

Determination of Cadmium and Zinc Concentration in Fish and Water from Sungai Kelantan

*Nadzifah Yaakub, Muhammad Nizam Abd Raoff, Muhamad Najmi Haris,
Aishatul Amiera Abdul Halim and Mazira Mansor*

Faculty of Bioresources And Food Industry, Universiti Sultan Zainal Abidin,
Besut Campus, 22200 Besut, Terengganu

Abstract: This study aimed to determine the concentration of Cadmium (Cd) and Zinc (Zn) in meat and gills of freshwater fishes and water samples from three different sampling along Sungai Kelantan. Sampling was carried out during monsoon and dry seasons and the fish were caught using gills net. Concentration of heavy metals in freshwater fish samples were analyzed using Inductively Coupled Plasma-Mass Spectrometer (ICP-MS). It was found that the concentration of Cd (mg/kg) in water samples is 0.0010 ± 0.0020 (Gua Musang), 0.0020 ± 0.0030 (Tanah Merah) and 0.0020 ± 0.0010 (Kota Bharu) while concentration of Zn (mg/kg) in water samples is 0.0020 ± 0.0040 (Gua Musang), 0.0030 ± 0.0020 (Tanah Merah) and 0.0040 ± 0.0020 (Kota Bharu). The Cd level (mg/kg) in meat of freshwater fishes was 0.0181 ± 0.0250 (Gua Musang), 0.0304 ± 0.0194 (Tanah Merah) and 0.0275 ± 0.0120 (Kota Bharu) while in gill was 0.0335 ± 0.0340 (Gua Musang), 0.0304 ± 0.0145 (Tanah Merah) and 0.0275 ± 0.0245 (Kota Bharu). The Zn level (mg/kg) in meat were 0.0165 ± 0.0172 (Gua Musang), 0.0136 ± 0.0192 (Tanah Merah) and 0.0275 ± 0.0245 (Kota Bharu) while in gill was 0.0232 ± 0.0317 (Gua Musang), 0.0230 ± 0.0215 (Tanah Merah) and 0.0275 ± 0.0245 (Kota Bharu). The level of Cd and Zn showed no significant different ($p > 0.05$) in both meat and gill of freshwater fishes during monsoon and dry season. The concentration of Cd and Zn in fish were below the permissible limit according to Malaysia Food Act (MFA, 1983), US Food and Drug Administration (USFDA, 1993), European Commission (EC, 2001) and Food Agriculture Organization (FAO, 2012) and is considered safe to be consumed.

Key words: Heavy metals • Water quality • Freshwater fish • Sungai Kelantan

INTRODUCTION

Sungai Kelantan is the longest river in the state of Kelantan, spanning a length of 248 km long and drains an area of 11,900 km², which occupy more than 85% of the State of Kelantan [1]. Sungai Kelantan has been used as medium for transportation, irrigation, industrial and food source for the local people. In recent years, the water quality of Sungai Kelantan is deteriorating which as reported by Department of Environment (DOE) Malaysia as polluted based on the suspended solid index. Among most of the pollutants discharged into the aquatic environment, heavy metals are regarded as one of the most serious pollutants due to their environmental persistence and tendency to accumulate in aquatic

environment [2]. Heavy metals that present in the water may accumulate in aquatic species, enter the food chain and cause serious harm to human health when the contamination content and exposure are significant [3]. Once introduced into the aquatic environment, trace metals are redistributed throughout the water column, deposited or accumulated in sediments and consumed by biota [4]. In recent years, focus has been directed to the level of bioaccumulation of heavy metals in aquatic organisms like freshwater fish and their risk to the human health.

Detection of bioaccumulation of heavy metal in aquatic organisms is one of the ways of identifying the impact of heavy metals presence in river ecosystem. The entry of heavy metals contaminants into the environment

Corresponding Author: Nadzifah binti Yaakub, Faculty of Bioresources And Food Industry,
Universiti Sultan Zainal Abidin, Besut Campus, 22200 Besut, Terengganu, Malaysia.
Tel: +60192692501.

can be caused by human and natural activities that come from municipal, industrial and agricultural activity [5]. Although some of the heavy metals are essential as micronutrients to fish, high concentration in the food chain can cause toxicity and create environmental impacts that endanger aquatic ecosystems and human consumers [6]. Fish can absorb an increased amount of heavy metals that present in the water directly through the epithelial absorption via their gills which will be deposited in the tissues of their body [7].

