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ANALYSIS OF HEAVY METAL ACCUMULATION IN FISHES FROM THE COAST OF LAUTOKA, FIJI

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Abstract. The concentrations of the accumulated heavy metals such as cadmium, lead, copper, mercury, zinc, and chromium have been determined in the muscles, gills and liver of the fish species of *Clupea pallasii*, *Macolor niger* and *Pristipomoides filamentosus* collected from the waters of the South Pacific Ocean around Lautoka in Fiji. Results of this study show the order of metal accumulation in tissues of all fishes as follows: Zn >Cu > Cr >Cd >Pb >Hg. Overall, the contents of heavy metals in all the samples were below the permissible limits, except for chromium that is slightly higher than the limits of 0.15 mg/kg and 0.05 mg/kg set by Food and Agriculture Organization of the United Nations and World Health Organization regulations respectively. As anticipated, the muscles contain the lowest concentration of all metals. Significant variations in heavy metal concentrations were found between different tissues within each species of fish.

Keywords: heavy metal, accumulation, fish, marine environment, Pacific Ocean.

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Introduction

In recent decades, industrial and urban activities have contributed to the increase of heavy metal contamination in the marine environment and have directly affected coastal ecosystems [1]. For this reason, the level of contaminated substances in fish tissue is often related to those found in their surroundings. Some of the heavy metals are essential to the physiological processes of fish, while others are toxic even at low concentrations. These heavy metals can enter the fish through feeding, the respiratory tract or the skin [2]. Sources of heavy metal accumulation come from food, water and sediments. In general, higher the concentration of metals in water, the more they are accumulated and stored by the fish [3,4]. The relationship between metal concentration in fish and water has been studied in field and laboratory experiments [5]. Various fish species can store different amounts of metals from the same aquatic environment. In terms of metal accumulation, these differences between fish species may be related to their lifestyle and diet [6]. Studies on many fish have shown that these metals alter the physical activity and biochemical parameters of tissues and blood. The toxic effects of heavy metals have been reviewed, among which many researchers include their bioaccumulation [7,8].

The presence of heavy metals in fish may pose a substantial risk to fish consumers such as human beings that include serious threats like renal failure, liver damage, cardiovascular diseases and even death [9,10]. Because of the dangers associated with the consumption of heavy metals, their concentration in commercial fishes in Fiji should be periodically examined to evaluate the possible risks linked to the consumption of contaminated fish. During the last twenty to thirty years, the concentrations of heavy metals in fish have been comprehensively investigated in various parts of the globe [11-13]. Most of the studies were concentrated mainly on the heavy metals in the fish muscles *i.e*. the edible part of the fish. Furthermore, few studies reported the distribution of metals in different organs such as bone, liver, gonads, kidneys, digestive tract and the brain [14,15].

Lautoka, (17.6242° S, 177.4528° E) is the second largest city of Fiji. It is in the west of the island of Viti Levu. The South Pacific Ocean encircles almost half of Lautoka and is the main source of fisheries. No analysis has been done to date on the presence of heavy metal ions in the fish species generally found at the coast of Lautoka. Therefore, the objective of the current study is to determine the heavy metals concentration (Cd, Pb, Cu, Hg, Zn, Cr) in the muscles, gills and liver of the three different fish species such as *Clupea pallasii, Macolor niger, Pristipomoides filamentosus* collected from the waters of the South Pacific Ocean around Lautoka in Fiji.

Experimental

Generalities

All solutions were prepared with analytical grade reagents and ultra-pure water (18 MΩ cm) generated by purified distilled water with the Milli-QTM PLUS system (Millipore, Bedford, MA, USA). High grade HNO_3 and $HClO_4$ were obtained from Merck, Germany. Standard solutions were prepared from stock standard solutions of the metals (Merck, multi-element standard, Germany) by diluting the stock solutions in mg/mL [16]. All glass and plastic items were hosed overnight with 10% (vol./vol.) nitric acid, washed with distilled water as well as deionized water and dried before use.

Fish sampling

The samples of the fish species *Clupea pallasii* (Pacific Herring), *Macolor niger* (Black Snapper), and *Pristipomoides filamentosus* (Crimson Jobfish) were purchased from the fishermen, at the key fish landing areas on the South Pacific Ocean around Lautoka in Fiji. A total of 15 fish samples, comprising 3 species and for each species five samples were collected on different dates between April to November 2018. The fishes were then classified with the help of an expert zoologist. The collected fish were instantly stored in an icebox kept in a polystyrene bag and shifted to the laboratory where they were weighed and the total length measured. They were kept in a deep freezer at around -25°C for further analysis. Lengths to nearest cm and weights to nearest g of the investigated fishes are presented in Table 1. The stated weight and length are given in the range from minimum to maximum of the three measurements taken for each species.

