggshells

Trace element concentrations in sea turtle tissues depend on their diet and feeding area (Gardner et al. 2006). Therefore, we compared TE concentrations in the eggshells of green

turtles to those in environmental media, seagrass, and algae, and PCA was used to investigate their distributions (Fig. 3). The Log_{10} -transformed concentrations of TEs in seawater, sediments, and seagrass/macroalgae are shown in Fig. 4, together with those from green turtle eggshells previously reported by Jian et al. (2021).

The PCA results showed that 12 TEs were reduced to three dimensions (PC1, PC2, and PC3), which collectively explained 87.1% of the total variation (Fig. 3). The element cluster with high-representation element Sr (loading = 0.56) along PC1 explained 61.0% of the total variation, Fe and Ni (loadings: Fe = 0.46, Ni = 0.58) together along PC2 explained 15.7% of the total variation, and Cd and As along PC3 (loading > 0.5), explained 10.1% of the total variation, suggesting that these TEs strongly influenced each respective component. A scale 3 correlation biplot was used to separate the 12 TEs into four clusters of strongly correlated elements. Seawater and sediment TE profiles differed from those of seagrass and algae. Additionally, the TE profile of green turtle eggshells differed from the sediment profile. Iron and Ni clusters were more closely associated with sediments, while Cu, Zn, and Se were closely associated with eggshells.

Overall, TE bioaccumulation in seagrass/algae did not uniformly correspond to their enrichment in the surrounding media. Boxplots confirmed that Cr, Sr, Pb, and Hg levels in seagrass and algae were lower than those in the surrounding seawater and sediments, and both the BAF and BSAF of the seagrass and algae for Cr, Sr, Pb, and Hg were < 1, indicating that these elements were confined to the seawater and sediments, and transfer to the biosphere was not observed (Table 3). The BSAF values of seagrass and algae for Fe, Ni, and Mn were also < 1, whereas the BAF value for Fe, Ni, and Mn were 8.58, 15.6, and 3.27, respectively, indicating bioaccumulation from seawater. Furthermore, the Cu, Zn, As, and Cd concentrations in seagrass and algae were higher than those in seawater and sediments, and both the BAF and BSAF of seagrass and algae for these elements ranged from 1.31 to 18.9, indicating bioaccumulation. Specifically, the BSAF of seagrass for Cu and the BSAF of rhodophytes for Zn, Se, and Cd were greater than ~ 10, indicating strong bioaccumulation.

The concentrations of Cr, Zn, Se, and Cu in green turtle eggshells were higher than those of seagrass and algae (3.4-fold for Cr, 1.5-fold for Zn, 3.4-fold for Se, 9.1-fold for Cu) (Fig. 3), indicating that female turtles possibly bioaccumulate these elements via forage and then transfer them to their offspring. Meanwhile, the Mn, Fe, Ni, Sr, Cd, and As contents in green turtle eggshells were 43–74-fold lower than those in seagrass and algae. Overall, we did not observe strong biomagnification for most of the TEs through eggshells. In particular, the contents of essential elements (Cu, Zn, and Se) increased slightly

Table 2 Trace elements concentrations in seagrass and algae samples from this study compared to seagrass and algae metal concentrations reported from other regions and global literature ($\mu\text{g}\cdot\text{g}^{-1}$, dry weight). ^aMedian (min–max value); ^bmean \pm SD; ^cRang; ^d0.2-truncated mean.

nd is not detected. *Chl* Chlorophyta, *Rho* Rhodophyta, *Pha* Phaeophyta. HWK is Howicks in Queensland, Australia; UPS is Upstart Bay in Queensland, Australia

