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Assessment of trace metal concentrations in Indian Ocean silky sharks *Carcharhinus falciformis* and their toxicological concerns

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ABSTRACT

This study assessed the concentrations of nine trace metals from juvenile *C. falciformis* caught from Indian Ocean. This study also discussed the metal pollution index (MPI) and bioconcentration factor (BCF) of each element, and their correlations. Further, the potential health risks of consuming shark muscles (THI) were evaluated. Results showed the mean concentrations of 9 elements as follows: Cu (0.36 ± 0.17), Zn (5.19 ± 16.6), Pb (0.12 ± 0.23), Cd (0.17 ± 0.21), Cr (0.57 ± 1.61), Ni (0.086 ± 0.51), As (1.36 ± 0.83), Co (0.000073 ± 0.0074), and V (0.0024 ± 0.0094) mg/kg ww. The BCF values of the elements were higher than 1, with Co and V being the lowest indicating their bioaccumulative behavior. Correlation analysis showed MPI to be highly correlated with Cu, indicating its greater contributions to the total pollution load. Principal components analysis explained 81.0% of the variability in biometric characteristics and metal concentrations. Health risk assessment for consuming shark muscle in Taiwanese male and female adults suggests potential chronic non-carcinogenic health hazards.

1. Introduction

Silky shark *Carcharhinus falciformis* is one of the most common pelagic sharks in tropical waters, continental and island shelves, continental slopes, and offshore waters worldwide. In addition, silky sharks are the most common bycatch shark in tropical tuna longline and purse seine fisheries (Amandé et al., 2008). Globally, *C. falciformis* is endangered (Bonfil et al., 2009), while it is considered vulnerable in the east-central and southeastern Pacific regions. They are a member of the requiem or gray sharks of the family Carcharhinidae which is one of the largest in the genus—reaching up to 330 cm in total length (Bonfil et al., 2009). Adult *C. falciformis* are piscivorous (Compagno, 1984), with juveniles feeding on pelagic fishes and crustaceans (Filmalter et al., 2017), and form large feeding aggregations when food is plentiful (Bonfil, 2008). *C. falciformis* has become the focus of many biological monitoring studies due to its importance in fisheries, with the increasing trend of global shark catches. *C. falciformis* is by far the most caught species, representing up to 90% of the elasmobranch bycatch worldwide (Gilman, 2011). Curnick et al. (2020) noted that over 56% of the species caught by longlines and purse seines were juveniles. With *C. falciformis* exhibiting diel vertical migrations and oscillatory diving behavior, spending more than 99% of their time in the top 100 m, and diving to the depths of greater than 300 m, they directly overlap with the typical deployment areas of purse seine and longlines in the Indian Ocean (Curnick et al., 2020).

Due to the bioaccumulation potential of heavy metals, they could be transferred through the food chain where large predatory fish like tuna, shark, and swordfish accumulate the highest concentrations (Burger and Gochfeld, 2011; Domingo et al., 2006). Notably, some metals biomagnify and lead to a critical accumulation in top predators such as sharks (Storelli, 2008; Adel et al., 2016). In recent years, marine pollution has become an issue of great concern (Anbuselvan et al., 2018; Sanjaigandhi et al., 2020). There is a range of persistent chemical contaminants such as heavy metals, ubiquitously distributed in marine environments and detected in a wide variety of marine organisms (Chiuchiolo et al., 2004). Ghani et al. (2012) reported that arsenic and vanadium contamination in sediments of Mediterranean coast showed an average of 29.90 ± 19.21 and 184.81 ± 134.42 mg/kg, respectively, mainly attributed from offshore oil fields and industrial wastes. These pollutants released in the marine environment could be magnified through the food chain e.g., from water/sediments–plankton–small pelagics–higher predators (Chouvelon et al., 2019; Albarico, et al., in press). Therefore, sharks being an apex ocean predator are highly susceptible to heavy metal bioaccumulation (Adel et al., 2016; Kim et al.,

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Table 1
Biometric characteristics and trace metal concentrations of *C. falciformis* caught from the Indian Ocean.

