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ASSOCIATION OF TOTAL MERCURY AND CADMIUM CONTENT WITH CAPTURE LOCATION AND FISH SIZE OF SWORDFISH (*Xiphias gladius*); INDIAN OCEAN

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ABSTRACT

Mercury (Hg) and cadmium (Cd) are non-essential trace elements that transfer through the trophic chain which ultimately bio-accumulate and biomagnify in the upper trophic level. The total Hg (THg) and Cd levels of the muscle tissues of swordfish, *Xiphias gladius* caught in three different areas in the Indian Ocean around Sri Lanka were determined. THg and Cd levels were <0.07-4.30 mg/kg and <0.006-0.180 mg/kg, (wet weight basis) respectively. Of the analyzed samples, 13.3% fish were over 1 mg/kg for THg while not any single sample exceeded 0.30 mg/kg for Cd which is EU, and FDA action limits. The results indicate that the catching locations do not govern the THg and Cd levels and it showed a weak positive relationship between the length and weight of the fish.

Keywords: Atomic Absorption Spectrometer, Catching location, Length-Weight relationship, Indian Ocean

Introduction

Contamination of the marine ecosystem by non-essential trace metals is a worldwide problem because they can be toxic even at a very low concentration (Le Croizier *et al.*, 2018). Mercury (Hg) and cadmium (Cd) are well known global environmental pollutants which occur naturally and also released to the marine environment by anthropogenic activities such as coal combustion, mining industry by-product, agriculture fertilizer and waste incineration (Jinadasa *et al.*, 2013, Nicklisch *et al.*, 2017).

Mercury exists in the environment in several chemical forms including elemental Hg (Hg°), inorganic Hg (Hg^+ and Hg^{2+}) and organic Hg ($MeHg^+$, $EtHg^+$, $PeHg^+$ *etc.*) (Zhu *et al.*, 2017). The adsorption, transport, bioaccumulation, metabolism, and toxicity of Hg are governed by its speciation form (Arroyo-Abad *et al.*, 2016). All forms of Hg can bio-accumulate and biomagnify through the food chain; $MeHg^+$ has a greater ability than other forms (Jia *et al.*, 2013). The microorganisms convert elemental Hg to $MeHg^+$ through the methylation process in the marine environment (Morrissey *et al.*, 2005). Methylmercury is a known neurotoxin and currently considers that fish is the main path for the Hg exposure to human. In the pelagic, highly migratory, slow growing and apex predator fish such as swordfish (SF), yellowfin tuna and a shark, about 90% of total Hg (THg) are in $MeHg^+$ chemical form (Silvia *et al.*, 2010).

Cadmium occurs in the marine environment as cadmium chloride ($CdCl_2$) (Engel and Fowler, 1979). The International Agency for Research on Cancer (IARC) categorized Cd as a group 1 human carcinogen (Guan *et al.*, 2015). In addition, Cd toxicity is responsible for various impairment of organisms such as kidney disorder, osteoporosis, damages of the liver, the central and peripheral nervous systems (Pastorelli *et al.*, 2018, Al-Saleh and Abduljabbar, 2017). There is an evidence to support that Cd might be related to hypertension, stroke, and heart failure which is counter to the cardio defensive property of eating the fish (Guan *et al.*, 2015).

The Food and Agriculture Organization (FAO), European Union (EU) and Food and Drug Administration of United States (USFDA) regulations stipulate the maximum level for THg and Cd for SF as 1 mg/kg and 0.30 mg/kg respectively (Jinadasa *et al.*, 2014, Bosch *et al.*, 2016). The THg and Cd levels in fish depend on numerous factors including species, body size, sex, migratory biology, trophic position, foraging behavior and environmental factors such as pH, salinity, water temperature and dissolved oxygen (Guan *et al.*, 2015, Ray, 1984).