Fish transfer the heavy metals from the environment into humans when they are consumed and therefore form the final link for the transfer of heavy metals in the water to humans [1]. Consumption of heavy metals above the permissible limit can lead to toxic effect to the human body, as they possess carcinogenic, mutagenic and neurotoxic properties. Cadmium (Cd) is a highly toxic heavy metals found in industrial factory and does not usually present in the environment in its pure metallic condition but rather as a combination of minerals. Higher temperatures promote accumulation of cadmium especially in the most burdened organs: kidneys and liver [8]. Potential exposures to Cd are highest through food intake but its absorption into human body is relatively as low as 3%. However, once cadmium is ingested, it remains in the human body for a long time and is retained in the kidney and liver. Itai-itai disease resulting from high exposure to cadmium has been recorded in Japan during 1940 until 1960 which was caused by exposure to Cd contaminated rice through Cd discharge into the river. People affected by this disease showed *osteomalacia* in bones and were characterized by extreme bone fragility and renal abnormalities. Compared to several other metal ions with similar chemical properties, zinc is relatively harmless. Only exposure to high doses has toxic effects, making acute zinc intoxication a rare event. In addition to acute intoxication, long-term, high-dose zinc supplementation interferes with the uptake of copper. Hence, many of its toxic effects are in fact due to copper deficiency. Consumption of fish that is contaminated with heavy metals is a serious issue because people who lives near the river and uses the river in daily basis consume fish everyday as a source of protein. High content of beneficial nutrients in fish help elevate good health in humans. Chronic toxicity of heavy metals is dangerous because it possesses high risk to human health. This study was carried out to determine the concentration of Cadmium (Cd) and Zinc (Zn) in meat and gills of freshwater fishes and water samples from three different sampling along Sungai Kelantan.

MATERIALS AND METHODS

Sampling for fish and water samples was carried out during the wet season (December 2016) and dry season (February 2017). Three stations were chosen along the Sungai Kelantan which covers all the length of Sungai Kelantan from upstream to downstream. The stations are located at Gua Musang (05.07'12.8, 102.01'11.5), Tanah Merah (05.46'31.6, 102.09'05.6) and Kota Bharu (06.09'07.0, 102.13'55.5).

Sample Collection: Fish were caught at each sampling station using several gill nets set up in the respective area which were left for 48 hours. The placement of the gill nets were chosen based on the ideal location in the river based on several factors. The gill nets were checked every 24 hours and all the fish caught were measured for length and weight before the fish were immediately stored in ice box with ice and kept cool. Water samples were taken at the respective sampling stations using water sampler and the water samples were stored in polyethylene bottles before kept in ice box for preservation purpose. Upon arrival at the laboratory, the fish were stored in the cool room at -20°C until further analysis.

Samples Analysis: The fish samples were dissected using scissors, scalpel and knife. The meat and gills part of the fish were dissected and weighed at 10 g per samples before the samples were oven-dried at 100°C for 24 hours until they achieved constant weight. Samples were then allowed to cool before the dry weights of the samples were taken. Acid digestion method was used based on the Association of Official Analytical Chemists (AOAC) [9]. Each samples of dried meat were put in the digestion tube where 10 mL of 69% Nitric acid were added and they were left overnight. Then, 10 ml of 70% nitric acid was added and left in room temperature overnight. On the next day, the samples were digested in block thermostat at 100°C for 2 hours before were left to cool. After that, 2 ml of 30% hydrogen peroxide (H₂O₂) was added. The digestion tubes were heated again for 1 hour until the color of solution change from orange to yellow and form clear solution. Then, the digestion tubes were allowed to cool before the solutions were filtered through 0.5 nm filter paper into a 25 mL volumetric flask. The solution was diluted with deionized water until the volume reached 25 mL. The water samples were taken out from the cool room and filtered with 0.5 nm filter paper into a 25 mL volumetric flask. The concentration of Cd and Zn were determined using Inductively Couple Plasma – Mass Spectrometry (ICP-MS).

Statistical Analysis: Two-way analysis of variance (ANOVA) was used to indicate significant differences of metal levels in fishes among different sampling sites and different seasons. All analysis was done at significant level of $p < 0.05$. All statistical analysis calculation was done using Microsoft Excel for Windows 7.