	Sex	TL	Weight	Condition factor	Cu	Zn	Pb	Cd	Cr	Ni	As	Co	V	MPI
		(cm)	(kg)	(*K*)	(mg/kg wet weight)									
S1	F	106	8.0	6.7	0.94	13.4	0.43	0.79	1.89	0.25	3.30	0.0044	0.025	1.31
S2	F	107	8.0	6.5	0.61	12.7	0.22	0.86	0.84	0.20	3.33	0.0024	0.026	0.97
S3	M	163	36	7.8	0.36	5.2	0.12	0.83	4.27	0.79	1.36	0.0072	0.027	0.98
S4	M	126	19	9.2	0.71	18.1	0.22	0.79	0.90	0.32	4.23	0.0066	0.021	1.15
S5	–	–	–	–	0.41	5.7	0.15	0.92	1.58	0.30	2.23	0.0031	0.019	0.86
S6	F	190	–	–	0.52	13.3	0.16	0.81	0.64	0.09	1.72	0.0001	0.015	0.70
S7	M	70	2.5	7.6	0.74	16.1	0.31	0.95	1.74	0.19	2.00	0.0061	0.019	1.13
S8	M	130	25	11.0	0.50	21.7	0.16	0.93	1.04	0.30	2.54	0.0042	0.016	1.03
S9	M	125	–	–	0.68	21.3	0.17	0.91	1.92	0.15	3.63	0.0028	0.017	1.14
S10	F	162	–	–	0.61	12.8	0.14	0.78	0.57	0.13	2.52	0.0018	0.017	0.77
S11	F	162	29	6.4	0.69	14.1	0.28	0.83	1.14	0.21	1.36	0.0091	0.002	0.96
S12	M	70	2.5	7.6	0.58	32.1	0.14	0.97	7.53	2.40	3.33	0.0303	0.002	2.05
S13	M	130	25	11.0	0.42	22.7	0.18	0.59	1.82	0.12	1.74	0.0058	0.018	0.87
S14	M	125	–	–	0.65	63.6	1.11	0.17	0.86	0.30	2.18	0.0022	0.047	1.24
S15	M	162	–	–	0.82	45.0	0.27	0.82	1.16	0.25	2.53	0.0049	0.023	1.29
S16	F	162	29	6.4	0.82	47.3	0.15	0.73	0.57	0.29	2.02	0.0084	0.016	1.05
S17	M	147	25	7.5	0.81	36.8	0.23	0.85	2.90	0.17	2.31	0.0061	0.024	1.32
S18	M	108	12	9.4	0.54	19.5	0.14	1.01	1.14	0.14	1.92	0.0016	0.030	0.90
S19	M	101	7.0	6.8	0.96	56.2	0.59	1.28	1.72	0.21	2.90	0.012	0.021	1.70
S20	M	130	17	7.5	0.65	20.7	0.23	0.92	2.39	0.69	3.93	0.023	0.014	1.52
Min					0.36	5.2	0.12	0.17	0.57	0.09	1.36	0.0001	0.002	
Max					0.96	63.6	1.11	1.28	7.53	2.40	4.23	0.030	0.047	
Ave					0.65	24.9	0.27	0.84	1.83	0.38	2.55	0.0071	0.020	
SD					0.17	16.6	0.23	0.21	1.61	0.51	0.83	0.0074	0.009	

–: Data is not available; NA: Not detected; Min: minimum; Max: maximum; Ave: mean values; SD: standard deviations; MPI: including seven toxic elements (Cu, Zn, Pb, Cd, Cr, Ni and As).

2019).

There have been a few studies on the trace metal concentrations and bioaccumulation in *C. falciformis*. Three of the literatures focused on mercury concentrations from *C. falciformis* caught along Mexico (Terrazas-López et al., 2016; O'Bryhim et al., 2017; Rodríguez-Gutiérrez et al., 2020). Similarly, a recent study in Mexican waters reported the bioaccumulation and biomagnification of various metals (Hg, Pb, Cd, Cu, As, Se, Zn, Cr, V, Fe, Ni, Co, and Mn) from shark muscle tissues (Álvaro-Berlanga et al., 2021). However, there were no studies in the Indian Ocean region which motivated this research to be undertaken. This study aimed to (1) describe the concentrations of heavy metals from *C. falciformis* muscle tissue, (2) determine the correlations of heavy metals, and (3) assess heavy metal bioaccumulation relative to that of seawater (background). The results of this study provide a baseline on the heavy metal concentrations in the muscle tissue of *C. falciformis* caught from Indian Ocean region.

2. Materials and methods

2.1. Study area and sample collection

Muscle tissues from the caudal peduncle (Fig. S1) were collected from juvenile *C. falciformis*; caught from the Indian Ocean using longlines between 2015 and 2018 ($n = 20$). Basic information on *C. falciformis* include sex, total length, and body weight (Table 1). Biological data from S5 was missing; therefore, although metal concentrations were measured, data from this individual was not used in the correlation analysis between biological parameters and metal contents. The total length (TL) ranged between 70 and 190 cm. The weight ranged between 2.5 and 36 kg. The condition factor (K) was also measured in this study.