In the Indian Ocean, SF is primarily caught from the area off Somalia and from the southwest Indian Ocean by longline fisheries. Recent years, this fishery has moved towards off Sri Lanka (IOTC, 2014). Several studies carried out of SF have revealed that there is a relationship of THg and Cd with the body size (length and weight) (Jinadasa *et al.*, 2013, Mendez *et al.*, 2001, Kojadinovic *et al.*, 2007), however, few studies are reported about the relationship with capture location. Among the large pelagic fish species in the marine environment, SF is of primary importance, as they are the important export fish species from Sri Lanka (Jinadasa *et al.*, 2014). Therefore, it is appropriate to evaluate the THg and Cd content of SF with the capture location.

This paper reports, THg and Cd level of SF caught in three main areas in the Indian Ocean around Sri Lanka and the relationships of THg/Cd concentration with catching location, length, and weight.

Materials and Methods

Sample Collection

A total of 75 SF samples were collected from commercial seafood exporter during Jan-Dec 2017 caught from the Indian Ocean. The capture coordination (latitude and longitude), capture dates, vessel number were taken from fisherman logbook and confirmed from the satellite-based vessel monitoring system (VMS). Then the coordination was fed into the Google map software (online) for clustering. The total dressed weight (kg) and dressed length (cm) were recorded (without sword-like snout) and 250 g of muscle tissues were separated and transported to the laboratory under chilled condition.

Mercury and Cadmium Analysis

Analytical and general laboratory procedure was based on Jinadasa *et al.* (2014) with slight modification. Briefly, all glassware used was decontaminated 24 hrs with 10% HNO_3, washed with ultrapure water and dried properly. All the standards and reagents were prepared using ultrapure water (Barnstead, Easy pure LF system, Dubuque, USA). All the chemicals were analytical reagent grade or better (Sigma Aldrich, USA). The standard solutions of Hg and Cd at 1,000 mg/L (Fluka, Switzerland) were used for the construction of calibration curves. Approximately 1 g of homogenized samples were accurately weighed and pre-digested by treating with 10 mL of 65% (v/v) HNO_3 acid for 15 min at room temperature. Pre-digested samples were digested using a microwave accelerated system, MARS-6 (CEM, Matthews, USA). The digested samples were used to prepare 50.00 mL aqueous solutions. All the experiments were carried out in duplicate. Atomic absorption spectrophotometer (AAS) (Varian240 FS, Varian Inc., Mulgrave, Victoria, Australia) equipped with vapor generation accessory (Varian VGA 77) with closed end cell was used for determination of THg while AAS with Varian graphite tube atomizer (GTA-120) was used for determination of Cd.

The accuracy of the analytical procedure for THg and Cd was determined by analyzing certified quality control material (CQM), the samples (n=10) were analyzed in the same manner (canned fish offal, T/07243, and canned crab meat, T/07279QC from Food Analysis Performance Assessment Scheme, FAPAS, Sand Hutton, York, UK). The average field blank, derived from sample field blanks, and three times of its standard deviation were used to evaluate the limit of detection (LOD). The limit of quantification (LOQ) was 3 × LOD.

Data Analysis

All THg and Cd results were reported in a wet weight basis. To measure the correlation between THg and Cd content with length and weight of SF, linear regression analysis and Pearson correlation coefficient applied. To find out the metal content among the different locations, we used one-way analysis of variance (Anova) followed by Levene's test. All the data were analyzed using SPSS software version 17 (SPSS Inc., Illinois, United States).

Results and Discussion

The suitability of the method was evaluated in terms of their respective LOQ, recovery value using CQMs. As the standard operating procedure, the recoveries were maintained between 80-120% and the relative standard deviation value (RSD) was less than 15%. The accuracy of the analytical procedures was verified through the CQM values (Table 1). The method quantification limit (reporting level) for THg and Cd was 0.07 and 0.006 mg/kg in wet weight basis respectively.

Table 1. Obtained and certified concentrations (μg/kg, wet weight) in CQM

CQM		THg	Cd
T/07243	Certified	707 (469-946)	800 (535-1065)
	Obtained	722.55±50.58	782.40±70.41
T/07279	Certified	106 (59-152)	7.55 (5.76-9.33)*
	Obtained	107.48±6.45	7.40±0.59*

*mg/kg

Overall, the length value ranged from 40.0-200.0 cm (mean 103.5 cm, median 101.0 cm) while the weight ranged between 13.3-92.6 kg (mean 42.4 kg, median 39.9 kg).

