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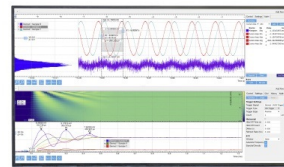
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Determination of Selected Toxic Metal (As, Cd, Pb) and Essential (Zn, Cu) Elements in Local Canned Seafood Products

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Abstract. In this study, the concentrations of selected toxic metals (Pb, As, Cd) and essential elements (Zn, Cu) of local canned seafood including sardines, tuna, anchovies, shellfish (mussels and cockles), prawns, mackerel and cuttlefish were determined. All 30 samples were purchased from supermarkets in Nilai, Negeri Sembilan. The highest concentration of trace metals in all samples was Zn which is 38.96 ± 0.56 mg/kg, followed by Cu (2.40 ± 0.57 mg/kg), Pb (1.71 ± 0.71 mg/kg), As (0.96 ± 0.70 mg/kg) and Cd (0.25 ± 0.70 mg/kg). The average concentration of toxic elements were the highest in anchovy species with concentration of 6.93 mg/kg followed by shellfish (4.24 mg/kg), mixed seafood (4.17 mg/kg), cuttlefish (3.63 mg/kg), prawns (3.28 mg/kg), sardines (2.65 mg/kg), tuna (1.68 mg/kg) and the lowest which is mackerel (1.36 mg/kg). The results were compared with the previous studies.

INTRODUCTION

Food is the primary source of essential elements for humans and it is a source of exposure to heavy metals elements [1]. Some of the heavy metals are toxic, persistent and not easily biodegradable. Heavy metals can be categorized in two categories which are toxic and essential elements. Toxic elements such as arsenic (As), cadmium (Cd) and lead (Pb) are natural trace components of the aquatic environment, but their concentrations have increased due to industrial, agricultural and mining activities [2]. These toxic elements are known to have no role in biological systems and the presence of these elements even at trace amounts can give negative effect to human body while essential elements such as zinc (Zn) and copper (Cu) can lead to toxicity when ingested in excess quantity over a long time period through food or water [3].

Heavy metal pollution in marine ecosystem has gained a special attention due to serious environmental and health issues. Marine life will also be affected as heavy metals contamination are widely contaminated into the ecosystem due to agricultural, industrial and mining activities. The results from accumulation of heavy metal consumed by marine organism will give adverse impact on the human health [2]. Since canned food is commonly consumed in daily life, therefore, the knowledge of the toxic heavy metal concentrations in canned food samples like fish (sardine and tuna) is important for assessing their risk on human health when consumed excessively.

Canned food is any edible material that has undergo preservation process for long period of time and packed in the airtight container. Canned seafood is one of the common canned food produced in Malaysia. Consumption of canned seafood is expanded in many parts of the world as seafood provide vitamins, proteins, fatty acid (omega-3) and contain a low trans-fat quantity [4]. Canned seafood are well eaten in the developed world because it is convenient and affordable for most working families [4]. A variety of canned food with variety style of cooking can be found in Malaysia and can be easily purchased at any supermarket or grocery store throughout the country.

The purpose of this study is to determine the level of selected toxic and essential elements in canned seafood produced and marketed in Malaysia. As, Cd, Pb were chosen to be studied because they are natural trace components of the aquatic environment, but their levels have increased due to industrial, agricultural and mining activities.

However, for Zn and Cu, they are among of essential elements that have to be taken in its appropriate amounts as excess or lack intake of them can give effects to body. Levels of trace heavy metals (As, Cd, Pb, Zn and Cu) in canned seafood (anchovy, shellfish, mixed seafood, cuttlefish, prawns, sardines, tuna and mackerel) that are produced and marketed in Malaysia were determined using flame atomic absorption spectrometry (AAS) and graphite furnace atomic absorption spectroscopy (GFAAS) as the concentration in these samples were estimated to be in mg/kg level.