RESULTS AND DISCUSSION

A total of 45 fishes from nine different species were caught during the sampling period and were tabulated in the Table 1. All the fish were collected from three stations along Sungai Kelantan which are Station 1 (Gua Musang), Station 2 (Tanah Merah) and Station 3 (Kota Bharu) during wet season and dry season. The concentration of heavy metals in the freshwater fishes in this study was determined on dry weight basis because the values were more reliable and consistent than the wet weight value [10].

During the sampling in wet season, fishes that were collected at Station 1 (Gua Musang) include *Hemibagrus nemurus*, *Hampala macrolepidota*, *Hemibagrus wyckii* and *Barbonymus schwanenfeldii*. At Station 2 (Tanah Merah), *Hampala macrolepidota*, *Barbonymus gonionotus* and *Puntius daruphani* were collected and at Station 3 (Kota Bharu), collected fishes were *Pangasius micromenus* and *Clarias gariepinus*. Metal concentration in meat and gill of each fishes collected were determined. During sampling in dry season, species of fishes that collected in Station 1 (Gua Musang) were *Barbonymus schwanenfeldii*, *Barbonymus gonionotu*, *Hampala macrolepidota* and *Hemibagrus wyckii* while at Station 2 (Tanah Merah), *Hemibagrus nemurus*, *Pangasius micromenus* and *Puntius daruphani* species were collected. At Station 3 (Kota Bharu), *Clarias gariepinus*, *Channa striatus* and *Pangasius micromenus* species were collected. Metal concentration in meat and gill of each fishes collected were measured. The concentration of

heavy metals Copper and Lead were detected in the muscle and gill tissues of fish samples collected from Sungai Kelantan and are summarized in the Table 2.

Bioconcentration of Cadmium and Zinc in Meat and Gill During Wet Season:

The Cd levels in meat of fish samples during monsoon season at Station 1, Station 2 and Station 3 are 0.0181 ± 0.0076 , 0.0304 ± 0.0126 and 0.0275 ± 0.0021 mg/kg and while in gill are 0.0335 ± 0.0083 , 0.0304 ± 0.0126 and 0.0275 ± 0.0021 mg/kg. The highest Cd level in meat was observed at Station 2 (0.0304 mg/kg) and the least value of Cd was found at Station 3 (0.0275 mg/kg).

Fish collected at Station 1 were also found with the highest Cd levels in gill (0.0335 mg/kg) while Station 3 showed the lowest value (0.0275 mg/kg). The result indicates that the fishes at Station 2 contained the highest Cd in meat while at Station 1 contained the highest Cd level in gill than other station in monsoon season. The level of Cd showed no significant different ($p > 0.05$) in monsoon season.

Zn level in meat of fishes at Station 1, Station 2, Station 3 showed 0.0165 ± 0.0062 , 0.0136 ± 0.0009 and 0.0275 ± 0.0021 mg/kg, while in gills showed 0.0232 ± 0.0102 , 0.0230 ± 0.0071 , 0.0275 ± 0.0021 mg/kg (Table 4.6). The highest Zn level in meat was observed at Station 3 (0.0275 mg/kg) and the least value of Zn was found at Station 2 (0.0136 mg/kg). The highest Zn level in gill observed at Station 3 (0.0275 mg/kg) and the least value of Zn level was found at Station 2 (0.0230 mg/kg). The result indicates that the fishes in Station 3 contained the highest Zn level in meat and gills of fishes. The level of Zn showed no significant different ($p > 0.05$) in meat and gill of fishes at three different station along Sungai Kelantan.

The river at Kota Bharu is located close to the city center and receives heavy metals from both natural and anthropogenic sources including industrial release,

Table 1: Distribution of fish caught in three sampling station in both wet and dry seasons

Species (n)	Common name	S1	S2	S3
<i>Hemibagrus nemurus</i> (4)	Baung	+	+	-
<i>Hampala macrolepidota</i> (3)	Sebarau	+	+	-
<i>Hemibagrus wyckii</i> (7)	Baung	+	-	-
<i>Barbonymus schwanenfeldii</i> (5)	Lampam sungai	+	-	-
<i>Barbonymus gonionotus</i> (6)	Lampam jawa	+	+	-
<i>Puntius daruphani</i> (7)	Ikan krai	-	+	-
<i>Pangasius micromenus</i> (6)	Ikan juara	-	+	+
<i>Clarias gariepinus</i> (5)	Keli	-	-	+
<i>Channa striatus</i> (2)	Haruan	-	+	+

Note: + present, - not present, (n) number of individual fish caught

Table 2: Mean concentration (mg/kg± SD) of cadmium (Cd) and Zinc (Zn) in meat and gill at three different stations in Kelantan River during wet and dry season.