TE	Location	Seagrass	Chl	Rho	Pha	Total	Reference
Cr	Xisha Islands, China ^a	0.25 (0.18–0.32)	0.30 (0.05–0.32)	0.43 (0.42–0.55)	0.48 (0.22–0.69)	0.25 (0.05–0.69)	Present study
	Queensland, Australia ^b	5.42 \pm 1.88	NA	NA	NA	NA	Thomas et al. 2020
	Mersin, Turkey ^b	22.9 \pm 0.14	20.4 \pm 0.11				Çelik et al. 2006
	Florida, USA ^b	0.70 \pm 1.1					Lewis et al. 2007
	Mediterranean ^c	0.01–123	0.31–104	0.17–26	0.07–82.8		Bonanno and Orlando-Bonaca, 2018
	Global ^d	3.2	2.73	1.29	2.51	2.33	Sánchez-Quiles et al. 2017
Mn	Xisha Islands, China ^a	6.24 (6.21–14.4)	3.67 (3.51–5.78)	6.86 (6.81–14.3)	5.22 (3.43–7.86)	6.24 (3.43–14.4)	Present study
	Queensland, Australia ^b	35.0 \pm 7.67					Thomas et al. 2020
	Baja California, México ^c	33.9–78.6	7.30–61.4	10.5–61.4			Riosmena-Rodríguez et al. 2010
	Mediterranean ^c	4.22–1600	6.30–416	5.60–475	7.89–757		Bonanno and Orlando-Bonaca, 2018
		Global ^d	25.0	93.1	35.6	71.7	55.9
Fe	Xisha Islands, China ^a	57.5 (49.3–68.0)	75.6 (69.2–115)	81.9 (61–102)	45.5 (27.9–124)	68.0 (27.9–124)	This study
	Queensland, Australia ^b	1700 \pm 827					Thomas et al. 2020
	Mersin, Turkey ^b	129 \pm 26.2					Çelik et al. 2006
	Baja California, México ^c	51.1–630	224–524	142–772			Riosmena-Rodríguez et al. 2010
		Global ^d	412	487	293	382	422
Ni	Xisha Islands, China ^a	6.12 (4.97–8.59)	2.10 (2.10–2.20)	7.10 (6.61–9.39)	5.26 (2.11–8.11)	5.31 (2.05–9.39)	Present study
	Queensland, Australia ^b	3.04 \pm 0.90					Thomas et al. 2020
	Mersin, Turkey ^b	23.8 \pm 0.22	21.8 \pm 0.12				Çelik et al. 2006
	Baja California, México ^c	2.80–3.10	1.80–7.90	1.10–11.0			Riosmena-Rodríguez et al. 2010
	Florida, USA	< 3.50					Lewis et al. 2007
		Mediterranean ^b	0.20–123	0.54–32.0	0.33–52.6	0.63–50.5	
	Global ^d	6.44	4.27	3.41	6.31	5.13	Sánchez-Quiles et al. 2017

Table 2 (continued)

TE	Location	Seagrass	Chl	Rho	Pha	Total	Reference
Cu	Xisha Islands, China ^a	4.58 (2.91–4.93)	0.99 (0.88–1.40)	4.38 (1.41–9.78)	0.99 (0.30–2.51)	1.41 (0.30–9.78)	Present study
	Queensland, Australia ^b	2.47 ± 3.30					Thomas et al. 2020
	Mersin, Turkey ^b	4.52 ± 1.01	4.92 ± 0.25				Çelik et al. 2006
	Baja California, México ^c	0.40–1.60	1.00–7.30	0.50–2.60			Riosmena-Rodríguez et al. 2010
	Florida, USA ^c	5.00–20.5					Lewis et al. 2007
	Mediterranean ^c	0.19–148	0.45–253	0.35–45.2	1.00–103		Bonanno and Orlando-Bonaca, 2018
	Global ^d	9.88	7.67	5.19	6.93	7.94	Sánchez-Quiles et al. 2017
Zn	Xisha Islands, China ^a	15.3 (14.5–20.0)	15.1 (8.40–19.3)	37.6 (13.6–80.5)	8.70 (2.01–26.7)	13.6 (2.01–80.5)	Present study
	Baja California, México ^c	13.5–16.5	5.70–14.2	6.40–13.4			Riosmena-Rodríguez et al. 2010
	Florida, USA ^c	3.40–7.30					Lewis et al. 2007
	Mediterranean ^c	5.00–787	2.81–369	0.14–248	1.60–780		Bonanno and Orlando-Bonaca, 2018
		Global ^d	39.6	37.7	29.8	58.9	40.5
Se	Xisha Islands, China ^a	0.41(0.41–0.55)	0.47(0.18–0.72)	3.02(2.07–5.76)	0.98(0.29–3.46)	0.72(0.18–5.76)	Present Study
	Black Sea (Turkey) ^c		0.02–0.69	0.01–0.25	0.03–0.09		Tuzen et al. 2009
Cd	Xisha Islands, China ^a	0.35 (0.25–0.53)	0.33 (0.32–0.70)	0.81 (0.67–0.98)	0.18 (0.06–0.94)	0.33 (0.06–0.98)	Present study
	Queensland, Australia ^b	0.20 ± 0.07					Thomas et al. 2020
	Mersin, Turkey ^b	1.40 ± 0.09	1.31 ± 0.07				Çelik et al. 2006
	Baja California, México ^c	nd–2.20	1.20–2.30	1.90–4.80			Riosmena-Rodríguez et al. 2010
	Mediterranean ^c	0.01–85.7	0.01–31.5	0.02–31.0	0.01–16.9		Bonanno and Orlando-Bonaca, 2018
		Global ^d	0.99	0.49	0.59	1.13	0.83
Sr	Xisha Islands, China ^a	1760 (403–2391)	680 (192–1523)	657 (157–1108)	1685 (1248–2049)	1248 (157–2391)	Present study
	Queensland (HWK) ^b	3208 ± 658					Thomas et al. 2020
	Queensland (UPS) ^b	315 ± 142					