In our study, the muscle parts of swordfish were used to detect the THg and Cd levels. The literature reveals that the liver tissues usually contain a higher value of THg and Cd (Damiano *et al.*, 2011), Our analysis, focused on the muscle part only because the liver is usually not an edible part of the fish. On the other hand, muscle act as a good reservoir for accumulation of environment contaminants (Damiano *et al.*, 2011).

Three clusters were identified as areas of Bay of Bengal (B), off Dondra (D) and Arabic sea (A) (Figure 1) when the catching locations were fed into Google map. Based on the map 27, 39 and 9 fish belonged to the Bay of Bengal, off Dondra and Arabic sea areas. The THg concentration in swordfish caught in off Dondra area (average: 0.69 mg/kg and the range <0.06-4.30 mg/kg) show a difference than other two capture areas, Bay of Bengal (average 0.55 mg/kg and the range <0.06-2.73 mg/kg) and Arabic sea area (average 0.51 mg/kg and the range 0.21-1.10 mg/kg), that is however not the statistically significant difference ($p>0.05$). Esposito *et al.* (2018), analyzed THg level of swordfish from 11 FAO fishing areas and observed the highest THg levels in fish from the Indian Ocean (0.955±0.118 mg/kg from West Indian Ocean; FAO 51 and 0.604±0.082 mg/kg East Indian Ocean; FAO 57). Further, it is generally believed that the THg concentration in fish from the Mediterranean Sea area is higher than in other oceans, because of the numerous deposits of Hg ore found in the surrounding countries (Esposito *et al.*, 2018, Storelli *et al.*, 2005). However, Esposito *et al.* (2018), reported a comparatively lower THg level in this area. Damiano *et al.* (2011), studied the THg, Cd and Pb level of SF from the Mediterranean Sea and Atlantic areas, observing a significant difference between the values in these two areas. The same kind of difference was observed in SF caught from two Atlantic Ocean areas by Branco *et al.* (2007) and highlighted that the most plausible reasons for this difference as the quantity and type of food eaten. Mendez *et al.* (2001), were highlighted that the THg level of SF depends not only on the size of fish but also the Hg content of the diet.