MATERIAL AND METHODS

Chemicals and Reagents

All the chemicals used for the determination of toxic and essential elements in local canned seafood product were analytical reagent grade: nitric acid (HNO₃) (Fisher Chemical, United States), hydrochloric acid (HCl) (Fisher Chemical, United States), lead(II) nitrate (MERCK, Darmstadt, Germany), cadmium sulfate hydrate (Fluka, Macedonia), arsenic (MERCK, Darmstadt, Germany), zinc sulfate and copper sulfate (Bendosen, Selangor, Malaysia). The standards 1000 mg/kg of Pb, Cd Cu, and Zn solutions were prepared by dissolving 1.598 g of Pb(NO₃)₂, 2.7441 g of CdN₂O₆.4H₂O, 2.744 g of CuSO₄.5H₂O and 4.397 g of ZnSO₄.7H₂O respectively in a 1000 mL volumetric flask, diluted until reaching the mark with distilled water. Standard 1000 mg/mL of As solution was diluted to 100 mg/mL from the standard solution in 100 mL volumetric flask. Standard solutions of mixtures were produced by diluting each stock solutions in 5% nitric acid to preserve the metals.

Sample Preparation and Acid Digestion Method

30 samples of different types of local canned seafood products were purchased from supermarkets in Nilai, Negeri Sembilan. The recommended procedures were used in this study with slight modifications as follows [5]. The samples were homogenized by using ceramic knife, mortar and pestle. All samples were dried overnight in drying oven at the temperature of 120-125 °C because zinc has the lowest volatility temperature which is 127 °C. 2 g of dried samples were weighed into a porcelain crucible and heated in the muffle furnace at the temperature of 200 °C for 90 minutes and gradually heated until the temperature achieved 350 °C for 4 hours as to change the physical properties of sample into the form of ash. Then, the samples were digested into 10 ml of mixed concentrated HCl and HNO₃ (HCl:HNO₃ = 1:1) followed by heating on an electric hot plate until all samples were evaporated to near dryness. Samples were then filtered, transferred and diluted into 25 ml volumetric flask with 3% HNO₃.

Instrumentation

Atomic absorption spectrometer (AAS), equipped with a 240 Fast Sequential Atomic Absorption from Agilent (Santa Clara, US) was used to determine the concentration of Cd, Pb, Al, Zn, and Cu. For the determination of As, 240 Zeeman Atomic Absorption from Agilent (Santa Clara, United State) was used. This offers high sensitivity and proven trace-level detection limit with 4 lamp positions [6]. The flow gas is air/acetylene; the source of energy used is the hollow cathode lamp for each tested element.

RESULTS AND DISCUSSION

Toxic and Essential Elements Concentration

In this study, concentration of toxic element (As, Cd and Pb) and essential elements (Zn and Cu) of 30 local canned seafood samples were successfully analyzed. Cd, Pb, Cu and Zn were determined using flame atomic absorption spectrometry (AAS) while As was analyzed using graphite furnace atomic absorption spectrometry (GFAAS). All metal concentrations were determined on a dry weight basis. The contents of examined trace metals in local canned seafood were found to be in the range of 0.00 - 7.06 ± 1.13 mg/kg for As, 0.46 ± 0.93 - 3.87 ± 0.71 mg/kg for Pb, 0.01 ± 0.40 - 1.66 ± 0.63 mg/kg for Cd, 0.58 ± 0.71 - 8.31 ± 0.71 mg/kg for Cu and 17.57 ± 0.71 - 84.32 ± 0.31 mg/kg for Zn were summarized in Table 1. According to the result found, the highest concentration of trace metals in all samples was Zn with 38.96 ± 0.56 mg/kg, followed by Cu (2.40 ± 0.57 mg/kg), Pb (1.71 ± 0.71 mg/kg), As (0.96 ± 0.70 mg/kg) and Cd (0.25 ± 0.70 mg/kg). Concentrations obtained in this study were higher maybe due to the water pollution that

leads to marine life contaminated with toxic metals that come from industrial and domestic waste water. The summary of the metal concentrations are tabulated in Table 1.

Arsenic (As)

Human will consumed Arsenic from animals and plants that ultimately be taken up by animals and plants [7]. As shown in Table 1, canned seafood with the highest concentration of As was in S15 with the concentration of 7.06 ± 1.13 mg/kg. Other samples worth noting were some tuna samples which are S5, S19 and S26 since there were no As detected in these canned seafood samples. From Table 2, anchovies was found to have the highest concentration of As compared to other type of seafood. Toxicological study was made by World Health Organization (WHO) and has estimated $0.1-3$ $\mu\text{g}/\text{kg}$ bw/day as the maximum concentration of As allowed for a human body. Concentration that exceed this limit can cause cancers of the urinary bladder and lung and skin lesions (hyperkeratosis, hyperpigmentation and hypopigmentation). Arsenic contamination results from activities such as metals and alloys manufacturing, refining petroleum and burning fossil fuels and wastes [8].