Table 2 (continued)

TE	Location	Seagrass	Chl	Rho	Pha	Total	Reference
As	Xisha Islands, China ^a	5.12 (5.00–6.86)	6.84 (6.65–8.87)	5.95 (5.75–14.1)	4.94 (3.89–18.4)	5.95 (2.00–18.4)	Present study
	Queensland, Australia ^b	5.97 ± 5.81					Thomas et al. 2020
	Mediterranean ^b	0.31–34.8	0.10–23.0	0.84–31.0	0.80–242		Bonanno and Orlando-Bonaca, 2018
	Global ^d	9.16	8.94	10.5	38	15.2	Sánchez-Quiles et al. 2017
Pb	Xisha Islands, China ^a	0.03 (0.02–0.10)	0.09 (0.08–0.09)	0.15 (0.10–0.17)	0.16 (0.06–0.24)	0.10 (0.02–0.24)	Present study
	Queensland, Australia ^b	1.12 ± 0.33					Thomas et al. 2020
	Baja California, México ^b	nd–2.5	nd–0.7	nd–0.6			Riosmena-Rodríguez et al. 2010
	Mersin, Turkey ^b	8.67 ± 0.64	6.26 ± 0.68				Çelik et al. 2006
	Mediterranean ^c	0.03–900	0.01–737	0.01–878	0.01–617		Bonanno and Orlando-Bonaca, 2018
Global ^d	5.11	6.02	3.16	5.91	5.27	Sánchez-Quiles et al. 2017	
Hg	Xisha Islands, China ^a	0.02 (0.01–0.02)	0.02 (0.01–0.02)	0.01 (0.01–0.09)	0.01 (0.01–0.03)	0.01 (0.01–0.09)	Present study
	Mediterranean ^c	0.01–1.28	0.05–26.1	0.04–0.11	0.04–0.27		Bonanno and Orlando-Bonaca, 2018
	Global ^d	0.05	0.10	0.09	0.07	0.06	Sánchez-Quiles et al. 2017