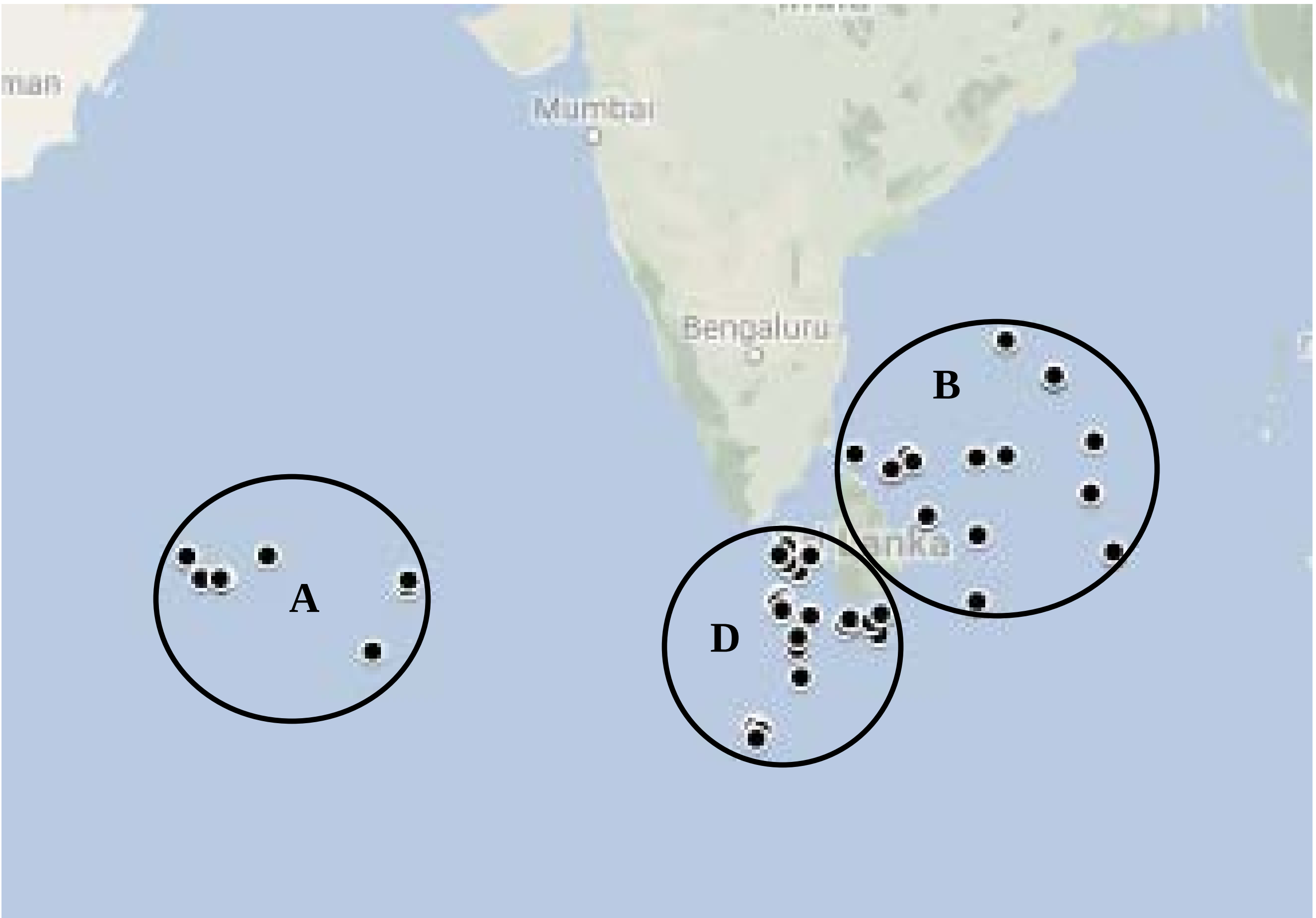


Figure 1: Location of sampling; D-off Dondra sea, B-Bay of Bengal sea, A-Arabic sea

Even though the results point to high variability, the statistical analysis showed that there is no significant difference in THg and Cd levels of SF muscle tissue in this 3 location ($p>0.05$). The THg level ranged from <0.07-4.30 mg/kg while the mean and median values were 0.62 and 0.50 mg/kg respectively. The highest value of THg (4.30 mg/kg) was from the off Dondra area, which was from a 28 kg, 64 cm weight fish. Furthermore, the THg found in this work are similar to those reported in the literature for swordfish of various origins (**Table 2**). The high bioaccumulation of the Hg in swordfish is generally endorsed by their top position in the food web and long lives (Branco *et al.*, 2007).

Among the analysed samples, 10 individuals (13.3%) exceeded the THg maximum allowable value set by EU and USFDA (1 mg/kg). Considering the number of fish exceeding 1 mg/kg level in 3 locations, 3 (11.1%), 6 (15.38%) and 1 (11.1%) represented in order B, D and A. The 31 individuals (41%) contained 0.50-1.00 mg/kg, 20 individuals (27%) 0.25-0.50 mg/kg while only 14 individuals (19%) below 0.25 mg/kg.