2.2. Sample preparation and determination of elements

Total concentrations of nine heavy metals (Cu, Zn, Pb, Cd, Cr, Ni, As, Co and V) were analyzed using inductively coupled plasma mass spectrometry (ICP-MS; Thermo Fisher Scientific iCAP RQ ICP-MS, USA). Approximately 0.050 g of homogenized dried shark muscle was digested by adding 10 mL HNO_3 and heating at 95 °C for 4 h. Digested samples were cooled at room temperature and diluted with double distilled water; and the dilution factors were calculated based on the dry sample weight. The ICP-MS analysis was carried out in KED mode. This method provided better accuracy and precision, and the trace elements showed definite differences. Blank was carried out for instrument calibration. The accuracy and precision of the results were checked using standard reference materials (DORM-4) supplied from the National Research Council of Canada (CNRC). The certified reference material DORM-4 (Dogfish muscle reference material, NRC Canada) was used to guarantee method accuracy. The observed values are presented in Table S1. All metal results were expressed in wet weight concentrations.

2.3. Data analyses

Statistical analysis was conducted using Microsoft Excel 2019 and SPSS Statistics (SPSS software 20.0). Data were summarized as min, max, and mean, along with standard deviations. Allometric growth model was conducted to describe the length-weight relationship of *C. falciformis* with the formula: $W = aL^b$, where W is the body weight, L is the total length, a is the intercept constant, and b is the length-weight relationship index (Le Cren, 1951). Condition factor was calculated using the formula: $K = W/L^b$ to evaluate the biological and physiological condition (Devaraj, 1973; Acharya and Dwivedi, 1985; Sharma et al., 2005). The metal pollution index (MPI) which is a toxic element content index was used to compare the metal loads in *C. falciformis* muscles. MPI was calculated using the following formula (Usero et al., 1997): $MPI = (cf_1 \times cf_2 \times cf_3 \times \ldots \times cf_n)^{1/n}$, where cf_n is the concentration of metal n in the examined tissue (mg/g dry weight). The bioconcentration factor (BCF) is defined as the net result of the absorption, distribution, and elimination of a substance in any organism after exposure via water. The BCF was also used to evaluate the element accumulation capability of *C. falciformis* exposed to contaminated water. BCF was calculated using the equation: $BCF = C_s/C_w$, where C_s and C_w denote the concentrations of elements in the muscular tissues of *C. falciformis* (mg/kg wet weight) and seawater (mg/L), respectively (Ju et al., 2021). Because the concentration of elements in seawater usually remains within a narrow

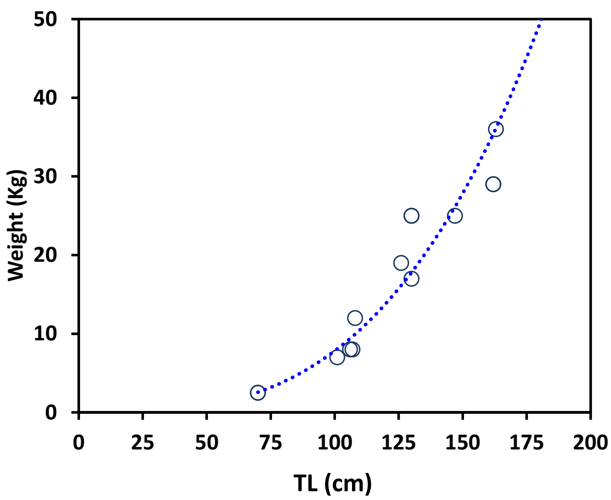

Fig. 1. Relationship between total length (L) and body weight (W) of *Carcharhinus falciformis*.

range, the difference in the element concentration between inshore seawater and ocean seawater is usually less than one order of magnitude (Lau et al., 1998). Thus, the seawater metal concentrations were based on the information given by Neff (2002). The concentrations used for each element were: Cu (0.35), Zn (0.12), Pb (0.1), Cd (0.1), Cr (0.55), Ni (1.0), As (3.0), Co (0.19), and V (1.9) mg/L. Spearman's rank correlation coefficient (r) was used to evaluate the correlations between various parameters. Shark characteristics were presented using descriptive statistics. Principal components analysis (PCA) were performed for trace metals using Varimax Method with a component rotation having an eigenvalue > 1.

2.4. Human health risk assessments

The potential human health risk of consuming *C. falciformis* muscles was measured in terms of the HQ and THI. The *HQ* is a proportion of estimated daily intake (EDI, μg/kg/day) to chronic oral reference dose (*RfD*, μg/kg/day); HQ = EDI/RfD. The EDI was estimated according to the element concentration (CE, μg/g wet weight), the daily consumption (DCfish, g/day) of *C. falciformis* muscles, and the mean body weight (BW, kg) of Taiwanese adults; EDI = (CE × *DCfish*) / BW was determined according to the data of the Integrated Risk Information System (https://www.epa.gov/iris) developed by the USEPA in 2020 (USEPA, 2020), except the doses of Pb and Co, which were determined according to the studies reported by Orisakwe et al. (2017) and Finley et al. (2012). In this study, the dataset followed our previous report (Ju et al., 2021).