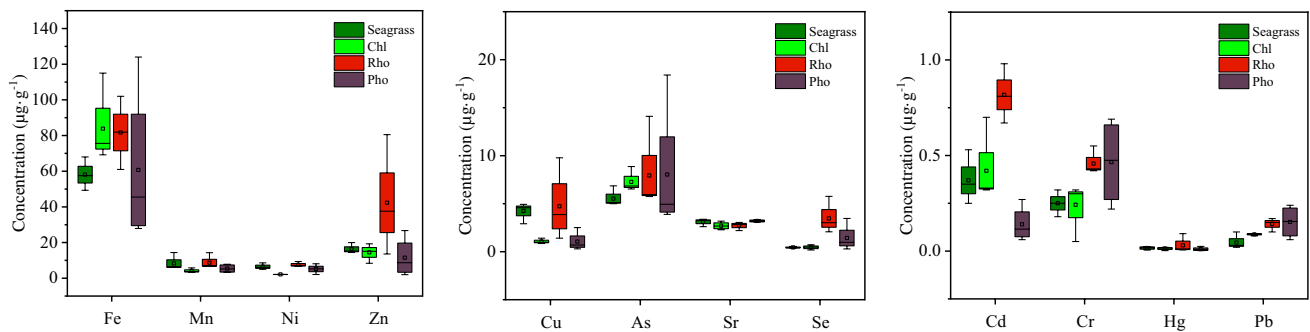


Fig. 2 Trace element concentration in seagrass and algae. Sr is expressed as a log 10 value. Seagrass and algae are marked by color; seagrass = bottle green; Chlorophyta = Light green; Rhodophyta = red; Phaeophyta = brown. The boundary of the box closest to zero indi-

cates the 25th percentile, a black line within the box marks the median, and the boundary of the box farthest from zero indicates the 75th percentile

with trophic level, indicating trophic transfer, especially for Cu. Notably, the nonessential elements Cd and As accumulated at high concentrations (As, 5.95 µg·g⁻¹; Cd, 0.33 µg·g⁻¹) in seagrass and algae but were present at low levels in green turtle eggshells (As, 0.07 µg·g⁻¹;

Cd, 0.01 µg·g⁻¹), indicating that these elements are not effectively transferred through food or from mothers to eggshells.

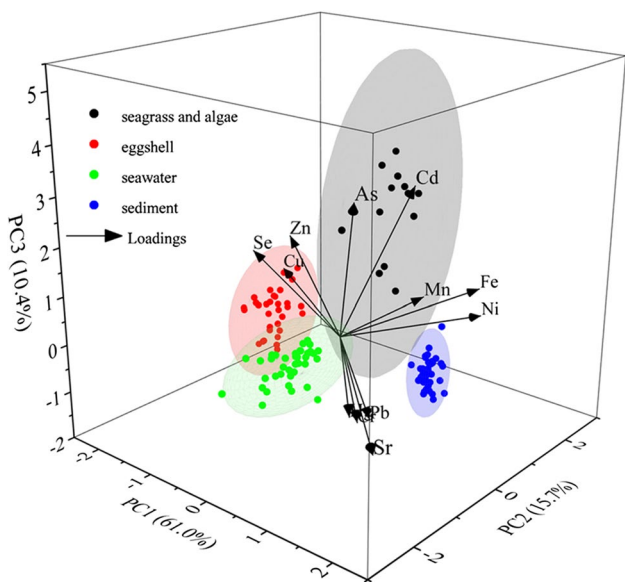


Fig. 3 The relative variation biplot for seagrass/algae, seawater, sediments, and green turtle eggshells. The data of Ni and Hg is from present study; other TMs are from Jian et al. (2021). Different samples are marked by color; seagrass and algae=black; green turtle eggshells=red; seawater=green; sediments=blue. PC1 explains most (61.0%) of the total variation; PC2 and PC3 explain 15.7% and 10.4% of the total variation

Discussion

TE transfer to primary producers

Significant interspecific differences in the accumulation of TEs in algae within the same region could be attributed to internal factors, such as different uptake capacities and retention (Fariás et al. 2007). In this study, rhodophyte species accumulated more Se than phaeophytes and Chlorophyte taxa ($p < 0.05$), which is consistent with the findings of Maher et al. (1992). Marine algae take up and bioaccumulate selenate from seawater as a sulfur analog, and Phaeophyta typically contain smaller amounts of amino acids and proteins than Rhodophyta (Vriens et al. 2016). High TE concentrations in Phaeophyta have been related to cell wall polysaccharide alginate, which exhibits a high chelating action for free metal cations (Sánchez-Quiles et al. 2017). In the Xisha Islands, we found higher concentrations of Fe, As, Pb, Cr, and Sr reported for Phaeophyta (especially in *Ishige sinicola*) as compared to the other clades, while there was only a significant difference for Cr contents ($p < 0.05$). Overall, most of the TEs were not significantly different among the four species of algae in the Xisha Islands, which may be related to intraspecific difference. In general, intraspecific variations in TE composition may be related to differences in biochemical composition, thallus morphology, and growth strategy (Malea and Kavrekidis 2014).