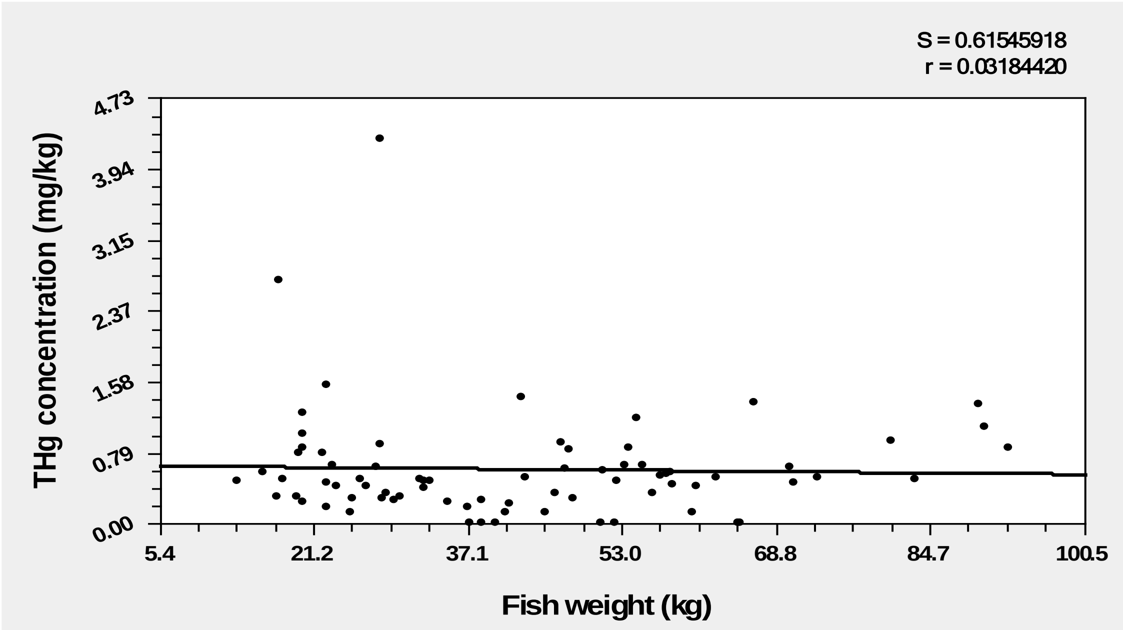
Cadmium in SF measured in muscle tissues ranged from <0.006-0.180 mg/kg (mean; 0.044, median; 0.033 mg/kg), Not a single individual exceeded the EU or USFDA maximum allowable level (0.30 mg/kg). The highest concentration of Cd (0.180 mg/kg), was recorded in a fish caught from the Bay of Bengal area and the size was 65.1 kg and 137.0 cm These results are consistent with the other study conducted in the Indian Ocean reported by Jinadasa *et al.* (2014) and with the other studies reported by Damiano *et al.* (2011) considering Mediterranean and the Atlantic Ocean with respect to the same species.

The concentrations of THg and Cd in muscle tissues of SF were investigated (fig. 2 and table 3). The linear fit models were run (y=a+bx) while the positive correlation of the metal concentration with the size of the fish was observed (x is fish weight or length and a, b are a parameter related with the fitted equation and the value given in Table 3). The fish length and weight were weakly associated with THg. A similar weak relationship has been observed between the edible part of swordfish and weight of fish (Mendez *et al.*, 2001), muscle tissue with length of the swordfish (Kojadinovic *et al.*, 2006) while strong relationship observed the THg and Organic Hg with the fish length (Chen *et al.*, 2007) and the dorsal muscle tissue THg and with the length and weight of swordfish (Jinadasa *et al.*, 2013).

In the study of Gewurtz *et al.* (2011), showed that the independent variables such as fish length and weight did not impact of their analysis and they highlighted that model fit typically get worse for larger sized fish, in general, there was a similar probability of under- and over-prediction.

Table 2: Summary of THg levels, origin, and a number of samples (n) in swordfish reported in the literature.

Origin	THg (mg/kg), wet weight			n	Reference
	Minimum	Maximum	Average		
Western Indian Ocean	0.241	1.880	0.955±0.118	21	Esposito *et al.* (2018)
Eastern Indian Ocean	0.091	1.400	0.604±0.082	19	Esposito *et al.* (2018)
Mediterranean and Atlantic ocean	0.66	2.41	__	56	Damiano *et al.* (2011)
Atlantic Ocean	0.031	9.8	__	52	Branco *et al.* (2007)
Sri Lanka, Indian Ocean	0.18	2.58	0.90±0.52	176	Jinadasa *et al.* (2013)
Indian and Atlantic Oceans	0.56	3.97	1.30±0.97	56	Chen *et al.* (2007)
Mediterranean Sea	0.02	0.15	0.07±0.04	58	Storelli *et al.* (2005)
Spain	0.177	1.227	__	__	Olmedo *et al.* (2013)
Spain	0.413	2.110	0.958±0.475	27	Torres-Escribano *et al.* (2010)
USA	0.15	3.07	1.40±0.18	18	Burger and Gochfeld (2006)
Mediterranean Sea	0.30	1.80	0.78±0.48	30	Barone *et al.* (2018)
Sri Lanka, Indian Ocean	<0.07	4.30	0.62±0.61	75	This study