Lead (Pb)

Lead are found in the environment both from natural sources and anthropogenic activities [9]. The canned seafood products shows concentration of Pb above the guidelines of Malaysian Food Act and Regulation were S2, S10, S11, S12, S13, S15, S16, S17, S21, S23 and S27. The highest Pb concentration which is 3.87 ± 0.71 mg/kg corresponded to canned seafood product is in the S2. On the other hand, S5 with the concentration of 0.46 ± 0.93 mg/kg shows the lowest concentration of Pb. However the lowest concentration in this study (0.46 ± 0.93) which is S5 exceeds the level in guideline of Joint FAO/WHO Expert Committee on Food Additives (JECFA) which is 0.03 mg/kg but it was lower the guideline of Malaysian Food Act and Regulations (1996) which is 2.0 mg/kg. Thus, all 30 samples exceed the guideline of JECFA (1999) but more than 50% of samples analyzed showed Pb levels below the guideline of Malaysian Food Act and Regulations (1996) which is 3.0 mg/kg. According to Table 2, Tuna canned seafood has the highest average concentration of Pb than the other type of seafood. High concentration of lead in marine life can be due to water pollution that leads to fish contaminated with toxic metals, from many sources such as industrial and domestic wastewater. Toxicological study that reported by JECFA showed that neurodevelopment for children and systolic blood pressure adults were the effects for excess intake of Pb.

Cadmium (Cd)

Cadmium enters organisms primarily through consumption and respiratory tract [10]. With regard to Cd, sample S17 (cuttlefish) presented the highest concentration of Cd which is 1.66 ± 0.63 mg/kg. The average concentration of Cd in cuttlefish (0.99 ± 0.51). On the other hand, sample showing the lowest concentration of Cd were S25 and S28 (tuna samples) which are 0.01 ± 0.71 mg/kg and 0.01 ± 0.40 mg/kg respectively. Toxicological study was made by JECFA to get the best estimated concentration for maximum amount allowed of Cd ranging from 0.0022 to 0.012 mg/kg bw/month. From Table 1, samples with the lowest concentration of Cd (S25 and S28) in canned seafood samples were below the estimated concentration made by JECFA. For the rest of canned seafood analyzed, Cd concentration was rather high, as all 28 samples exceed the estimated concentration made by JECFA. According to JECFA, excess intake of Cd can cause osteoporosis in women, height loss in men, kidney damage, elevated blood pressure and cardiovascular.

Zinc (Zn)

Deficiency of zinc can induce to loss of appetite, growth retardation, skin changes and immunological abnormalities [11]. The highest concentrations of Zn was found in S13 with the concentration of 84.32 ± 0.31 mg/kg. In contrast, S7 showed the lowest concentration of Zn (2.05 ± 1.26 mg/kg). Concentrations of Zn in all samples were generally below the concentrations in the guideline prescribed by JECFA and Malaysian Food Act and Regulation (1996) which were 150 and 100 mg/kg respectively. With the average concentration of 63.23 ± 0.72 mg/kg, prawn samples has the highest concentration of Cd compared to sardines, tuna, anchovies, shellfish, mackerel, cuttlefish and mixed seafood (Table 2). However, studies with experimental animals that was made by JECFA had shown that high

concentration of dietary intake of Zn can cause anemia as well as decreased concentrations of Cu and Fe absorption and reduction in the activities of several important enzymes in various tissues.

Copper (Cu)

The presence of Cu in canned seafood has been acknowledged as an important issue for human health. However, Cu has a low availability in many foods because of its tendency to bind with organic matter and Cu is rarely contaminates seafood in marine environment [12]. In the present study, the highest Cu concentration was observed in S13 (8.31 ± 0.71 mg/kg). All canned seafood analyzed presented Cu concentrations far below the guideline of JECFA (1982) and Malaysian Food Act and Regulations (1996) which both were 30 mg/kg. As illustrated in Table 2, type of food that shows the highest concentration of Cu is prawns. According to JECFA, copper is not carcinogenic in either humans or animals. Highly-exposed populations do not turn out to be adversely affected, nor does Cu appear to be a cumulative toxic hazard for man, except for the one with Wilson's disease.