The bioaccumulation of TEs is likely related to their availability in the surrounding media. Dissolved TEs in

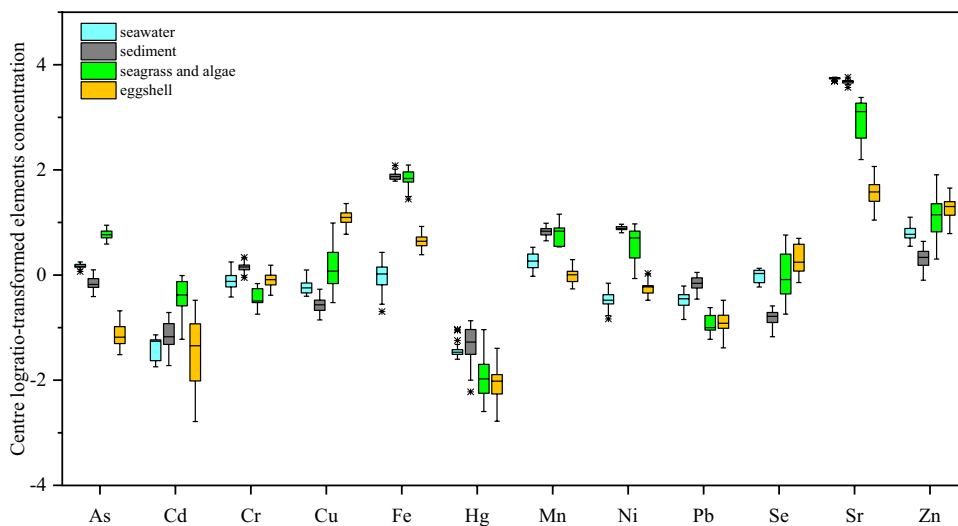


Fig. 4 Boxplots of log₁₀-transformed TEs concentrations in seagrass/algae, seawater, sediments, and eggshells. Different samples are marked by color; seagrass and algae=green; green turtle eggshells=yellow; seawater=blue; sediment=gray. The boundary of the box closest to zero indicates the 25th percentile, a black line within

the box marks the median, and the boundary of the box farthest from zero indicates the 75th percentile. The black line above and below the box indicates “minimum” and “maximum.” Asterisk above and below the box indicate outliers

Table 3 The bioaccumulation factors and biological sediment accumulation factors of TEs in seagrass and algae

TE	BAF					BSAF				
	Seagrass	Chl	Rho	Pha	Total	Seagrass	Chl	Rho	Pha	Total
Cr	0.32	0.39	0.56	0.62	0.32	0.17	0.21	0.30	0.34	0.17
Mn	3.27	1.92	3.59	2.73	3.27	0.90	0.53	0.99	0.76	0.90
Sr	0.32	0.12	0.12	0.31	0.23	0.36	0.14	0.14	0.35	0.26
Fe	8.58	8.58	8.58	8.58	8.58	0.77	1.01	1.09	0.61	0.91
Ni	18.0	6.18	20.9	15.5	15.6	0.60	0.13	0.58	0.13	0.19
Cu	7.27	1.57	6.95	1.57	2.24	16.4	3.54	15.6	3.54	5.04
Zn	2.30	2.27	5.65	1.31	2.05	7.08	6.99	17.4	4.03	6.30
Se	0.39	0.44	2.85	0.92	0.68	2.56	2.94	18.9	6.13	4.50
Cd	7.00	6.60	16.2	3.60	6.60	5.00	4.71	11.6	2.57	4.71
As	3.39	4.53	3.94	3.27	3.94	7.76	10.4	9.02	7.48	9.02
Pb	0.08	0.25	0.42	0.44	0.28	0.04	0.13	0.21	0.22	0.14
Hg	0.67	0.67	0.33	0.33	0.33	0.40	0.40	0.20	0.20	0.20