Places	Monsoon		Dry	
	Cd	Zn	Cd	Zn
Gua Musang				
Meat	0.0181± 0.0075	0.0165± 0.0062	0.0250± 0.0095	0.0172± 0.0067
Gill	0.0335± 0.0083	0.0232± 0.0102	0.0320± 0.0114	0.0317± 0.0153
Tanah Merah				
Meat	0.0304± 0.0126	0.0136± 0.0009	0.0194± 0.0069	0.0192± 0.0064
Gill	0.0304± 0.0126	0.0230± 0.0071	0.0145± 0.0007	0.0125± 0.0092
Kota Bharu				
Meat	0.0275± 0.0021	0.0275± 0.0021	0.0120± 0.0014	0.0120± 0.0014
Gill	0.0275± 0.0021	0.0275± 0.0021	0.0245± 0.0021	0.0245± 0.0021



Fig. 1: Sampling stations along Sungai Kelantan

domestic wastes and municipal sewage. In addition, metals in airborne particulates reach directly through atmospheric deposition and indirectly through surface runoff [11]. Concentrations of heavy metals at Gua Musang were almost comparable to the values observed at Tanah Merah, Station 1 and 2 are located in city upstream and receive rural and suburban influences. Downstream sites with urban influences showed higher heavy metals concentration.

Bioconcentration of Cadmium and Zinc in Meat and Gill during Dry Season: The Cd levels in meat of fish samples during dry season at Station1, Station 2 and Station 3 showed 0.0250±0.0095, 0.0194±0.0069 and 0.0120±0.0014 mg/kg, while in gill showed 0.0320±0.0114, 0.0145±0.0007 and 0.0245±0.0021 mg/kg (Table 4.6). The highest Cd level in meat was observed at Station 1 (0.0250 mg/kg) and the

lowest value of Cd was found at Station 3 (0.0120 mg/kg). Station 1 also found with the highest Cd levels in gill (0.0320 mg/kg) while Station 2 showed the lowest value (0.0145 mg/kg). The result indicates that the fishes at Station 1 contained the highest Cd in meat and gill than the other sampling location during the dry season. The level of Cd showed no significant different ($p>0.05$) in the dry season.

Zn level in meat of fishes at Station 1, Station 2 and Station3 showed 0.0172±0.0067, 0.0192±0.0064 and 0.0120±0.0014 mg/kg, while in gills showed 0.0317±0.0153, 0.0125±0.0092 and 0.0245±0.0021 mg/kg (Figure 4.6). The highest Zn level in meat was observed at Station 2 (0.0192 mg/kg) and the least value of Zn was found at Station 3 (0.0120 mg/kg). The highest Zn level in gill observed at Station 1 (0.0317 mg/kg) and the least value of Zn level was found at Station 2 (0.0125 mg/kg). The result indicates

Table 3: Mean concentration±SD of Cd and Zn in water at three different sampling at stations in both seasons (mg/kg).

Location	Monsoon		Dry	
	Cd	Zn	Cd	Zn
Gua Musang	0.0010±0.0000	0.0020±0.0000	0.0020±0.0000	0.0040±0.0000
Tanah Merah	0.0020±0.0000	0.0010±0.0000	0.0030±0.0000	0.0020±0.0000
Kota Bharu	0.0020±0.0000	0.0010±0.0000	0.0040±0.0000	0.0020±0.0000

that the fishes at Station 2 contained the highest Zn level in meat and Station 1 contained the highest Zn level in gills of fishes. The level of Zn showed no significant different ($p>0.05$) in meat and gill of fishes in three different station along Kelantan River.

The concentrations of heavy metals are different in meat and gill of fishes. It was observed that there is higher concentration of heavy metals presence in meat compared to in gills. There are several factors that can affect the accumulation of heavy metals [12]. Fish accumulates higher Cd and Zn concentrations during dry season compared to monsoon season and it vary between sampling stations (Tanah Merah< Kota Bharu< Gua Musang). This difference in heavy metals concentration is due to high temperatures during the dry season, which increased the activity, ventilation, metabolic rate and feeding session [12]. The lower heavy metal concentrations during monsoon season due to the dilution of metal levels associated with heavy rain. In general, the concentration of heavy metals should be higher in gills because the absorption of metals onto the gills surface as the first target for pollutants in water could be significant influence in the total metal levels of the gill. Target organs such as gills and intestine are metabolically active parts that can accumulate heavy metals in higher levels.