Preparation of samples

Before analysis, the frozen fish samples were moderately defrosted and then the fishes were dissected with a stainless steel knife to take out the muscles, liver and gills. Multiple samples of 2-5 g were used for ensuing analysis. The removed parts were kept in several Petri dishes and oven dried using a microwave digestion system (Multiwave 7000, Anton Paar GmbH, Germany) that ensures the novel pressurized digestion cavity (PDC) at temperature up to 300°C and pressure up to 199 bar, respectively. The internal temperature control was used to assist the acid digestion process to eliminate the content of water. This helps in achieving complete digestions of samples. The samples for heavy metal analysis were performed according to the method prescribed by the association of official analytical chemists [17].

Procedure: A quantity of 0.5 g of dried fish parts was placed in a 125 mL Erlenmeyer flask with glass beads and 25 mL deionized water. In this, 10 mL of 1:2 mixture of concentrated HNO_3 and $HClO_4$ were added. The sample was boiled until the solution became clear. The completely digested sample was allowed to cool to room temperature. The clear solution was transferred to a 50 mL volumetric flask to bring up to the volume with deionized water and mixed.

Determination of heavy metals

The samples were analysed for Cd, Pb, Cu, Zn, and Cr using the AA800 Perkin Elmer atomic absorption spectrophotometer (Norwalk, CT, USA) with an air/acetylene flame. The instrumental parameters were adjusted in order to achieve a maximal signal-to-noise ratio. The accuracy and precision of the analytical procedure were tested with a reference material (DORM-2, dogfish muscle, National Research Council, Canada). The mean recoveries of Cd, Cr, Cu, Zn, and Pb were 98.6, 104.3, 100.8, 89.2, and 96.5%, respectively. The instrument limit of detection (LoD) was calculated as the concentration was associated with thrice the standard deviation of the background noise recorded on seven measurements of the procedural blank [18]. The instrument limit of quantification (LoQ) was calculated as the concentration was associated with 10 times the standard deviation of the background noise [19]. The methods LoD and LoQ were calculated in a similar way by using real sample digest with concentrations of metals in the range of one to five times the instrument LoD.

Table 1

Length and weight of examined fish species.

Common name	*Scientific name*	*Length (cm)*	*Weight (g)*
Pacific Herring	*Clupea pallasii*	34—41	500—550
Black Snapper	*Macolor niger*	27—34	280—450
Crimson Jobfish	*Pristipomoides filamentosus*	26—42	400—800

The Hg was determined using the Milestone DMA-80 direct Mercury Analyzer (Milestone Srl, Italy). The sample weight required is between 0.020 and 0.006 g, with drying temperature of 300°C for 60 seconds. The samples were analysed at the Institute of Applied Sciences, University of South Pacific, Fiji.

The blank sample was analysed together with each batch of samples. All samples were analysed three times. The concentrations of heavy metals were given in mg/kg, wet weight (wet wt.) for the fish sample.

Results and discussion

The increase in human population and economic growth contributed significantly to the current global decline in water quality, including the periodic accumulation of heavy metals such as Cd, Pb, Cu, Hg, Zn, and Cr in the waters of the South Pacific, Lautoka Bay, Fiji. The non-essential metals have no metabolic function; but as a consequence of their bioaccumulation in fish, these metals may be toxic to human beings even at very small concentrations [20]. The exposure to heavy metals in water leads to the accumulation in the body of all aquatic organisms but fishes are generally more affected with respect to any other organisms [21,22]. The Food and Agriculture Organization (FAO) of the United Nations and World Health Organization (WHO) have established the allowed limits of heavy metals in fishes, these are presented in Table 2. The comparison among the three fish species in the Lautoka waters according to their metal accumulation levels in muscles, liver and gills are given in Tables 2, 3 and 4, respectively.

The accumulation of heavy metals in the tissues of the different species was found to be almost similar. Therefore, the accumulation concentration of zinc and copper were the highest in all tissues of the species, except for copper in gills of *Pristipomoides filamentosus* whereas the accumulation concentration of cadmium, lead and mercury were at the lowest levels. For example, the order of metal accumulation in muscles of *Clupea pallasii* was Zn >Cu >Cr >Cd >Pb >Hg. The similar order was also followed in the tissues of all other species.

However, the trends of the accumulation of heavy metals in all the three different species were similar but there were large differences in the amounts of heavy metal accumulation in the examined tissues of the species. This shows that the three different species of the fishes in the identical region accumulated diverse levels of heavy metals in their tissues. The results presented in Tables 2, 3 and 4 show that all the fish contain the lowest concentrations of metal in the muscles except for copper and zinc whereas almost all the species of fish have the highest concentrations of chromium in liver.

Table 2

Heavy metal concentration (mg/kg) in muscles of fish species.