seawater are most easily accessible to living organisms, including algae. Studies have shown that seagrass can absorb dissolved TEs (e.g., Zn, Cd, Cu, and Mn) from the surrounding water and store it in their tissues, especially in their blades (Li and Huang, 2012). It has also been demonstrated that macroalgae (Phaeophyta and Rhodophyta) can take up and bioaccumulate arsenate and Se from surrounding seawater (Fariás et al. 2007; Schiavon et al. 2017). In this study, the relatively high concentrations of Zn, Cu, As, and Se in seawater and BSAF values < 1 for these elements in seagrass and algae indicate that seagrass and algae may absorb dissolved TEs from seawater.

The bioavailability and mobility of TEs in sediments depend on their chemical speciation, which is controlled by several physicochemical factors, such as pH, the reduction–oxidation potential, and organic matter (OM) content (Stumm and Morgan, 1995). The pH values of seawater (8.08–8.28) and sediments (7.43–7.97) in the Xisha Islands ranged from neutral to weakly alkaline. Under alkaline or near-neutral and oxidizing conditions, Fe and Mn are present as insoluble ferric (Fe^{3+}) and manganic (Mn^{4+}) oxides and hydroxides (Stumm and Morgan, 1995). These hydroxides further coprecipitate metal ions (e.g., Zn and Cd), and thus, their availability decreases. This may explain why the BSAF values of seagrass/algae for Fe and Mn were < 1, and a significant relationship was observed between Cd and Mn ($r=0.506$, $p<0.01$, $n=38$) and between Zn and Fe ($r=0.324$, $p<0.05$, $n=38$) in the sediment. Additionally, TEs adsorbed on the negatively charged sites of clays and OM are more exchangeable (Stumm and Morgan, 1995), and Cu can easily form complexes with OM because of the high stability constant of organic Cu compounds (Stumm and Morgan, 1995).

Trace elements are bound within the crystalline lattices of primary and secondary minerals, which are not normally accessible. The average TE contents (e.g., Cr, Ni, and

Pb) in sediments from the Xisha Islands were lower than the sediment quality guidelines, with a small RSD. Thus, these elements may be derived from a natural source, as TEs from natural minerals tend to be retained in residual sediments and exhibit low mobility (Stumm and Morgan, 1995). Strontium exhibited comparatively low BAF and BSAF values in seagrass and algae, but a high total content, which corresponds to the results of previous reports (Malea and Kavrekidis 2014). Because Sr is an alkali metal that participates during metabolic processes with Ca (Moiseenko et al. 2008), it is typically enriched in carbonate environments containing aragonitic organisms, such as carbonate-accumulating seagrass (e.g., *Thalassia testudinum*) (Enriquez and Schubert, 2014).

TE transfer to green turtle eggshells

Zn, Cu, and Se are essential for the normal growth and metabolism of living cells and are present at higher quantities in female sea turtle blood compared to nonessential elements (Cd, Hg, and Pb) (Sinaei and Bolouki, 2017). These essential elements (with higher maternal transfer rates) are easily transferred from female sea turtles to their eggs (Páez-Osuna et al. 2010). In this study, we found that the contents of essential elements (Cu, Zn, and Se) in eggshells were relatively higher than those in seagrass/algae; especially, the Cu content in eggshells was approximately 10x higher than that in forage. Generally, the Cu content has been reported to be higher in eggshells than in albumen and yolk (Páez-Osuna et al. 2010). Additionally, we previously demonstrated that Cu concentrations in green turtle eggshells from Xisha islands exceeded the toxic reference value for bird eggs and Se concentrations were between the worst- and best-case scenario hazard quotients (Jian et al. 2021). Notably, females may accumulate Cu and Se through forage, and eggshell formation is a pathway for the discharge of these elements.