Bioconcentration of Cd and Zn in Water: The concentration of Cd and Zn in water samples at Station 1 (Gua Musang), Station 2 (Tanah Merah) and Station 3 (Kota Bharu) were tabulated in Table 3. The concentration of Cd in water samples in Station 1, Station 2 and Station 3 were 0.0010±0.0000, 0.0020±0.0000 and 0.0020±0.0000 mg/kg while concentration for Zn were 0.0020±0.0000, 0.0010±0.0000 and 0.0010±0.0000 mg/kg during monsoon season. Concentration of Cd at Station 1 is the lowest (0.0010 mg/kg) compare to Station 2 and Station 3 (0.0020 mg/kg). Station 1 has the highest concentration of Zn (0.0020 mg/kg) compared with other two sampling station.

The results shows the concentration of Cd in water samples at Station 1, Station 2 and Station 3 were 0.0020±0.0000, 0.0030±0.0000 and 0.0040±0.0000 while concentration for Zn were 0.0040±0.0000, 0.0020±0.0000

and 0.0020±0.0000 mg/kg during monsoon season. Concentration of Cd at Station 3 is the highest (0.0040 mg/kg) compare to Station 2 (0.0030 mg/kg) and Station 1 (0.0020 mg/kg). Station 1 has the highest concentration of Zn (0.0040 mg/kg) compared to Station 2 and Station 3. Level of Cd and Zn based on INWQS is at natural level. It is which could be due to the natural land composition type.

Level of heavy metals is slightly higher at Station 1 (Gua Musang) and Station 2 (Tanah Merah) compared to Station 3 (Kota Bharu). Gua Musang is located in southern of Kelantan and is the largest district in Kelantan. The land use in this study area is mainly for rubber plantation, logging activity and located near with Lojing which is where logging activity is high. Station 3 is located in downstream area or estuarine area. Kota Bharu is one of the main districts in Kelantan and categorized as the highest population district in Kelantan. Kota Bharu is considered as the largest town in Kelantan and has been undergoing rapid urbanization in the recent time. Nevertheless, there are also agricultural land use in the study area is mainly paddy grown here other than mixed horticulture, rubber and orchards. The bedrock cropping out in the central ranges and lying beneath the river basin comprises granites, metamorphic rocks and minor tuff of Paleozoic identified as phyllite. The bedrock underlying the basin in the study area to the east of the Kelantan River is mainly consist of granite but changing to phyllite towards west. Phyllite and other metamorphosed sediments are fine grained and mostly weathered to clay and silt [13]. Sampling stations 2 in this study are located within Tanah Merah which is normally affected by human activities and the water has changed to a brownish color. In addition, sampling undertaken after the monsoon season and most rivers are normally more turbid within this period compared to dry seasons. Sampling Station 2 has slightly lower in concentration of Cd and Zn due to location near to upstream area. The river basin was forest, opened areas of roads or plantation, river and water bodies [6]. As the river flow declines in dry season, the rate of sedimentation and consequently the concentration is enhanced. In monsoon season, on the other hand, increased river flow causes a dilution effect

Table 4: Comparison mean concentrations of heavy metals (mg/kg) in water sample from the Kelantan River during the monsoon and dry seasons with the previous study and the standard criteria

Study	Cd	Zn
Gua Musang	0.0010±0.0020	0.0020±0.004
Tanah Merah	0.0020±0.0030	0.0030±0.0020
Kota Bharu	0.0020±0.0010	0.0040±0.0020
Previous Study		
Galas River		
Zarith & Mohd (2015)	ND	0.0030±0.0010
Kelantan River		
Ahmad <i>et al.</i> (2009)	0.3700	1.5000

*ND=not detected

Table 5: Heavy metals content (mg/kg) in meat and gills of fishes in three sampling stations

Species	Cd	Zn
<i>Hemibagrus nemurus</i>	0.0246±0.0113	0.0192±0.0080
<i>Hampala macrolepidota</i>	0.0217±0.0067	0.0177±0.0072
<i>Hemibagrus wyckii</i>	0.0365±0.0120	0.0145±0.0007
<i>Barbonymus schwanenfeldii</i>	0.0216±0.0052	0.0159±0.0054
<i>Barbonymus gonionotus</i>	0.0198±0.0145	0.0190±0.0054
<i>Puntius daruphani</i>	0.0127±0.0012	0.0163±0.0059
<i>Pangasius micromenus</i>	0.0270±0.0017	0.0230±0.0079
<i>Clarias gariepinus</i>	0.0240±0.0012	0.0192±0.0056
<i>Channa striatus</i>	0.0215±0.0091	0.0183±0.0079

and consequently, metal concentration in sediment declines. Although at the onset of monsoon season the first flush effect may enhance the concentration, the dilution effect predominates as the season progresses [11].