	Cu	*Zn*	*Cr*	*Cd*	*Pb*	*Hg*
Clupea pallasii	0.771±0.013	12.402±0.01	0.291±0.001	0.233±0.003	0.112±0.001	0.021±0.001
Macolor niger	0.472±0.011	18.331±0.06	0.344±0.003	0.151±0.002	0.091±0.003	0.092±0.002
Pristipomoides filamentosus	0.521±0.002	15.614±0.02	0.312±0.002	0.081±0.001	0.021±0.001	0.073±0.001
WHO*	30	40	0.05	1	2	0.5
FAO**	30	30	0.15	0.5	0.5	0.5

*The maximum allowed heavy concentration established by *WHO and **FAO.*

Table 3

Heavy metal concentration (mg/kg) in gills of fish species.

	Cu	*Zn*	*Cr*	*Cd*	*Pb*	*Hg*
Clupea pallasii	0.491±0.015	7.403±0.01	0.391±0.008	0.192±0.001	0.364±0.003	0.164±0.001
Macolor niger	0.422±0.01	9.201±0.04	0.313±0.005	0.261±0.002	0.243±0.001	0.071±0.001
Pristipomoides filamentosus	0.372±0.013	9.902±0.02	0.421±0.006	0.331±0.001	0.292±0.002	0.181±0.001

Table 4

Heavy metal concentration (mg/kg) in liver of fish species.

	Cu	*Zn*	*Cr*	*Cd*	*Pb*	*Hg*
Clupea pallasii	0.683±0.007	13.702±0.02	0.441±0.004	0.332±0.001	0.181±0.001	0.061±0.001
Macolor niger	0.612±0.004	23.112±0.04	0.473±0.006	0.291±0.001	0.304±0.002	0.103±0.001
Pristipomoides filamentosus	0.451±0.003	17.803±0.02	0.401±0.004	0.223±0.002	0.251±0.002	0.142±0.001

The gills were found to have the maximum concentration of lead. The highest concentrations of mercury show a discrepancy between gill and liver in the three different species with the maximum value in the tissue of *Pristipomoides filamentosus*. Many studies have shown that active metabolic organs like gill and liver can accumulate a greater concentration of heavy metals as compared to the tissues of muscles [23]. The variations in the metal concentrations in tissues may be due to their ability to induce inter-metal binding proteins such as metallothioneins, as reported in many earlier studies [24,25]. The gill is an important site for the entry of the heavy metals and is the first target organ for exposure in fish [26]. The high concentration of metals in the gills is due to the metals complexation with the mucus. The concentration of metals in the gill reflects the level of the metals in the waters where the fish live, whereas the concentration in liver represents the storage of metals [27]. Thus, the gills in fish are more often recognized as the environmental indicator organs of water pollution than any other fish organs [28].

Copper being identified as a vital element, it is necessary for a wide variety of enzymes and other cellular components with important functions in all living beings. However, the excessive consumption of copper is harmful to human health as it leads to toxicity, and other related ailments. The current investigation shows the concentrations of copper in muscles was the highest (0.77 mg/kg) in *Clupea pallasii* whereas the species *Macolor niger* shows the lowest amount (0.47 mg/kg). The concentration of copper in gills was lower in comparison to the muscles and the minimum concentration (0.37 mg/kg) was present in *Pristipomoides filamentosus*. As far as liver is concerned, the copper accumulation was found similar to that of muscles and gills. The safe and adequate daily intake of copper is estimated at 1.5-3.0 mg for adults [8]. The mean concentration of copper in fish tissue samples ranged from 0.37-0.77 mg/kg, which is similar to 0.251-0.907 mg/kg according to the available literature [29], but for human consumption, it was not greater than the allowed level (Table 2) by FAO/WHO [30]. The high copper concentration can be attributed to oil dropping by frequent boating activities or may be due to too much fishing activities around the sampling site.

Zinc is harmful to human health if taken in excess and may cause intoxication [31]. Zinc levels found in the tissues of fish species are less than the FAO permitted limits (Table 2) of 30 mg/kg [32]. The species *Macolor niger* has the maximum amount of zinc in liver (23.1 mg/kg) as well as in muscles (18.3 mg/kg) in comparison with the other two species. In the gills, the highest concentration of zinc (9.9 mg/kg) was found in *Pristipomoides filamentosus*.

Chromium is among the common heavy metal pollutants that enters the water through the effluents from electroplating, dyeing, or printing industries. Chromium is an affective teratogenic and carcinogenic agent for human. The allowed limit established by WHO for chromium is 0.05 mg/kg [33]. The concentrations of chromium were comparatively lower for all the organs of the species. The lowest concentration (0.29 mg/kg) was in the muscles of *Clupea pallasii*, while the maximum concentration (0.47 mg/kg) was determined in the livers of *Macolor niger*. Therefore, the chromium concentration for all samples was higher than the limit (0.05 mg/kg) prescribed by WHO (Table 2).