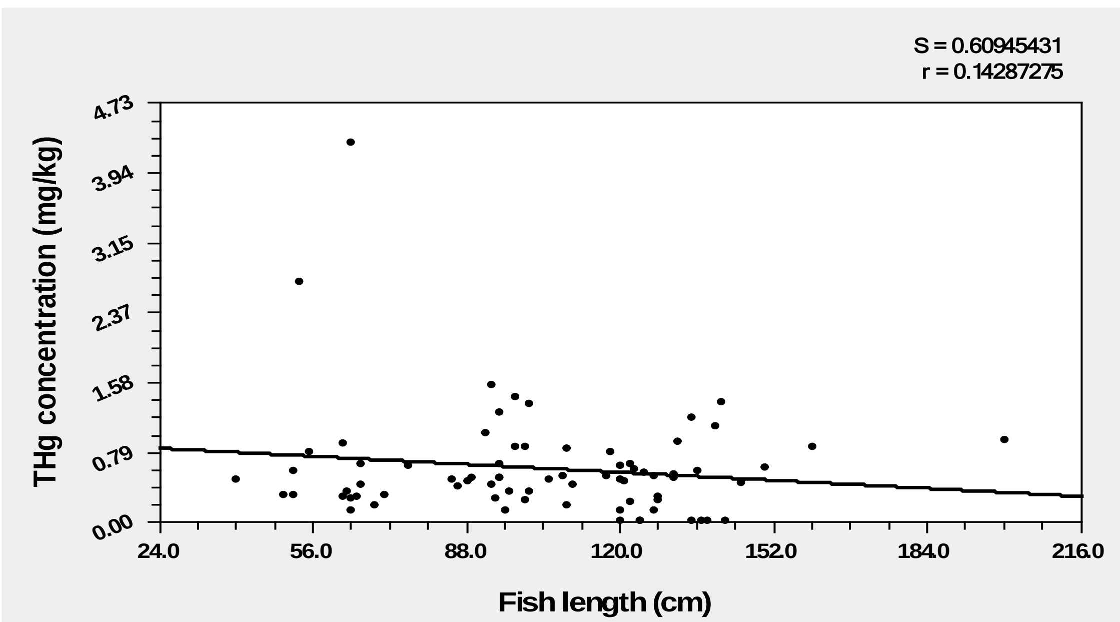
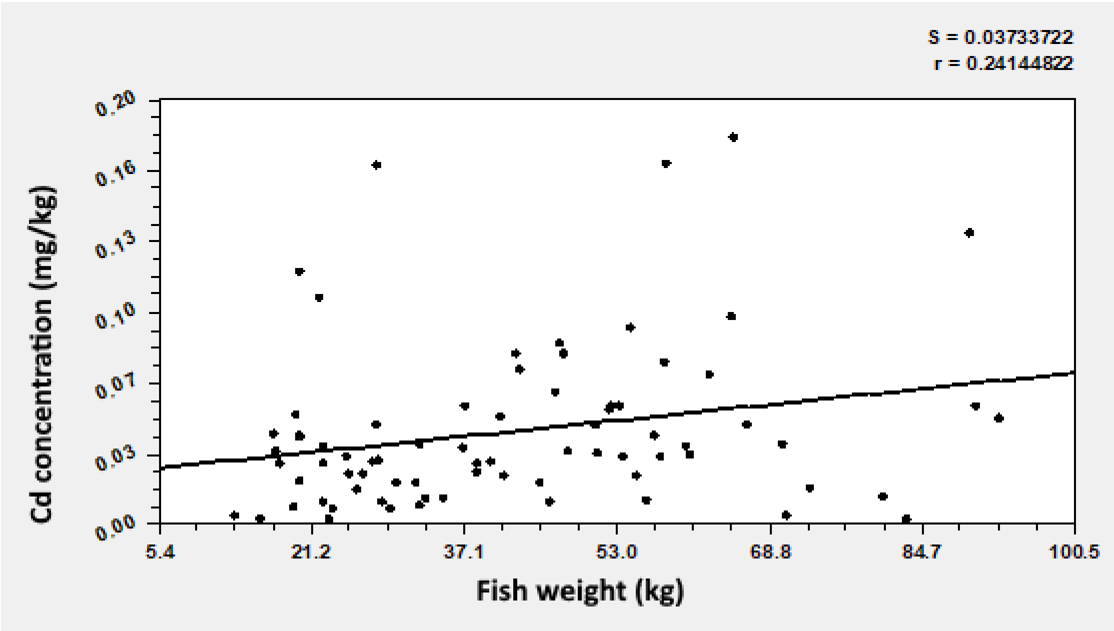