The concentration of Cd and Zn in this study was compared with the result from Ahmad *et al.* [1]. The concentration of Cd and Zn in water is higher in this study compared to previous study. This study showed that concentration of Cd is at natural level while Zn in between class I to IIB. According to Ahmad *et al.* [1], the concentration of Cd and Zn in Kelantan River classified in class II to III. This study showed that concentration of Cd and Zn almost the same with study at Kelantan River by Ahmad *et al.* [1] referred to Table 4.

Mean concentration of Cd in fish species were *P. daruphani* < *B. gonionotus* < *C. striatus* < *B. schwanenfeldii* < *H. macrolepidota* < *C. gariepinus* < *H. nemurus* < *P. micromenus* < *H. wyckii* (Table 5). The highest mean concentration of Cd was detected in *Hemibagrus wyckii*, *Pangasius micromenus* and *Hemibagrus nemurus* with values of 0.0365, 0.0270 and 0.0246 mg/kg, respectively but still below permissible limit according to World Health Organization (WHO). Species that have least concentration of Cd is *Puntius daruphani* with value of 0.0127 mg/kg. Overall, Cd

concentration in all species was below permissible limit and safe to be consumed. Cd is a non-essential element and can be toxic to humans when ingested or inhaled in high doses [14]. Cd causes kidney damage (renal tubular damage), lead to lung cancer and prostate cancer [15].

Mean concentration of Zn in fish species were *H. wyckii* < *B. schwanenfeldii* < *P. daruphani* < *H. macrolepidota* < *C. striatus* < *B. gonionotus* < *C. gariepinus* < *H. nemurus* < *P. micromenus*. The highest mean concentration of Zn was detected in *Pangasius micromenus* with the value of 0.0230 mg/kg. Species that have lowest concentration of Zn is *Hemibagrus wyckii* with the level of 0.0145 mg/kg. Zn toxicity is rare, but at concentrations in water up to 40 mg/kg, may induce toxicity, characterized by symptoms of irritability, muscular stiffness and pain, loss of appetite and nausea [16]. Zn is one of the essential metals that needed by the body to regulate metabolic process, embryonic development, cell differentiation and cell proliferation. Nonetheless, if the level exceeds the maximum residual limits, it will cause problem to human health.

The level of Cd and Zn concentration is different between meat and gill. It is because due to the variation in the environment where the fish lives in. The absorption of metals onto the gill surface as the first target for pollutants in water could also be a significant influence in the total metal levels of the gills. Target organs such as gills and intestine are metabolically active parts that can accumulate heavy metals in higher levels. There are several factors that can affect the accumulation of heavy metals. It also due to detoxifying ability of liver that tend to accumulate large amount of Cd and Zn in tissues [12].

Detection of heavy metals that present in freshwater fishes is important for environmental management and human consumption safety. In this study, the meat and gill of freshwater fishes from three different sampling locations alongside the Kelantan River were used for Cd and Zn detection. In general, the concentration of Cd and Zn should be below the permissible limit as these metals do not naturally exist in high concentration in the nature. For Cd, it was not an essential element, thus the concentration should be lesser than Zn. Nonetheless if the levels of heavy metals exceeds the maximum residual limits, it will cause problem to human health. Cd is the most toxic and non-essential. Heavy metal has wide distribution in the earth's crust and aquatic environments [17]. Cd occurs naturally in some phosphate rock and may this account for the high concentration of Cd in some phosphate fertilizers such as super phosphate fertilizer with increased urbanization and industrialization [2].