Cadmium is not an essential element however it competes with calcium ion at the enzyme sites of the organism. Cadmium is considerably accumulated in the liver, followed by gills and muscles. According to FAO standard, the maximum admissible limit of the concentration of cadmium is 0.5 mg/kg [32]. Cadmium tends to have pronounced nephrotoxic potential and the excessive intake may lead to renal and hepatic toxic effects [34]. Since fish muscles are the main edible part, the concentration of cadmium was found to be lowest in the muscles except gills of *Clupea pallasii* of all the species investigated. Cadmium concentrations in the samples of all the species of analyzed fish were below the legal limits (Table 2).

The maximum concentration of lead detected in this investigation was in the gills of *Clupea pallasii* (0.36 mg/kg), whereas the lowest concentration (0.02 mg/kg) was determined in the muscles of *Pristipomoides filamentosus*. Henceforth, regardless of the investigated fish species, lead concentration in all the analyzed samples was lower than the level 0.5 mg/kg specified by FAO [35] given in Table 2.

The consumption of fish containing mercury above the permissible limit may badly affect the central nervous system and the endocrine system. The concentration of mercury in all the investigated samples was found to be lower than the permissible limits prescribed by FAO which is 0.5 mg/kg [36]. However, the accumulation of mercury even at very low concentrations is toxic.

Table 5

Comparative results for metal accumulation in fishes (mg/kg) from different locations.

Location	*Cu*	*Zn*	*Cr*	*Cd*	*Pb*	*Hg*	*Ref.*
Gulf of Oman, Oman	0.24-19.5	1.82-67.3	0.054-0.072	<0.005	0.011-0.005	<0.5	[38]
Hainan coastal area, South China Sea, China	0.93±0.006	9.95±0.99	1.82±0.31	0.002±0.001	0.07±0.02	-	[39]
Coastal waters of Terengganu, Malaysia	0.007±0.001	0.109±0.04	-	0.005±0.002	-	-	[40]
Bulgarian Black Sea Coast, Bulgaria	0.71±0.18	8.74±0.98	0.03±0.01	0.08±0.001	0.06±0.01	0.16±0.02	[41]
Rio de Janeiro State Coast, Brazil	2.18±0.19	4.71±0.60	-	0.02±0.008	0.2±0.1	-	[42]
The Coast of Lautoka, Fiji	0.532±0.006	12.631±0.03	0.382±0.004	0.231±0.001	0.204±0.001	0.091±0.001	Present study

The concentration of mercury is generally greater in the liver of fishes than in the muscles tissue [37]. However, the present investigation does not follow the trend and the maximum concentration of 0.18 mg/kg was determined in the gills of *Pristipomoides filamentosu*s and the minimum concentration was present in the muscles of *Clupea pallasii* (0.02 mg/kg).

The comparison of literature results for metal accumulation in fishes studied from different locations has been summarized in Table 5. The average concentration of copper in this study was approximately the same as compared to the reported values. However, the average accumulation of zinc was the highest in the current investigation compared to the other studies, except the results reported by de Mora, S. *et al*. [38]. In case of the average concentration of chromium, it was only less than that determined in fish samples from Hainan coastal area, South China Sea. The average cadmium concentrations level reported in the present study were higher than the previous studies in different locations. For the average level of lead, it was found to be higher than reported results except for the Rio de Janeiro State Coast, Brazil. The average mercury level in different parts of fish in this research was found to be below than the levels reported in the selected literature.

Conclusions

The heavy metals concentration (Cd, Pb, Cu, Hg, Zn, Cr) was determined in the muscles, gills and liver of three different fish species such as *Clupea pallasii*, *Macolor niger*, *Pristipomoides filamentosus* collected from the waters of the South Pacific Ocean around Lautoka in Fiji.

The obtained results indicate that the highest concentration was detected for zinc (23.112 mg/kg) in the liver of *Macolor niger* and the lowest concentration was found for mercury (0.021 mg/kg) in the muscles of Clupea *pallasii*. There was no single type of fish that was consistently high for all metals. The concentration range of the investigated metals including copper (0.372-0.771 mg/kg), zinc (7.403-23.112 mg/kg), cadmium (0.081-0.332 mg/kg), lead (0.021-0.364 mg/kg) and mercury (0.021-0.181 mg/kg), was within the permissible limits recommended by FAO/WHO, thus makes them available for human consumption. However, the concentration of chromium among all the three species was in the range of 0.291-0.473 mg/kg, higher than the WHO limit of 0.05 mg/kg.

The results of this study show considerable variations in the bioaccumulation in muscles, gills and livers for the specific heavy metals in the fishes from the waters of South Pacific Ocean around Lautoka in Fiji.

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