The THI is a summation of HQ of all metal elements. When the HQ or THI is less than 1.0, consuming *C. falciformis* muscles will not pose a significant non-carcinogenic risk. Possible health risks increase as HQ or THI increases. When the THI is greater than 10, the non-carcinogenic risk is considered extremely high (Ju et al., 2021).

3. Results and discussion

3.1. Biometric characteristics of C. falciformis

Biometric characteristics such as length and weight are important parameters in shark research. Joung et al. (2008) estimated that 50% of silky sharks from northern Taiwan waters mature at 212.5 cm total length, using the logistic curve describing the relationship between total length and the proportion of maturity in each length interval. The latter results suggest that *C. falciformis* selected for trace metal analysis in this study were juveniles. To clarify the effect of metal bioaccumulation on the physiological condition of *C. falciformis*, the condition factor (*K*) was used. The condition factor can be estimated using weight (W) and length (L) of the *C. falciformis* following Sharma et al. (2005), where W is the shark bodyweight in grams, L is the total length in cm, and b is the exponent of the length-weight relationship, which can be obtained from the allometric growth model described by Kumar et al. (2018) where a is the constant of intercept. Fig. 1 shows the relationship between total length (L) and body weight (W) for *C. falciformis*. The R^2 shows a higher value of 0.9545. The model $W = aTL^b$ defined, resulted to 4.26×10^{-6}, and b value of 3.132. The condition factors (K) are presented in Table 1. The level of the physiological condition K ranged between 6.4 and 11.0.

3.2. Trace element concentrations in C. falciformis

The trace metals, Cu, Zn, Pb, Cd, Cr, Ni, As, Co and V were determined using ICP-MS. The values for trace metals measured in the muscle are illustrated in Table 1. The Cu concentration ranged between 0.36 and 0.96 mg/kg, with an average concentration of 0.65 ± 0.17 mg/kg. These concentrations were far lower compared to *C. falciformis* in Mexico having 26.8 ± 27.9 mg/kg (Álvaro-Berlanga et al., 2021), and other species like *C. obscurus* and *Isurus oxyrinchus* (Table 2). The regulatory maximum limits indicated for the copper ion is 5 mg/day (SCF, 2006) suggesting that the metal concentrations were within the allowable levels. In this study, the Zn concentrations ranged from 5.2 to 63.6 mg/kg. These results were correspondingly lower compared to Zn concentrations (168.3 ± 101.53 mg/kg, *n* = 30) in *C. falciformis* caught from Mexico (Álvaro-Berlanga et al., 2021), but are higher than other shark species worldwide (Table 2). Similarly, Pb concentrations (0.12–1.11 mg/kg) were lower than *C. falciformis* in Mexico, but higher than other species. However, other shark species like *G. galeus* in Azores, Portugal (Torres et al., 2016), and *C. obscurus* and *C. umbratile* in Korea (Kim

Table 2
Comparison for metal concentrations (mg/kg ww) among different shark worldwide.

	C. falciformis	*C. falciformis*	*C. obscurus*	*C. umbratile*	*G. galeus*	*I. oxyrinchus*	*I. oxyrinchus*	*M. manazo*	*P. glauca*	*T. scyllium*
Area	Indian Ocean	Mexico	Korea	Korea	Portugal	Korea		Korea	Portugal	Korea
Cu	0.65 ± 0.17	26.8 ± 27.9	2.73 ± 3.67	0.57	0.24 ± 0.01	4.45 ± 4.67	0.38 ± 011	0.43 ± 0.35	0.70 ± 0.21	0.18 ± 0.08
Zn	24.9 ± 16.6	168.3 ± 101.5	8.16 ± 4.01	13.56	6.24 ± 0.22	3.23 ± 1.60	4.48 ± 0.43	2.09 ± 0.08	3.95 ± 0.15	3.41 ± 0.62
Pb	0.27 ± 0.23	4.08 ± 6.57	0.08 ± 0.06	0.07	–	0.06 ± 0.02	–	0.01 ± 0.01	–	0.02 ± 0.01
Cd	0.84 ± 0.21	1.44 ± 1.50	–	–	–	–	–	–	–	–
Cr	1.83 ± 1.61	3.46 ± 5.87	0.37 ± 0.53	0.16	1.28 ± 0.08	–	1.06 ± 0.17	0.07 ± 0.08	0.50 ± 0.03	0.31 ± 0.40
Ni	0.38 ± 0.51	3.58 ± 8.28	–	–	–	–	–	–	–	–
As	2.55 ± 0.83	152.2 ± 87.7	7.85 ± 0.24	8.09	20.68 ± 1.52	2.06 ± 1.07	1.71 ± 0.43	17.59 ± 3.63	10.02 ± 0.69	8.32 ± 0.37
Co	0.0071 ± 0.0074	0.19 ± 0.19	–	–	–	–	–	–	–	–
V	0.02 ± 0.009	1.2 ± 1.14	–	–	–	–	–	–	–	–
Ref.	This study	Álvaro-Berlanga et al. (2021)	Kim et al. (2019)	Kim et al. (2019)	Torres et al. (2016)	Kim et al. (2019)	Hussey et al. (2017)	Kim et al. (2019)	Torres et al. (2016)	Kim et al. (2019)

–: Data is not available.