Table 6: Comparison mean concentrations of heavy metals (mg/kg) in meat of fish from the Kelantan River during the monsoon and dry seasons with previous study and the permissible limit in fish

Study	Cd	Zn
	Meat	Meat
Gua Musang	0.0181±0.0250	0.0165±0.0172
Kuala Krai	0.0304±0.0194	0.0136±0.0192
Pasir Mas	0.0275±0.0120	0.0275±0.0245
Rohasliney Hashim <i>et al.</i> (2014)	0.0490±0.0350	-
Zarith & Mohd (2015)	0.002	0.0434
Abdulali <i>et al.</i> (2003)	0.0020	0.0206

Table 7: Comparison mean concentrations of heavy metals (mg/kg) in gill of fish from the Kelantan River during the monsoon and dry season with the previous study

Study	Cd	Zn
Gua Musang	0.0335±0.0340	0.0232±0.0317
Tanah Merah	0.0304±0.0145	0.0230±0.0215
Kota Bharu	0.0275±0.0245	0.0275±0.0245
Makedonski <i>et al.</i> (2015)	0.0080±0.0310	0.0073

Table 8: Permissible limit of metal for freshwater fish (mg/kg)

	Permissible limit of Heavy Metals in fish	
	Cd	Zn
Malaysia Food Act (1983) and Regulation (1985)	1	100
FAO (2012)	0.05	30
USFDA (1993)	0.01-0.21	-
EC (2001)	0.05	-

The concentration of Cd in meat in this study is lower compared to Rohasliney *et al.* [19] and Zarith & Mohd [20] showed in Table 4.8. This study showed that concentration of Cd in fish meat is safe to be consumed. According to Rohasliney *et al.* [18], the concentration of Cd in fish in Kelantan River is slightly below the limits of the permissible level with 0.0490 mg/kg, respectively.

The concentration of Cd and Zn in gill in this study is higher compared to previous study by Makedonski *et al.* [20]. According to Makedonski *et al.* [20], the concentration of Cd and Zn in gill of fish at black sea, is 0.0080 and 0.0073 mg/kg. Excess of Zn can affect the gastrointestinal tract [4]. The concentration of Cd and Zn in fish gill is within the range of permissible limit according to EC [21] and Malaysia Food Act and Regulation [22].

The limits of heavy metal concentration in fish have been set to safeguard public health. Malaysia, for example, has set maximum limits of contamination for Cd and Zn based on permissible limits recommended by the Malaysian Food Act (MFA) [22]. In this study, however, the heavy metal permissible levels were also compared with others such as the WHO (1985), USFDA (1993), EC (2001) and FAO (2003). This study showed the Cd and Zn concentration in meat and gill of freshwater fishes along Kelantan River was below the permissible limit of

Malaysia Food Act and Regulation [22], WHO/FAO [23], USFDA and EC [21]. The result obtained in this study proves that the fish was safe to human consumption. From the results, the determination level of Zn in fish is lower than level of Cd in both meat and gill.

CONCLUSION

The result of this study showed that Zn and Cd in the fish and water samples was below the permissible limit of heavy metals recommended by Malaysia Food Act (MFA, 1983)[22], US Food and Drug Administration (USFDA, 1993), European Commission (EC, 2001)[21] and Food and Agriculture Organization (FAO, 2012)[23] and the fish are considered safe to be consumed. Although the level of heavy metals that were detected is low there is a risk of future increases in heavy metals concentration, along with the increasing human activity along Sungai Kelantan. A regular monitoring of heavy metals concentration in water and fish are needed to minimize heavy metals toxicity in freshwater ecosystem.

REFERENCES

- Ahmad, A.K., I. Mushrifah and M.S. Othman, 2009. Water quality and heavy metal concentrations in sediment of Sungai Kelantan, Kelantan Malaysia: A baseline study. *Sains Malaysiana*, 38: 435-442.
- Benzer, S., H. Arslan, N. Uzel, A. Gül and M. Yılmaz, 2013. Concentrations of metals in water, sediment and tissues of *Cyprinus carpio*, 1758 from Mogan Lake (Turkey). *Iranian Journal of Fisheries Sciences*, 12: 45-55.
- Fernandes, C., A. Fontainhas-Fernandes, F. Peixoto and M.A. Salgado, 2007. Bioaccumulation of heavy metals in *Liza saliens* from the Esmoriz-Paramos coastal lagoon, Portugal. *Ecotoxicology and Environmental Safety*, 66: 426-431.
- Kljaković- Gašpić, Z., Herceg-Romanic, S., Kožul, D. & Veža, J. (2010). Biomonitoring of organochlorine compounds and trace metals along the Eastern Adriatic coast (Croatia) using *Mytilus galloprovincialis*. *Marine Pollution Bulletin*, 60, 1879-1889
- Kazi T.G., M.B. Arain, M.K. Jamali, N. Jalbani, H.I. Afridi, R.A. Sarfraz, J.A. Baig and Abdul Q. Shah, 2009. Assessment of water quality of polluted lake using multivariate statistical techniques: A case study, *Ecotoxicology and Environmental Safety*, 72: 301-309, 2009.