Although As may be an essential element for life, no definitive data are available on its importance for biological systems (Kunito et al. 2008), and Cd is a nonessential element. Despite the high burden of As and Cd in seagrass and macroalgae, As and Cd contents were found to be low in green turtle eggshells in this study. Marine algae contain As primarily as arsenosugars, whereas the livers of green turtles primarily contain As as arsenobetaine (AB) and a small percentage of dimethylarsinic acid (DMA(V)) (Kunito et al. 2008). Both AB and DMA(V), which are converted from arsenosugars, have short biological half-lives (Kunito et al. 2008), and the lack of an effective trophic As transfer was observed in other lower trophic-level marine animals (Signa et al. 2017). Although Cd can be efficiently transferred and biomagnified in invertebrates at contaminated sites (Signa et al. 2017), little bioaccumulation of Cd was observed in green turtle eggshells in this study. Generally, Cd accumulates in the kidneys and livers of sea turtles (Cortés-Gómez et al. 2017). Studies have shown that even a high Cd in the diet results in low blood concentrations in freshwater turtles (*Trachemys scripta elegans*) (Guirlet and Das, 2012). Moreover, egg-laying may not be the main method for sea turtles to exude harmful elements, such as Cd (Páez-Osuna et al., 2010), which suggests that Cd toxicity in green turtles may be prevented by excretory mechanisms. Thus, eggshells may be a poor bioindicator for some harmful elements with low maternal transfer rates (e.g., Cd).

No significant bioaccumulation of Pb and Hg was observed in the eggshells of green turtles in this study. The average content of Pb in eggshells ($0.09 \mu\text{g}\cdot\text{g}^{-1}$, wet.wt, Jian et al. 2021) was less than the worst-case scenario hazard quotients ($0.12 \mu\text{g}\cdot\text{g}^{-1}$, wet.wt). There are two explanations for the low values. First, the Pb and Hg contents in the seawater and sediments were low. Second, the concentrations of TEs in seagrass and algae reflect TEs availability in the surrounding media (Rai et al. 1981). In particular, the BAF and BSAF of forage for Hg and Pb were < 1 , and therefore, we assumed that Pb and Hg were unavailable in the environment.

Conclusions

In this study, we described the concentrations of 12 TEs in the seawater, coral sands, seagrass, and algae from the Xisha Islands, South China Sea. Overall, the TEs in seawater and coral sands had low ecological risks. The concentrations of most TEs in seagrass and algae were considerably lower than the global mean values and those at other sea turtle habitats, whereas the Cu, Zn, As, Se, and Cd concentrations were similar. The concentrations of Hg, Pb, and Cr in environmental media were higher than those in seagrass/algae and eggshells, indicating that their biological transfer

was limited, which appears to be influenced by the metal-specific bioavailability in coral reef ecosystems. In particular, we found that the bioaccumulation of TEs, especially the essential elements (e.g., Cu, Se, and Zn) in green turtle eggshells, corresponded well with the accumulation of these elements in forage, indicating that these elements are effectively transferred through forage, and then further from the mother to eggshells. Although the nonessential elements Cd and As accumulated at high concentrations in seagrass and algae, they were present at low levels in green turtle eggshells. Thus, eggshells may be a poor bioindicator for the exposure of female green turtles to this toxic element. As a non-invasive indicator, eggshells are good bioindicators for the exposure of female turtles to essential elements (e.g., Cu, Se, and Zn), while they cannot reflect the exposure to some toxic elements (e.g., As and Cd) via forage because the maternal transfer rates of these elements are limited, and bioaccumulation may be tissue- or organ-specific (e.g., liver or kidney). Further investigation is needed to explore these relationships using multiple approaches.

Author contribution Li Jian and Ting Zhang: conceptualization, methodology, investigation, formal analysis, writing-original draft. Liu Lin: formal analysis, writing-review and editing. Jinfang Xiong: investigation, methodology. Haitao Shi: conceptualization, formal analysis. Jichao Wang: conceptualization, writing-review and editing.

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Data availability Data will be made available on request.

Declarations

Ethics approval Not applicable.

Consent to participate All authors consent.

Consent for publication All authors consent when it is published.

Competing interests The authors declare no competing interests.

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