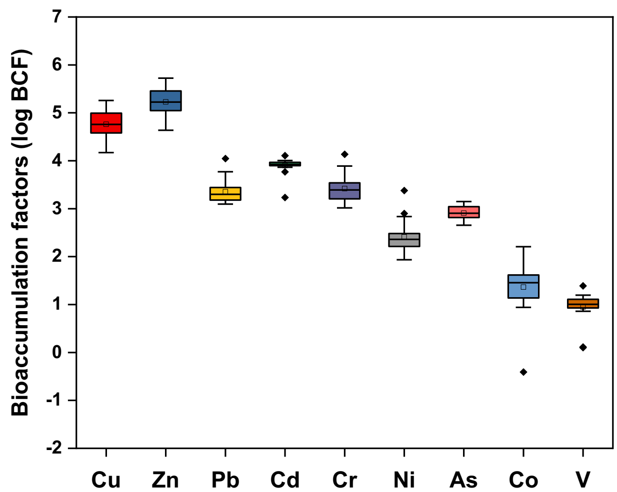

Fig. 2. Box plots showing the distributions of bioaccumulation factors of metals in *C. falciformis* muscular tissues.

et al., 2019) showed slightly comparable concentrations (Table 2). These concentrations were over the limits recommended by the UN committee for Pb concentrations (0.3 mg/kg), hence immediate attention is necessary. The Cd concentration (0.84 ± 0.21 mg/kg) showed similarly lower concentrations than its conspecifics in Mexico (Álvaro-Berlanga et al., 2021). However, Reátegui-Quispe and Pariona-Velarde (2019) observed that concentrations of Cd in blue shark *Prionace glauca* were 0.01–0.04 mg/kg which is lower than this study. The Cr concentration ranged between 0.57 and 7.53 mg/kg which is higher than the rest of the world (Table 2). Ni showed comparable concentrations with Pb and Cd in this study (0.38 ± 0.51 mg/kg). As displayed concentrations from 1.36 to 4.23 mg/kg with an average of 2.55 ± 0.83 mg/kg. In the same species, *C. falciformis*, Álvaro-Berlanga et al. (2021) showed a significantly higher concentration of 152.2 ± 87.7 mg/kg for As. Similarly, As concentrations in this study showed lower values than the rest of the shark species worldwide (Table 2). As compounds are mostly soluble in water, hence, As is more likely to be present in seafood. According to Food Safety Focus (2007), the level of As can range from 1 to 10 mg/kg in fish and up to 100 mg/kg in bottom feeders and shellfish. Co and V exhibited the lowest concentrations among the nine trace metals (Co: 0.0071 ± 0.0074; V: 0.02 ± 0.009 mg/kg). Generally, the differences on metal concentrations in sharks observed worldwide could be attributed to the ontogeny, trophic levels, and different foraging areas (Bonfil, 2008; Newman et al., 2012). The confidence intervals of simple linear regression for different metal concentrations in the muscular tissues of *C. falciformis* are found in Fig. S2. The data point of Cu and As were all in the confidence intervals (95%) of nine elements. The highest concentration of Cr, Co, Pb, Ni, and V was out of 95% of confidence intervals. On the other hand, the lowest concentration of Cd, Cr, Co, Pb, and Ni were out of 95% of the confidence intervals range. The Zn, Cr, Co, and Pb were showing in the same pattern.

3.3. Elemental bioaccumulation in sharks (MPI and BCF)

Sharks are used as human food (meat and soup in Asia, Europe, USA, Africa, and Australia), in the industries (skin, shark liver oil), and for medicinal purposes (vitamin A) (Kibria and Haroon, 2015). Thus, the presence of metal pollution in sharks could compromise human health and product quality. Metal pollution index (MPI) was used to compare the metal loads of Cu, Zn, Pb, Cd, and As in *C. falciformis* (Usero et al., 1997). MPI ranged between 0.70 and 2.05, with an average of 1.15 ± 0.33. MPI was greatly contributed by Cr and Ni, which were defined as the MPI indicator in shark muscle. For the issue of food safety, the BCF was used to understand the metal accumulation in *C. falciformis*. When aquatic organisms absorb a substance at a rate higher than they can metabolize and eliminate, bioaccumulation occurs. Hence, toxic substances with longer biological half-life (e.g., trace metals) can pose a high chronic health risk due to bioaccumulation even if their concentrations in the water is extremely low (Ju et al., 2021). Estimates of BCF were shown in Fig. 2. The seawater metal information was cited from Neff (2002). Several studies discussed the environmental pollutant heavy metals in the Indian Ocean (van der Schyff et al., 2020; Tamele and Loureiro, 2020; Moffett and German, 2020). In this study, Zn and Cu have the highest contribution for trace metal accumulation in sharks. The log BCF value of elements was higher than 1 indicating the metal's bioaccumulative behavior (USEPA, 1991). Co and V were the lowest accumulation factor in the shark of the muscular tissues. Alarmingly, bioaccumulation factors for Zn > Cu > Cd > Cr > Pb > As > Ni were greater than 2 (BCF > Log 2.0) indicating significantly high metal accumulation.