6. Kane, S., P. Lazo and A. Vlora, 2012. Assessment of heavy metals in some dumps of copper mining and plants in Mirdita Area, Albania, in Proceedings of the 5th International Scientific Conference on Water, Climate and Environment, Eds., Ohrid, Macedonia.
7. Fatima, M. and N. Usmani, 2013. Histopathology and bioaccumulation of heavy metals (Cr, Ni and Pb) in fish (*Channa striatus* and *Heteropneustes fossilis*) tissue: A study for toxicity and ecological impacts. Pak. J. Biol. Sci., 16: 412-420.
8. Baldwin, D.H., J.F. Sandahl, J.S. Labenia and N.L. Scholz, 2003. Sublethal effects of copper on coho salmon: impacts on non-overlapping receptor pathways in the peripheral olfactory nervous system. Environmental Toxicology and Chemistry, 22: 2266-2274.
9. AOAC. 1984. Association of Official Analytical Chemists. In official methods of association analytical chemists. Washington, DC:AOAC, pp: 418.
10. Pe'rez-Lo'pez, M., M. Hermoso de Mendoza, A. pez Beceiro and F. Soler Rodri' guez, 2008. Heavy metal (Cd, Pb, Zn) and metalloid (As) contents in raptor species from Galicia (NW Spain). *Ecotoxicology and Environmental Safety*, 70: 154-162.
11. Pandey, J. and R. Singh, 2015. Heavy metals in sediments of Ganga River: up- and downstream urban influences. Applied Water Science, pp: 1.
12. Alweher, S.M., 2008. Levels of heavy metals Cd, Cu and Zn in three fish species collected from the Northern Jordan Valley, Jordan. Jordan Journal of Biological Sciences, 1: 41-46.
13. Mohammad, M.A.K., A.S. Nor, M.A.B. Arham and M.A. Baten, 2014. Impact of the Flood Occurrence in Kota Bharu, Kelantan Using Statistical Analysis. Journal of Applied Sciences, 14: 1944-1951.
14. Nwani, C.D., V.C. Nwoye, J.N. Afukwa and J.E. Eyo, 2009. Assessment of heavy Metals Concentration in the tissues (Gills and Muscles) of Six commercially Important Freshwater Fish Species of Anambra River South-East Nigeria. Asian Jr. of Microbiol. Biotech. Env. Sc. 11: 7- 12.
15. Nas-NRC, National Academy of Sciences-National Research Council, Food and Nutrition Board, Recommended Dietary Allowances, 8th Edition, National Academy Press, Washington DC, pp: 1974.
16. Prohaska, J. R. & Gybina, A. A. (2004). Inter-cellular copper transport in mammals. *The Journal of Nutrition*. 134:1003-1006.
17. Muthukumaravel, K. and M.G. Paulay, 2007. Toxic effect of cadmium on the electrophoretic protein patterns of gill and muscle of *Oreochromis mossambicus* EJournal of Chemistry, 14: 2884-286.
18. Rohasliney, H., H.S. Tan, M. Noor Zuhartini Md and P.Y. Tan, 2014. Determination of Heavy Metal Fishes from the lower reach of Kelantan River, Kelantan, Malaysia. Tropical Life Sciences Research, 25: 21-39.
19. Zarith, S.B. and Y.I. Mohd, 2015. Determination of heavy metal accumulation in fish species in Galas River, Kelantan and Beranang mining pool, Selangor. Procedia Environmental Sciences, 30: 320 -32.
20. Makedonski, L., K. Katya Peycheva and M. Stancheva, 2015. Determination of some heavy metal of selected black sea fish species. Food Control, pp: 1-6.
21. European Commission (EC) (Commission Regulation) 2001. Np 466/2011. Official Journal of European Communities, 1: 77/1.
22. Malaysian Food Act (MFA) Malaysian Food and Drug (1983). Kuala Lumpur: MDC Publishers Printer Sdn. Bhd.
23. Food and Agriculture Organization (FAO) 2012. From Heavy Metal Regulations Faolex: [http:// faolex.org/docs/ pdf/eri42405.pdf](http://faolex.org/docs/pdf/eri42405.pdf).