TABLE 1. Total concentration of toxic and essential elements according to the type of seafood (mg/kg)

Sample	As	Pb	Cd	Zn	Cu
Sardines					
S1	0.88 ± 1.34	1.35 ± 1.14	0.08 ± 0.71	42.73 ± 0.37	2.10 ± 0.60
S2	0.62 ± 0.29	3.87 ± 0.71	0.08 ± 0.91	32.52 ± 0.72	1.48 ± 0.92
S3	0.37 ± 0.12	1.17 ± 0.83	0.07 ± 1.24	39.67 ± 0.22	1.40 ± 0.58
S4	0.84 ± 0.66	1.36 ± 0.87	0.02 ± 0.91	40.64 ± 1.19	1.24 ± 1.05
S23	0.12 ± 1.09	2.36 ± 0.79	0.05 ± 0.08	54.83 ± 0.97	2.41 ± 0.08
Tuna					
S5	ND	0.46 ± 0.93	0.09 ± 0.68	36.92 ± 1.33	0.75 ± 0.30
S6	0.44 ± 0.71	1.18 ± 0.79	0.03 ± 0.88	18.81 ± 0.04	0.81 ± 0.73
S7	0.45 ± 0.71	0.69 ± 0.71	0.08 ± 0.30	2.05 ± 1.26	0.68 ± 0.12
S8	1.13 ± 0.71	1.46 ± 0.71	0.06 ± 1.37	10.03 ± 0.67	0.58 ± 0.71
S19	ND	0.99 ± 0.24	0.04 ± 0.43	21.92 ± 0.58	3.35 ± 1.19
S24	0.09 ± 0.71	1.36 ± 0.76	0.14 ± 1.18	23.46 ± 0.54	1.09 ± 0.78
S25	2.00 ± 0.71	1.57 ± 1.16	0.01 ± 0.71	17.57 ± 0.71	1.53 ± 0.42
S26	ND	0.69 ± 0.90	0.26 ± 0.00	34.69 ± 0.10	0.69 ± 0.15
S28	0.17 ± 1.30	1.91 ± 0.67	0.01 ± 0.40	39.10 ± 0.94	0.76 ± 0.32
S30	0.65 ± 0.58	0.86 ± 0.71	0.03 ± 0.72	59.08 ± 0.71	2.36 ± 0.71
Anchovies					
S10	0.45 ± 0.71	2.48 ± 0.56	0.27 ± 1.18	40.92 ± 0.47	1.57 ± 0.04
S15	7.06 ± 1.13	3.21 ± 0.51	0.38 ± 1.31	71.40 ± 0.65	4.67 ± 0.69
Shellfish					
S11	1.20 ± 1.06	2.60 ± 0.80	0.31 ± 0.27	58.92 ± 0.29	3.19 ± 0.74
S12	0.63 ± 0.96	3.23 ± 0.38	0.49 ± 0.75	44.21 ± 0.29	3.97 ± 0.82

Prawns					
S13	1.19 ± 1.18	2.99 ± 0.97	0.18 ± 0.85	84.32 ± 0.31	8.31 ± 0.71
S20	0.44 ± 0.40	1.43 ± 0.39	0.33 ± 1.41	42.13 ± 1.13	7.40 ± 0.57
Mackerel					
S9	0.36 ± 0.71	0.90 ± 0.76	0.07 ± 1.41	20.46 ± 0.13	1.15 ± 0.38
S14	0.79 ± 1.33	1.15 ± 0.00	0.02 ± 0.00	50.34 ± 0.25	1.14 ± 0.41
S22	0.13 ± 0.71	0.38 ± 0.47	0.29 ± 0.15	2.28 ± 0.08	0.78 ± 1.18
Cuttlefish					
S16	0.08 ± 0.71	2.23 ± 0.92	0.96 ± 0.08	55.92 ± 0.20	5.09 ± 0.59
S17	0.80 ± 0.91	2.22 ± 0.82	1.66 ± 0.63	55.29 ± 0.48	4.45 ± 0.36
S18	0.05 ± 0.71	1.41 ± 1.09	0.68 ± 1.26	35.61 ± 0.34	1.94 ± 0.50
S21	1.15 ± 0.71	2.63 ± 0.71	0.64 ± 0.05	50.16 ± 0.52	3.57 ± 0.85
Mixed Seafood					
S27	4.73 ± 0.71	2.70 ± 0.71	0.03 ± 1.09	22.21 ± 1.30	1.18 ± 0.31
S29	0.04 ± 0.71	0.56 ± 0.16	0.27 ± 0.03	15.11 ± 0.08	2.32 ± 0.35

Comparison between the Type of Seafood

Concentrations of toxic elements in different canned seafood under study are represented by average values in Table 2.