3.4. Spearman and PCA analysis with relations between characteristics and trace elements in muscle tissue

The relationship between body length and weight and metal contents were evaluated. Several studies have reported metal concentrations associated with body size in sharks (Kim et al., 2019; Turoczy et al., 2000). Table 3 describes the correlation between the total length, weight, metal concentrations, and MPI. Levels of Cd in muscle decreased significantly ($p < 0.05$) with shark length; in the case of shark weight, the Co concentration increased significantly ($p < 0.05$). The condition factor (K) had no significant correlation with other metal elements. The concentration of Cd ($r = 0.263$, $p > 0.05$) and V ($r = 0.064$, $p > 0.05$) had no significant correlation with the MPI ($p > 0.05$), whereas the concentration of the other seven metal elements showed a significantly positive correlation with the MPI ($r = 0.490$–0.615, $p < 0.05$). Highly

Table 3
Spearman's rho coefficient matrix between metals in *C. falciformis* the muscular tissues.

	TL	Weight	K	Cu	Zn	Pb	Cd	Cr	Ni	As	Co	V
Weight	0.324											
K	−0.220	0.592**										
Cu	−0.096	−0.031	−0.146									
Zn	−0.048	0.004	0.037	0.470*								
Pb	−0.228	−0.120	−0.073	0.675**	0.378							
Cd	−0.527*	−0.052	0.359	−0.058	−0.026	−0.092						
Cr	−0.277	0.166	0.370	−0.022	0.061	0.037	0.408					
Ni	−0.175	0.228	0.140	−0.032	0.085	−0.036	0.124	0.288				
As	−0.414	−0.244	−0.030	0.330	0.173	0.177	0.217	0.149	0.335			
Co	−0.054	0.516*	0.370	0.328	0.292	0.159	0.225	0.514*	0.568**	0.169		
V	−0.195	−0.046	0.057	0.082	−0.022	0.202	−0.108	0.005	−0.029	−0.064	−0.363	
MPI	−0.343	0.011	0.147	0.624**	0.603**	0.495*	0.263	0.511*	0.490*	0.615**	0.593**	0.064

* Correlation is significant at the 0.01 level (2-tailed).
** Correlation is significant at the 0.05 level (2-tailed).

Table 4
Principal components estimated on the variable item of *C. falciformis*.

Variable	Principal components			
	PC1	PC2	PC3	PC4
TL (cm)	−0.202	−0.919	−0.112	−0.211
Weight (kg)	−0.111	−0.855	−0.278	−0.340
Condition factor (K)	−0.141	0.740	−0.058	−0.514
Cu	−0.187	0.122	0.846	0.171
Zn	0.230	−0.167	0.801	−0.034
Pb	−0.238	0.293	0.792	0.099
Cd	0.163	0.436	0.573	0.068
Cr	0.848	0.171	−0.187	0.077
Ni	0.927	0.152	−0.175	0.210
As	0.111	0.213	0.161	0.884
Co	0.917	0.069	0.156	0.204
V	−0.739	0.115	−0.138	0.224
Eigenvalue	3.8	3.1	1.8	1.1
Variance (%)	31.6	25.5	14.7	9.2
Cumulative variance (%)	31.6	57.1	71.8	81.0

Rotation method: Varimax with Kaiser normalization.
Bold = loadings > 0.65.

significant correlations between Cu and Pb were determined ($r = 0.675$, $p < 0.01$). MPI and Cu had the highest correlation compared to other metals, hence, Cu can be an indicator element for MPI. Spearman's rank correlation showed there was no correlation between TL and Cd concentrations ($p < 0.05$) with Blue sharks (*Prionace glauca*) (Castro-Rendon et al., 2021).