Figure 2. Relationship between THg and Cd concentration and length and weight of swordfish (part 1)



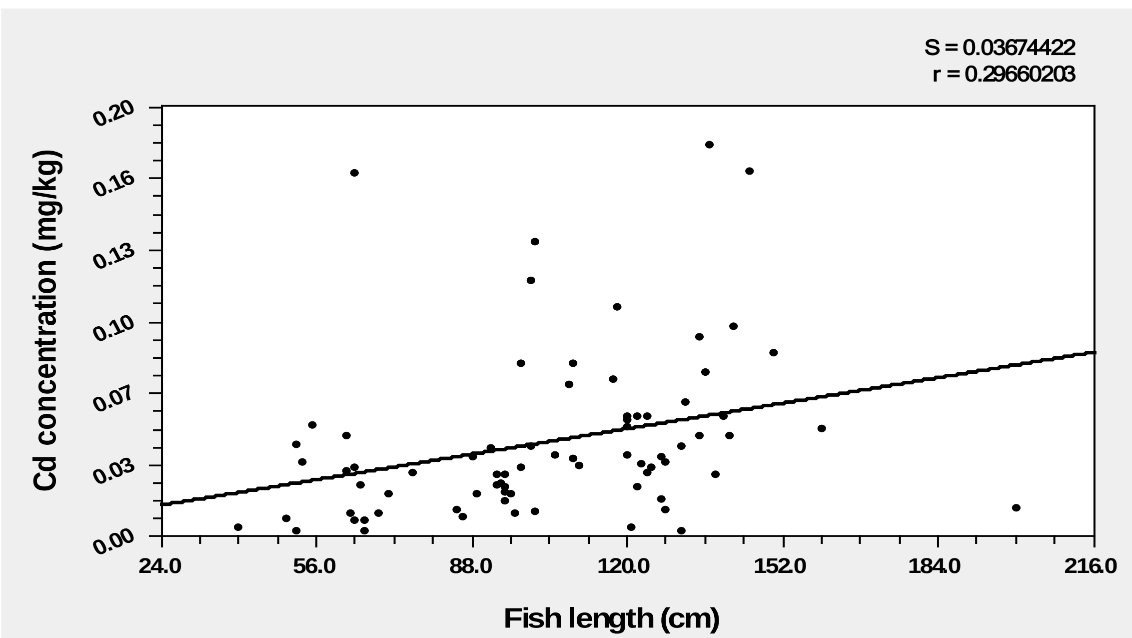


Figure 2. Relationship between THg and Cd concentration and length and weight of swordfish (part 2)

Table 3. Numerical value, standard error and correlation coefficient of the model run

Metal	Parameter	Standard Error	Correlation Coefficient	a	b
T-Hg	Weight	0.6155	0.0318	6.6013E-001	-9.8108E-004
	Length	0.6094	0.1429	9.1047E-001	-2.8197E-003
Cd	Weight	0.0373	0.2414	2.4306E-002	4.6480E-004
	Length	0.0367	0.2966	6.1452E-003	3.6576E-004

Conclusion

This study provided the picture of THg and Cd level of swordfish in the Indian Ocean around Sri Lanka. The mean THg and Cd level of swordfish were <0.07-4.30 mg/kg and <0.006-0.180 mg/kg respectively. The results indicated that the catching position is not a critical factor to estimate the THg or Cd level of swordfish. The weak positive significant relationship was observed with both studied metals with the length and weight of the fish.

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