TABLE 2. Average concentrations for each elements according to type of seafood (mg/kg)

Samples	As	Pb	Cd	Zn	Cu
Sardine	0.57 ± 0.7	2.02 ± 0.87	0.06 ± 0.77	42.08 ± 0.69	1.73 ± 0.65
Tuna	0.49 ± 0.54	11.17 ± 0.76	0.08 ± 0.67	28.35 ± 0.69	1.26 ± 0.59
Anchovies	3.76 ± 0.92	2.85 ± 0.54	0.33 ± 1.25	56.16 ± 0.56	3.12 ± 0.37
Shellfish	0.92 ± 1.01	2.02 ± 0.59	0.40 ± 0.51	51.57 ± 0.29	3.58 ± 0.78
Prawns	0.82 ± 0.79	2.21 ± 0.68	0.26 ± 1.13	63.23 ± 0.72	7.86 ± 0.64
Mackerel	0.43 ± 0.92	0.81 ± 0.41	0.13 ± 0.85	24.36 ± 0.15	3.07 ± 0.66
Cuttlefish	0.52 ± 0.76	0.53 ± 0.89	0.99 ± 0.51	49.25 ± 1.54	15.05 ± 0.58
Mixed Seafood	2.39 ± 0.71	1.63 ± 0.44	0.15 ± 0.56	18.66 ± 0.69	1.75 ± 0.33

The highest average concentration for As was detected in anchovies samples with the concentration 3.76 ± 0.92 mg/kg while the lowest average concentration was in mackerel samples which is 0.43 ± 0.92 mg/kg. For Pb, the highest average concentration for Pb element was in tuna samples (11.17 ± 0.76 mg/kg) and the lowest average concentration was in cuttlefish samples which was 0.53 ± 0.89 mg/kg. The highest and lowest average concentration for Cd element were in cuttlefish samples (0.99 ± 0.51 mg/kg) and sardine samples (0.06 ± 0.77 mg/kg) respectively. While for essential elements Zn and Cu, the highest average concentration were prawns samples (63.23 ± 0.72 mg/kg) for Zn element and cuttlefish samples (15.05 ± 0.58 mg/kg) for Cu element. The lowest average concentration for Zn element was in mixed seafood samples with the concentration 15.05 ± 0.58 mg/kg and for Cu element, tuna samples has the lowest average concentration which is 1.26 ± 0.59 mg/kg. The average toxic and essential elements (mg/kg) are in following order for sardine: Zn > Pb > Cu > As > Cd; tuna: Zn > Pb > Cu > As > Cd; anchovies: Zn > As > Cu

> Pb > Cd; shellfish: Zn > Cu > Pb > As > Cd; prawns: Zn > Cu > Pb > As > Cd; mackerel: Zn > Cu > Pb > As > Cd; cuttlefish: Zn > Cu > Cd > Pb > As; mixed seafood: Zn > As > Cu > Pb > Cd. For example, the results of the sardine samples indicated that the concentrations of heavy metals (mg/kg) were varied for As: from 0 to 7.06 with a mean of 0.96 mg/kg; Cd: from 0.58 to 8.31 with a mean of 0.25 mg/kg; Pb: from 0.46 to 3.87 with a mean of 1.71 mg/kg; Cu: from 0.58 to 0.87 with a mean of 2.40 mg/kg, Zn from 17.57 to 84.32 with a mean of 38.96 mg/kg.

CONCLUSIONS

In summary, concentration of the toxic elements (As, Cd and Pb) and essential elements (Cu and Zn) were successfully analyzed in 30 different samples of local canned seafood. The highest average concentration of trace metals in all samples was Zn which is 38.96 ± 0.56 mg/kg, followed by Cu (2.40 ± 0.57 mg/kg), Pb (1.71 ± 0.71 mg/kg), As (0.96 ± 0.70 mg/kg) and Cd (0.25 ± 0.70 mg/kg). According to the concentration of toxic elements in canned seafood purchased in Malaysia, canned seafood are not healthy to be consumed as the concentration was considered to be quite high. Anchovies was the species with the highest concentration of toxic elements which is 6.93 mg/kg while prawns was the type of seafood with the highest concentration of essential elements which was 71.08 mg/kg. Meanwhile, concentrations of these elements were broadly comparable to those found in the previous studies. In general, ingestion of toxic elements can cause harm to body even in the small concentrations while excess intake of essential elements can lead to toxicity.

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