The results of PCA (varimax rotation) obtained a total of four principal components (PC1 to PC4), which can explain 81.0% of the biometric characteristics and the trace metal concentration variability (Table 4). PC1 can explain 31.6% of all variation, among which Cr, Ni, Co, and V have relatively high factor loadings (larger than 0.70) and Cr, Ni, and Co are positive. Demirezen and Uruc (2006) described that Ni and Cr are biological essential elements because they play an important role in a variety of enzymes and other cellular components. Kowalska et al. (2020) noted that cobalt is a component of vitamin B12 (cobalamin), a coenzyme that is essential in the formation of proteins, nucleic acids, and red blood corpuscles. Moreover, the authors mentioned that the largest amounts of cobalt are found in muscles, approx. 43%. The PC2 explained 25.5% of the variation, in which TL (cm), weight (kg), and condition factor (K) showed relatively high loadings, implying that PC2 represents the level of biometric characteristics content in *C. falciformis*. The PC3 explained 14.7% of the variation, in which Cu, Zn, and Pb showed a relatively high positive load, PC3 represents the toxic elements in *C. falciformis* muscle tissue. The PC4 describes 9.2% of the variation. The metal As have relatively high loading. Therefore, the PC4 represents the concentration of As, which for marine animals increases with higher trophic levels, but may also be affected by dietary intake and the ability to absorb, metabolize, and accumulate As in their bodies (Ju et al., 2021). Fig. 3 shows the scatter diagram of PC1 and PC2 to PC4 scores for all 20 shark samples. Metals like Cr, Ni, Co, and V have relatively high factor loadings observed in this study. In Fig. 3A, shark 12, shark 14 were observed to have higher Cr and V concentrations. According to the distribution of PC1 and PC3 scores (Fig. 3B), Cu, Zn, and Pb showed relatively high positive load, while the S4, S12, and S14 can be observed with the higher concentrations of these three elements. The PC4 score of the S7, S12, and S14 have relatively higher As concentrations than that of other sharks (Fig. 3C).

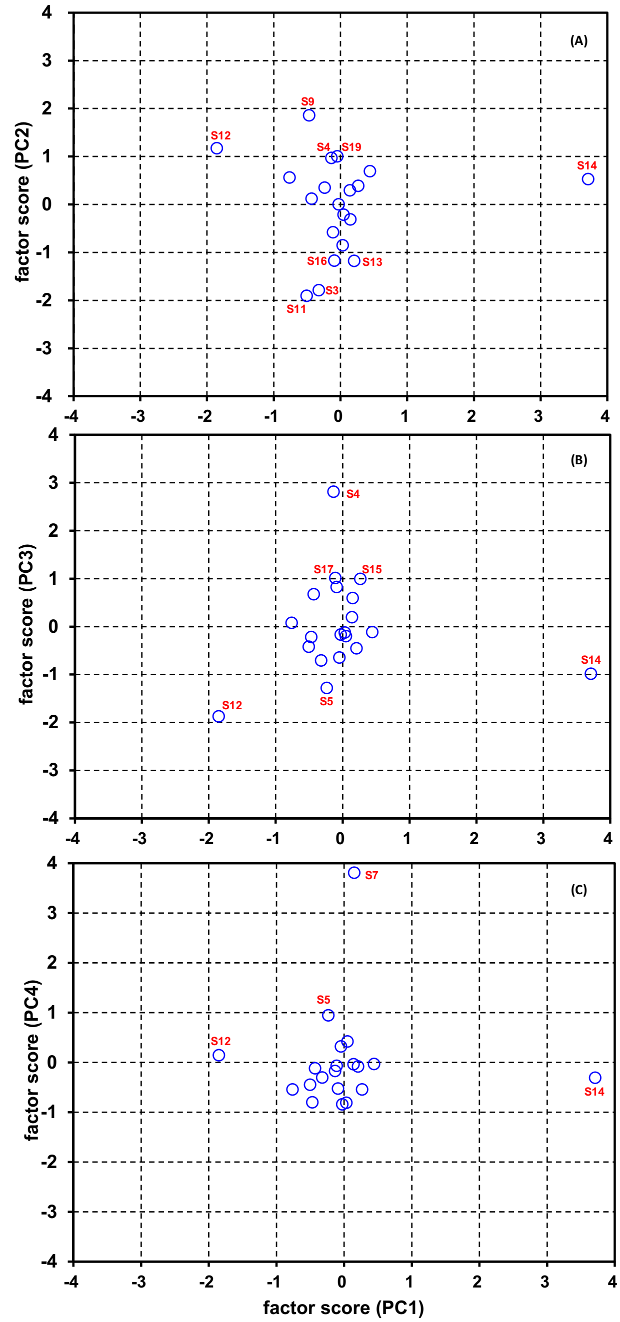

Fig. 3. The principal components analysis of metals in *C. falciformis*.

3.5. Estimation of human health risks by consumption

Shark meat consumption is a global trend (Dent and Clarke, 2015), especially in some countries in Asia, which could be related to cultural or traditional reasons rather than for real health benefits (Dell'Apa et al., 2014). Table 5 presents the HQ and THI values estimated based on the mean concentrations of various elements in the muscular tissues of silky sharks and the mean consumption of saltwater fishes by Taiwanese male and female adults. The content of inorganic As in fish usually accounts for 0.5% to 10% of total As; therefore, in this study, the HQ of As was estimated by converting 6% of the total As into inorganic As values (Rahman et al., 2012; Copat et al., 2013; Boldrocchi et al., 2019). The HQ of all estimated elements was higher than 1, with As found to be the main contributor (85% of total HQ in Best Case; 56% of total HQ in Worst Case) in this study. Moreover, THI was greater than 1 in both male and female adults; and the health risk faced by male adults (Best Case: THI = 4.22; Worst Case: THI = 20) were slightly lower than that of females (Best Case: THI = 8.01; Worst Case: THI = 38). The THI ratio of Worst/ Best Case scenarios were almost at 5. This implies that long-term consumption of sharks by adults may cause chronic non-carcinogenic health hazards. The accumulation of As in fish muscles and their high toxicity to humans are often considered noteworthy elements in health

Table 5
Risk assessment of consuming shark for Taiwanese male and female adults.

Trace element	C_m (Min)	C_m (Max)	RfD[a]	Hazard quotient (HQ) Worst case		Hazard quotient (HQ) Best case	
	(μg/g)	(μg/g)	(μg/kg/day)	Male	Female	Male	Female
Cu	0.36	0.96	40	0.019	0.036	0.01	0.014
Zn	5.19	63.55	300	0.17	0.32	0.01	0.026
Pb	0.12	1.11	3.5	0.25	0.48	0.03	0.054
Cd	0.17	1.28	1	1.0	1.9	0.14	0.259
Cr	0.57	7.53	3	2.0	3.8	0.15	0.289
Ni	0.09	2.40	20	0.1	0.2	0.00	0.007
As[b]	1.36	4.23	0.3	11.2	21.3	3.61	6.845
Co	0.00	0.03	30	0.0008	0.0015	0.00	0.000
V	0.00	0.05	0.007	5.3	10.1	0.27	0.518
Total hazard index (THI)				20	38	4	8

[a] RfD adopted from USEPA (2020), and Pb and Co from Orisakwe et al. (2017) and Finley et al. (2012).
[b] The value represents the As inorganic form that were calculated taking into account 6% of total As (Boldrocchi et al., 2019).

risk assessment (Ju et al., 2021). But usually, the risk is lesser than other metals because of the dominance of inert organic forms of As. However, V (HQ = 0.2738 and 0.5193) is also a remarkable element in this study, even though it is an essential element for humans and is quickly excreted through urine and bile after absorption in the human body (Bhattacharya et al., 2016). In other studies, the V values were observed mainly in the central area of the Adriatic Sea for anchovy (V 89.9 μg/kg ww), red mullet (V 79.1 μg/kg ww), and mackerel (V 43.5 μg/kg ww), which is estimated to contribute 11–34% of the daily vanadium ingestion with fish, calculated for the population of the Adriatic coast (Sepe et al., 2003). Based on these results of best or the worse case, silky sharks caught from the study sites could pose potential chronic non-carcinogenic health hazards to male and female Taiwanese adults (The worst case of THI is greater than 10, the non-carcinogenic risk is extremely high).

4. Conclusions

This study assessed the concentrations of nine trace metals in *C. falciformis* caught from the Indian Ocean. The juvenile *C. falciformis* in this study were selected for trace metal analysis. To clarify the effect of metal bioaccumulation on the physiological condition of *C. falciformis*, the condition factor (K) was used. The trace elements concentrations among shark species can be affected by ontogeny, trophic levels, and foraging areas, explaining the differences in the trace metal levels in shark specimens. The MPI was much higher in Cr and Ni than other metals (Cu, Zn, Pb, Cd, and As) which were defined as an MPI indicator in shark muscle. The BCF value of elements was higher than 1. The Co and V were the lowest accumulation factor in the shark of the muscular tissues. MPI and Cu were highly correlated compared to other metals, making Cu a potential indicator element for MPI. The PCA can explain 81.0% of the biometric characteristics and the trace metal concentration variability. For the human health risks by consumption, the Taiwanese male and female adults who regularly eat shark muscles are exposed to potential chronic non-carcinogenic health hazards.

CRediT authorship contribution statement

Ming-Huang Wang: Conceptualization, Methodology, Data curation, Writing – original draft, Formal analysis, Visualization. **Chiu-Wen Chen:** Resources, Supervision, Project administration. **Chih-Feng Chen:** Data curation, Formal analysis, Visualization, Writing – review & editing. **Wen-Pei Tsai:** Investigation, Validation, Methodology. **Cheng-Di Dong:** Writing – review & editing, Resources, Supervision, Funding acquisition.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.marpolbul.2022.113571.

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